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In This Issue

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CAMEL POPULATION/ CONFERENCES/ AWARD/ RESEARCH TRENDS IN 2020

A synthetic analysis of the world camel demographic changes was done critically by Dr B.Faye in his recent Springer open access publication* which drew attention of many of us. Accordingly, it is particularly important to accurately estimate the place of camel production in the world economy. In many countries, camel data are insufficiently recorded. Between 1961 and 2018, the world camel population multiplied by 2.75 which was higher than many other species, possibly due to renewed interest in the camel within this new global climatic context. The present large camelid population in the world is probably more than 40 million and could reach 60 million after 25 years from now if the current demographic trend is maintained.

*(Faye, B. How many large camelids in the world? A synthetic analysis of the world camel demographic changes. *Pastoralism* 10, 25 (2020). https://doi.org/10.1186/s13570-020-00176-z)

Many kudos to Doug Baum for organising "Southwest Camel Conference 2020" at Texas, USA on 28 October 2020 with three sessions, free, via zoom. The speakers were Dr TK Gahlot, India, Abdul Raziq Kakar, Al-Ain, UAE, Ahmed Eisa El Hag, Sudan, Gil Reigler, Oasis Camel Dairy, USA, Surong Hasi, Inner Mongolia, China, Ivan French, Oklahoma Mini Mill. Although it was aimed for limited participants but was highly interactive and educative because of live practical sessions at farm sites.

The triannual JCPR brought newer trends of research this year. Noteworthy is a thorough study of camels in Somaliland, Eritrea and Ethiopia by R.Trevor Wilson. He has also given a critical review of the literature and bibliography of dromedary camels. Research on Bactrian camels is coming up substantially. Scientists from China studied CYP2E1 gene, proteome profile of hump, effect of camel milk on the liver injuries, transcriptome analysis of adipose tissue and whole genome sequencing. Scientists from UAE performed excellent work in the diagnosis of infectious and parasitic diseases. Identification of *Schistosoma indicum*, Actinomycosis or lumpy jaw, trichomonosis, MERS and *Trypanosoma evansi* abortion was done. Scientists from Saudi Arabia studied vitamin B1, B2, B6 and B12 levels in serum and CSF of camels with diverse neurological disorders. Studies on gliotoxin intoxication was done by the scientists of Saudi Arabia and Egypt. Pharmacological studies on dUMP, UTP, UMP metabolising enzymes in camels and *Trypanosoma evansi* was done by the Saudi scientists. A series of studies on biomarkers for bone metabolism, cardiac troponin I, infection and inflammation was done by Saudi Scientist Dr M. Tharwat. Scanning microscopy of *Wohlfahrtia magnifica* was done by Chinese scientists. SEM of oesophagus and anatomy of tongue was studied by the Indian Scientists. Detailed lung lesions were studied by the Sudanese scientists. Additionally, scientists carried out research on microsatellite markers, immune-histochemical studies of poll gland, lactobacillus in skimmed milk, ultrastructure of ulbourethral (Cowper's) gland, insulin contents of milk, ultrasonography of digestive tract, castration, endotoxins of camel milk, altitude effect on camel milk properties, expression level of the MERS-CoV receptor and dipeptidase 4, total mix ration with roughages feeding, *Hyalomma dromedarii,* sarcoptic mange, characteristics of Shami camels, dermatophytosis, *in vitro* capacitation of spermatozoa, endocardial fibroelastosis (Llama), *Klebsiella oxytoca* isolation from nostrils and lameness.

In this Covid 19 pandemic year 2020 when everything was stand still, laboratories and institutions were closed, we were quite apprehensive about receiving manuscripts for various issues of JCPR. Astonishingly, there was an overwhelming response from the camel scientists who contributed excellent manuscripts for JCPR. As a reciprocating gratitude gesture Camel Publishing House will issue an award certificate bearing a title, "ACTIVE CAMEL RESEARCH SCIENTIST AWARD 2020" to all the authors of volume 27 of JCPR. This award is dedicated to those camel research scientists who continued to work in the year 2020 during Covid 19 pandemic despite of several constraints and preferred to get their work published in the Journal of Camel Practice and Research. All authors will receive a PDF of this award by email in the first week of January 2021. Congratulations to all. Hope to receive your continued support in the year 2021 also.

Wishing a Happy New Year to all authors and editors!

(Dr. T.K. Gahlot)
Editor

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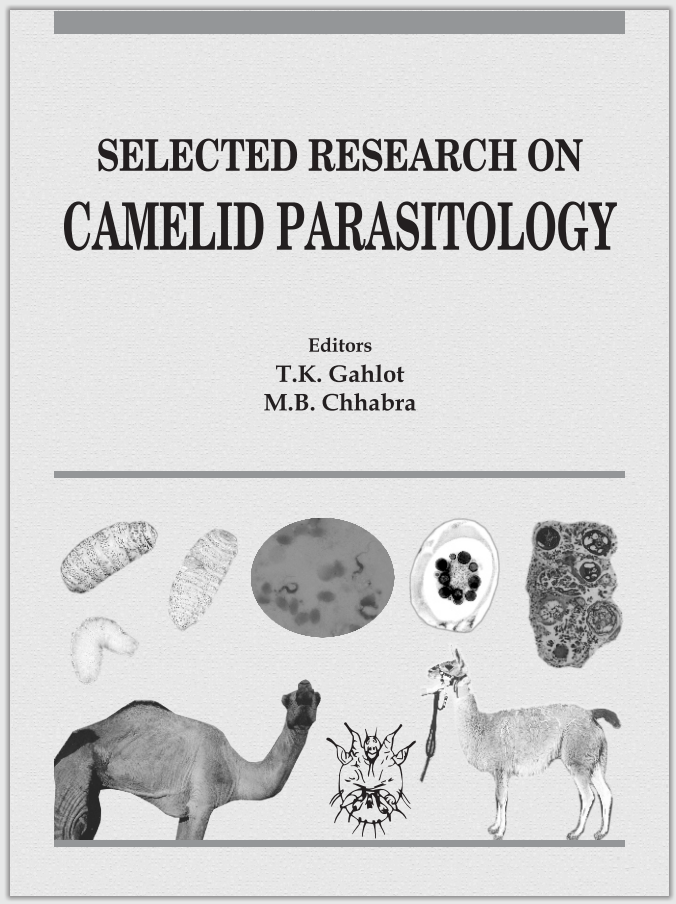
SELECTED RESEARCH ON
CAMELID PARASITOLOGY

Hard bound, 291 pages, few figures coloured

New research and experience always broaden our knowledge, and help us adopting new diagnostic methods and treatments. Camel Publishing House has taken a step forward to compile this knowledge in form of a book and this Herculian task was accomplished with the help of dedicated editors. viz. Drs. T.K. Gahlot and M.B. Chhabra. ***Selected Research on Camelid Parasitology*** is most comprehensive guide to Camelid Parasitology. The classic reference book serves as a one stop resource for scientific information on major aspects of Camelid Parasitology. Featuring abundant photographs, illustrations, and data, the text covers camelid protozoa, helminths, and arthropods of dromedary and New World camelids. This hard bound book of 304 pages contains seroepidemiological studies, immunological and other diagnostic procedures, and new treatments of parasitic diseases. There are at least 17 countries involved in camelid parasitology research, viz. Ethiopia, France, India, Iran, Jordan, Kenya, Libya, Mauritania, Nigeria, Sultanate of Oman, Pakistan, Saudi Arabia, Sudan, Sweden, United Arab Emirates, Uganda and U.S.A. As per published papers in Journal of Camel Practice and Research (JCPR), 173 authors have contributed 72 manuscripts which are appropriately placed in 5 sections. The text of each manuscript published previously in JCPR remains the same except the pattern of numbering the references in the body of text. This book indicates a swing of camelid research during period 1994-2008 and will help identifying the missing links of research in this subject.



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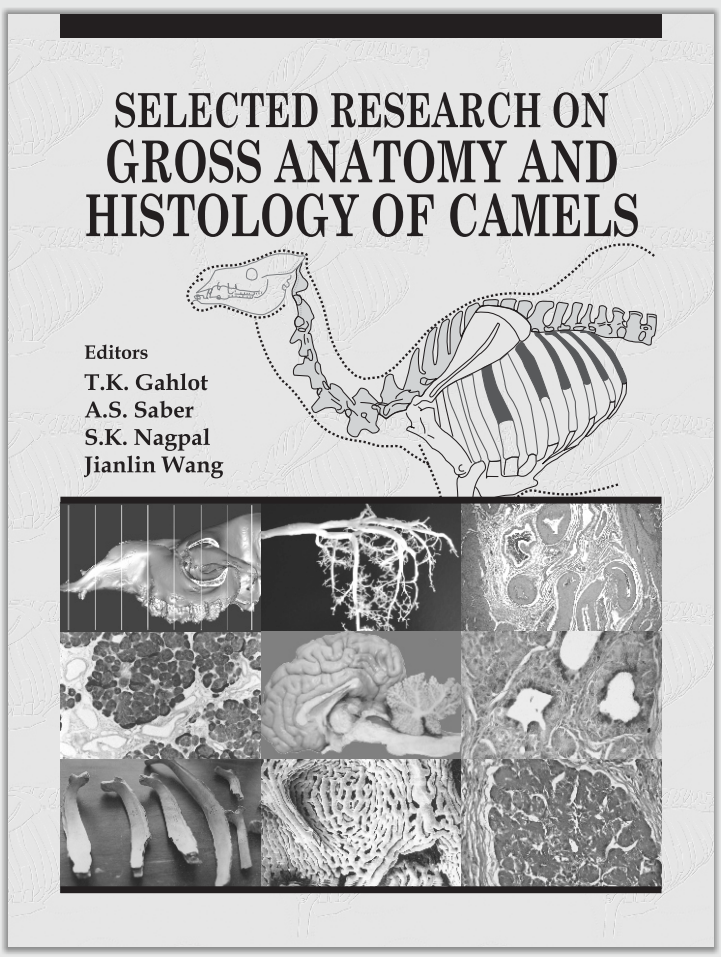
SELECTED RESEARCH ON
GROSS ANATOMY AND HISTOLOGY OF CAMELS

Hard bound, 452 pages, few figures coloured

Selected Research on Gross Anatomy and Histology of Camels is a unique reference book on anatomy of dromedary and bactrian camels. This book contains a first ever wide spectrum of histological description of various organs of camels which is depicted by special stains and scanning electronmicroscopy in addition to the gross anatomy, histochemical and immunohistochemical studies. The book has 92 manuscripts in 9 sections, e.g. radiographic anatomy, anatomy of various systems (skeletal, digestive, respiratory, circulatory, urogenital and nervous), common integument and miscellaneous. These manuscripts were published by 158 authors working in 37 laboratories or colleges or institutions from 14 countries in the Journal of Camel Practice and Research between June 1994 to June 2010. Bactrian camel anatomy research was exclusively contributed by the researchers of China. The countries involved in camel anatomy research were China, Egypt, India, Iran, Saudi Arabia, Iraq, Jordan, Japan, Pakistan, Sweden, United Arab Emirates, United States of America, France and Germany. Camel Publishing House has taken a step forward to compile this knowledge in form of a book and this herculian task was accomplished by its dedicated editors, viz. T.K. Gahlot (India), S.K. Nagpal (India), A.S. Saber (Egypt) and Jianlin Wang (China). This classic reference book will serve as a one stop resource for scientific information on gross anatomy and histology of camels.



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THE ONE-HUMPED CAMEL IN ERITREA AND ETHIOPIA: A CRITICAL REVIEW OF THE LITERATURE AND A BIBLIOGRAPHY

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ABSTRACT

Eritrea and Ethiopia, among the poorest in the world, are independent nations in north east Africa. Ethiopia's land area is ten times that of Eritrea and its population outnumbers that of Eritrea by a factor of eighteen. Ethiopia has the greatest number of livestock in Africa at an estimated 120 million ruminant animals of which 1.1 million are camels. In contrast Eritrea's livestock population is under 10 million in which camels may number 320 000. The one-humped camel possibly arrived in the area of study about 1900 years ago. The main areas of distribution are the arid lowlands below 1000 metres altitude although in recent years there has been some range expansion to higher elevations. In Eritrea camels are owned by Beja tribes near the border with Sudan, by Tigre clans in the north and by Afar and some Somali in the east along the Red Sea littoral. Ownership in Ethiopia is mainly by the Somali people and by the Afar in their respective Regional States in eastern Ethiopia and by the Boran in the south. Overall herd structure shows 40 per cent male and 60 per cent female. The genetic resource is generally referred to by the name of the ethnic group owning it but there are also classifications based on colour. The camel value chain includes milk, meat, hides, transport and medicines with milk for home consumption being the principal product. Welfare is poor by many standards. Camels suffer from many diseases including zoonoses. Trypanosomosis is a major problem as are respiratory diseases and bacterial infections. Ethnoveterinary knowledge is not well documented but is widely understood. Nutrition mainly derives from browse species but a wide range of feed resources is consumed. There is some supplementary feeding for commercial milk production. In the overall national and livestock economies the camel is of minor importance but is a major contributor to household wealth, welfare and food security to the many pastoral families inhabiting the driest and most impoverished areas of the two countries. The paper is complemented by an Annex (Bibliography) with more than 360 references.

Key words: Arid zones, camel trypanosomosis, *Camelus dromedarius*, disease, genetic resources

Background

The State of Eritrea and the Federal Democratic Republic of Ethiopia are independent nations in northeast Africa (Fig 1). "Eritrea" was formed in the late nineteenth century when Italy invaded the area and forcefully incorporated several independent and distinct kingdoms and sultanates into the Italian East Africa Colony or Italian Eritrea. Following defeat of the Italian colonial army in 1942 by Allied forces the country was administered by a British Military Administration until 1952. In that year the UN General Assembly decided that Eritrea would become an autonomous region of Ethiopia with a local Eritrean parliament: foreign affairs and defence would be federal in nature together with Ethiopia. In 1962, Ethiopia annulled the Eritrean parliament and annexed Eritrea. In 1991, after 30 years of continuous armed struggle for independence, the Eritrean Liberation Front achieved victory over the Ethiopian forces. The State of Eritrea came into being in 1993 after a referendum overwhelmingly voted for complete independence. Ethiopia, unique among African countries, was an independent country for many centuries except for a short period from 1939 to 1942 when it was invaded and colonised by Italy. The Ethiopian monarchy was overthrown in 1974 by a military coup and became a Marxist republic. The Marxists themselves were toppled in 1991 when the country became known as the Federal Democratic Republic of Ethiopia.

Both countries are among the poorest in the world and subject to frequent drought due to erratic and generally low rainfall. Famine is a consequence of these droughts coupled to poor agricultural practices. Eritrea is much smaller than Ethiopia with an area of about 117 600 square kilometres compared to the 1 104 300 square kilometres of Ethiopia. Eritrea is divided administratively into

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six Regions each with a number of Subregions. Its 2017 population was estimated at 5.9 million people. Gross Domestic Product (GDP) per caput in 2017 was estimated at the equivalent of USD 1600 at Purchasing Power Parity rate (PPP) (CIA, 2018a). Ethiopia's area is almost tenfold that of Eritrea. The country operates as a federal state with nine ethnically based Regional States (Addis Ababa and Dire Dawa being Chartered Cities) each comprising Zones (total 68), Districts ('woreda') and Neighbourhoods ('kebele'). Ethiopia's population of over 105.3 million inhabitants outnumbers that of Eritrea by a factor of eighteen. GDP per caput was USD 2200 at PPP in 2017 (CIA, 2018b).

Agriculture remains a major component of the economy in both countries and employs the greater proportion of the population. Crop production is important in the moister highland area above 1300 metres altitude whereas livestock dominate the economies of the arid lowlands. Cattle, sheep, goats and equines are all significant in the highlands with cattle and equines providing much of the energy needed to drive crop production. In the lowlands camels are a major species along with goats, sheep and cattle.

This paper expands on and updates an earlier one (57 references) written by the author more than 30 years ago that covered the then Ethiopia (including Eritrea) (Wilson, 1989). Many of the early papers related to Eritrea and were written by Italian veterinarians during their occupation of the country. A goodly proportion of the remainder were about Ethiopian camels and produced by French veterinarians of the Institut d'Elevage et de Médecine Vétérinaire des Pays Tropicaux. Only five papers, all university dissertations, were written by Ethiopian nationals. In 2012 a book entitled "Camel in Ethiopia" was published because "camel production and health has, for the past last three decades, featured in the curricula of Ethiopian Veterinary and Agriculture colleges [but] there has been no textbook on Ethiopian camels [and] this book is intended for undergraduate veterinary and animal science students, policy makers and researchers". Further, the authors "tried to distill the scattered and scanty literature on Ethiopian camel, the pastoralist, the environment, the market and camel health and welfare [and] relied heavily on [their] experience of the past 25 years of on and off teaching and research on camels, blended with results and experience from other countries" (Melaku and Getachew, 2012). This last book lists 154 references of which only 55 (35.7 per cent) are directly related to Ethiopia and of which 52 of these last (94.5 per cent) have at least one Ethiopian among the listed authors.

Since the early 1990s more than 230 articles or theses on camels have been produced. Only six (2.5 per cent) of these do not have an Eritrean/Ethiopian among the listed authors. Only five (2.1 per cent) of all these articles relate to Eritrea and two (40.0 per cent) of these were contributed by non-Eritrean authors. This paper provides a detailed analysis of camels and camel production in the two countries. It does not attempt to cite all the more than 360 references listed in the Bibliography but presents a representative selection of papers related to the various section headings.

Methodology

The methodology for this paper comprises two distinct parts. The first is based on the author's own knowledge and observations garnered over 45 years of (residential and intermittent consultancy) work in both Eritrea and Ethiopia starting in 1974. This part also benefits from the vast repository of knowledge of camel keepers throughout Eritrea and Ethiopia who shared their experiences with the author. Discussions with veterinarians, administrators and development workers also contributed to this paper.

The second part was a very detailed review of the literature which is provided as an Annex ("Bibliography") to the main text. Search terms for the literature review were: "camel", or "one-humped camel", or "dromedary", or "Eritrea", or "Ethiopia". It needs to be noted here that the search terms did not include "North East Africa", "Horn of Africa" nor "ruminants". It is known that many papers including these terms do contain information on the camel in the two countries under review but it is unlikely that they would add much, if any, knowledge additional to that already included in the analysis.

It also needs to be noted that Eritreans and Ethiopians have a system of naming that differs form the conventional "family" name system used in most western countries. In the area under study a child is given a personal name at birth, to which is added the father's given name and sometimes the paternal grandfather's given name. All people are addressed throughout life, even in official situations, by their given name as 'Ato' (Mr), 'Wezeiro' (Mrs) or 'Wezeirat' (Miss) and females do not change their patronym on marriage. This system is particularly problematic for citations as some journals will name the author by his Ethiopian form of address, others will treat the patronym as a "surname" and list the

author by this followed by his or her "initials", yet others will provide only initials for the first (given) name and add the patronym after it. It is thus no easy task to use a name search to identify items published by one person and, indeed, in some data bases and reference lists the same paper is cited twice (or more times) because of this confusion. There are additional problems with variable spellings of both given names and patronyms due in part to the way the geez alphabet has been transliterated In this paper, the author has attempted to standardise the presentation in the text citations and in the reference list using the Eritrean/Ethiopian system of given name plus patronym even when the original publication or references to it used a "western" system (thus, Mohammed YK, Kurtu MY and YK Mohammed have all been rendered as Mohammed Yusuf Kurtu). It is certain that the reference list, as presented, has not been completely successful in achieving this but citations by one person as the first author now appear together in the alphabetical reference list and most duplications have been removed. Searching on a given name or a patronym will in most cases lead to the author and the reference being found.

History and introductions

Palaeontological research in Ethiopia in the lower Omo valley discovered a molar tooth and a metatarsal bone dated at 2.6 million years ago (Arambourg, 1947). These seem to be of a Bactrian camel and are the first camel remains recognised from eastern Africa (Howell *et al,* 1969). The one humped camel appears to have been present in Ethiopia at least as early as 100 AD. This is evident from rock paintings in a cave at Laga Oda (Fig 2), some 30 km south-west of Dire Dawa in southeastern Ethiopia at about 9° 15′ N and 41° 18′ (Cervicek, 1971). A camel tooth found in Axum is dated at about 500 AD (Phillipson, 1995).

It is possible that modern camels arrived in Eritrea/Ethiopia from both the north and the east. The former route from Mesopotamia, across the Sinai and down the western side of the Red Sea and the latter from the same origin through the Arabian Peninsula and across the Red Sea north of its entry into the Gulf of Aden (Melaku and Getachew, 2012). There is, however, no DNA analysis to corroborate this hypothesis.

Numbers

Foreview and international sources

Some early attempts at enumerating livestock were made by the Italian administrations in Eritrea and during its short hegemony in Ethiopia (Marchi, 1929; Pirani, 1938; Roetti, 1938; Girardon, 1939; Salerno, 1939; Bonomo, 1940). These were, however, very partial and probably far from accurate.

Since then estimates of numbers over time have varied widely among years, sources of data or information and method of computation. A livestock census was taken in 1978, followed by sample surveys in most years since. Neither the census nor the subsequent surveys, carried out by the Central Statistical Authority (CSA) have covered the entire country. Eritrea and Tigray were not included in much of the 1980s, mainly because of the disturbances of the peace in those areas. The "pastoral" areas of the Afar and Somali Regions – whose human populations of the eponymous ethnic groups have been traditionally suspicious of and hostile to a central government and who have strong antipathy to having their wealth counted and where most camels are to be found — were also not included in the enumerations until the early years of the twenty-first century. The Food and Agriculture Organisation of the United Nations (FAO) produces statistics on an annual basis with data provided by official sources of the country or arrives at numbers based on an estimate from many years ago that are updated by a formula peculiar to that organisation.

In general terms, based on FAO data, Eritrea and Ethiopia are home to about 6.4 per cent of the world's one-humped camel population. Using three other criteria to determine the importance of camels in the livestock economy of a country, the contribution to total domestic herbivore biomass of Ethiopian camels is about 3.7 per cent (26^{th} in a league table of 36 countries with one-humped camel populations), the number of camels per person is 0.03 (12^{th} of 36) and the number of camels per square kilometre of land area is 0.87 (5^{th} of 36). By any of these criteria, therefore, camels are a valuable resource in the region. Their true value to particular sections of the population is nonetheless masked by these very crude estimates.

FAO sources provide time series data for camel numbers from 1961 to 1993 for the former Ethiopia. Since 1993 data have been provided separately for Eritrea and Ethiopia. From 1961 to 1976 FAO reported numbers from official data but from 1977 to 1992 numbers were FAO estimates. From 1993 to 2016 all numbers for Eritrea were FAO estimates whereas for Ethiopia numbers were FAO estimates from 1993 to 2004 since when it is claimed they were from official data (FAO, 2016). According to FAO camel

numbers in Ethiopia (prior to 1993 thus including Eritrea) rose from 930 000 animals in 1961 to 1 070 000 in 1992 with a marked fall in numbers between 1974 and 1975 followed again by an increase (Fig 3). In the 1994 edition of the FAO Production Yearbook the number of camels for the former Ethiopia for 1992 was 1.07 million (FAO, 1995). In 1993 and 1994 the data for Ethiopia showed 1.0 million and 0.069 million for Eritrea (FAO, 1995): i.e the previous Ethiopia camel population had been partitioned between the two new states. In the most recent FAO publication, however, camel numbers for Eritrea are shown as 312 000 in 1993, rising to 373 572 in 2016 (FAO, 2016). The FAO 2016 publication shows the "new" Ethiopia of 1993 having 320 000 (meaning that camel numbers for Eritrea plus Ethiopia had fallen from 1.07 million in 1992 to 0.69 million in 1993), a total that increased to 445 000 in 2004 (FAO, 2016). CSA data for the year 2000 indicate 262 000 camels for Ethiopia (CSA, 2000) whereas the FAO Yearbook for 1999 still uses the old estimate of 1.03 million (FAO, 1999). From 2005 to 2016 FAO claims it uses Ethiopian official data but in that first year it indicates 458 576 camels (Fig 3) whereas the CSA is already reporting 2 100 000 (CSA, 2006a).

Eritrea

The Eritrean Ministry of Information admits to a lack of reliable statistics on livestock populations as there has been no census since 1978 and numbers are based on estimates. In 2007 one secondary source estimated 9.4 million ruminants of which about 320 000 were camels (Bissrat and Woldeselassie, 2007). In 2012 the camel population was estimated at 318 914 (Fig 3). This may be a completely spurious value as is the official estimate of cattle numbers where annual vaccinations are often double the official estimate (MOI, 2012). An estimate of 75 000 camels appears in a slightly earlier paper but does not indicate its source (Dioli, 2006). An otherwise quite detailed paper by an Eritrean author working in the Ministry of Agriculture does not give any indication of numbers (Gebrehiwet, 1998). Another estimate indicates camel numbers at 373 952 (Banerjee, 2006).

Ethiopia

The regular Agricultural Sample Surveys carried out by the CSA (e.g. CSA, 2000; 2006b; 2008; 2010; 2017) have suffered from only partial coverage, being restricted to the settled agricultural areas. In the year 2000 the "agricultural" areas of the country had 242 410 camels, in 2006 the population was 437 606 camels, in 2008 the number was 1 009 040, in 2010 there was a fall in numbers to 807 581 but by 2017 the number had again risen to 1 418 457.

National estimates of livestock numbers for 2005 indicated that the country was home to 43.8 million cattle, 23.2 million sheep, 22.8 million goats, 1.5 million horses, almost 4.1 million donkeys, 356 thousand mules, 2.1 million camels and probably in excess of 50.0 million poultry (CSA, 2006a). These data include results of surveys in the two "pastoral" areas of Afar Regional State and Somali Regional State (CSA, 2004a; 2004b) which had not been included in earlier population data. The surveys, carried out on the ground in Afar and from the air and on the ground in Somali arrived at a total of 759 750 camels in the former State and 1 041 870 camels in the aerial survey plus 64 510 additional animals from the ground survey in the latter State. Consequent on the censuses in the Afar pastoral areas and the aerial surveys of the Somali ones the estimated camel population of Ethiopia is now considered to be well in excess of two million head.

Hindview

It needs to be noted that inclusion of the pastoral areas into the general census puts a considerable new light on Ethiopia's livestock populations with moderate increases in cattle numbers, large increases in sheep and goat populations and very large increases in the camel population. It is clear from the available data that no firm conclusions can be provided for actual numbers in Eritrea and Ethiopia. Over time there has been considerable "redaction" of previously published data especially by FAO. In addition the actual area covered, the methodology employed and the analyses of any results are often confusing. Camels are the least numerous of all the domestic herbivorous mammals in both Eritrea and Ethiopia except for mules and possibly horses. On the assumption, however, that the Ethiopian camel population is in excess of 2.0 million claims that it has the third largest population of this domestic animal on the African continent, after Somalia and Sudan, may be valid but it is possible that numbers in Kenya exceed those in the two countries under review. The enumeration of camels in Eritrea and Ethiopia is best considered as a "work in progress".

Distribution

Eritrea

Eritrean camels occupy the lowland northern arc of the country (Dioli, 2006). They thus are found

from the Southern Red Sea via the Northern Red Sea through Anseba and into the Gash Barca Regions (Fig 1).

Ethiopia

According to official data 42 per cent of the Ethiopian national camel herd is found in the Somali Region of eastern Ethiopia, 34 per cent in the Afar Region of northeastern Ethiopia and 24 per cent in the Oromia Region in the Borana and Kereyeu Zones in southern and southeastern Ethiopia (CSA, 2006) (Fig 1). Almost all Ethiopian camels are thus found in the northern, eastern and south-eastern lowlands at altitudes below the 1000 m contour except in the south where Boran camels are commonly found at 1500-1600 m altitude (Fig 4). In recent years under the pressure of drought and probable overstocking of the lowlands, Afar camels have been brought to feed in the dry season west of Dukam at 2000 m and only 30 km from Addis Ababa. In other highland areas some camels are used by sedentary farmers and traders for miscellaneous transport operations. Transport camels along with mules and donkeys also regularly traverse the 3200 m ridge of the west wall of the Rift Valley near Wukro in Tigray carrying salt from Dallol to the market in Mekelle (Fig 5) (Wilson, 1976). There are no camels in the southwestern or western areas and a very few in the northwest of the country.

The camel is traditionally an animal of the wide open spaces, constantly moving with its owners from place to place in search of feed and water and avoiding urban areas. Some Ethiopian pastoralists, due to demographic, socio-economic and political factors, are beginning to settle and in the process triggering an unprecedented growth of small towns and the creation of urban centres across the pastoral lands. Pastoralists have thus had to adapt to new situations or be left without sustainable incomes. An initiative of "town pastoralists" is camel dairy production in and around these new and expanding urban centres (Abdi Abdullahi Hussien *et al,* 2011)

Ownership

Eritrea

The main ethnic groups owning camels are the Beja tribes – Beni Hamer and Rashaida – in the north west and part of the north, the Tigre clans in the west, the north and the northeast, the Afar in the east and southeast and the Somali in the southeast (Dioli, 2006). Livestock production is the main economic activity for all these groups with camels generally being the most important species (Assefaw *et al,* 1999). Most camels are owned by Muslim lowlanders in Eritrea but the Kunama people in the southwest close to the border with Ethiopia are mainly Christian. Camels were introduced into the highlands during the war of independence for carrying trade goods and for transporting military materials. This has led to some camels being owned by Christian highlanders who keep them for transport but, except for the Saho tribe, do not drink their milk or eat their meat (Gebrehiwet, 1998).

Eritrean camels are always considered as clan property although individuals and families "own" their own animals. Camels are branded with a clan mark and a subsidiary symbol that represents the individual or family. It is the clan that decides on the distribution of animals and this unit also arranges their distribution to deprived families or individuals, thus ensuring that members who have lost their animals can recover from the disaster. An individual possessor has no absolute right to give or refuse to give his or her animals (Gebrehiwet, 1998).

A male child is given a young or neonate female animal on its birth. Gifts of animals are also made to the child by close relatives. As he grows thus his herd increases. On marriage he is given a further allocation from the family holding and a bride price of two to seven camels is paid to the father of his new wife (Gebrehiwet, 1998).

Ethiopia

Ethiopian camels are mainly owned by the Somali people of the eponymous Regional State, then by the Afar in Afar Regional State and parts of Tigray Regional State. These are followed by the Borana people of the south of Oromia State. In the far northwest of the country some camels are owned by a small population of Kunana people, who are of Nilotic descent and whose main numbers are in southwest Eritrea. These last are mainly Ethiopian Orthodox Christians whereas in all other areas camel owners nominally follow the Islamic faith.

In much of Ethiopia camels are clan property with families and individuals benefiting from them in trust in a manner analogous to the system described for Eritrea.

Herd sizes vary over a wide range from as little as one animal to as many as 150 head. Many of these larger "herds" are, however, most likely to be agglomerations of camels belonging to several families. In one study of 73 families in eastern Ethiopia the average herding unit comprised 25 camels but there was a very large standard deviation

that was greater than the mean (Eyassu Seifu, 2009). All 73 (100 per cent) of these families owned camels whereas 67.1 per cent owned cattle, 37.0 per cent owned goats and 13.7 per cent owned sheep[1].

Herd structure

Camels are used less in Ethiopia for transport and draught purposes than they are in some other countries. Their main purpose for the Somali and Afar as well as the Boran is for the production of milk. The national herd structure reflects these functions with 39.4 per cent males and 60.6 per cent females. In Somali Regional State the ratio of males to females was 36:64 in the herd as a whole but in older animals over 4 years is about 1:2 indicating some offtake for meat or sale to other uses for transport and perhaps draught. The Afar herd once again shows the dairy vocation of all species of livestock with only 22.7 per cent males and 77.3 per cent females. About one third of camels under four years old in Afar are males and in the age group of over four years the ratio of males to females falls to about one to four. In one study in Somali Regional State 19.0 per cent of the camel herd was lactating females, 20.2 was male and 19.0 per cent was calves (Eyassu Seifu, 2009).

In the agricultural areas three quarters (76.6 per cent) of camels are in the age group of 4 years and older. About two fifths (39.4 per cent) of camels are in the age group of 4 years and older in the pastoral areas. Camels are multipurpose animals as indicated by the census return that shows 48 per cent kept for milk production and about 37 per cent for transport and draught. What the census does not show — only 3.3 per cent of animals are recorded as being kept for meat — but that can be inferred from the 40/60 per cent male/ female population structure is that camels are also meat animals either being slaughtered locally or sold to the international market. Almost all camels are found in the lowland areas (Fig 4).

Genetic resources

Early attempts to classify Ethiopian camels no longer seem entirely satisfactory and are certainly incomplete. Some merely named camels according to the tribes owning them (Droandi, 1921; 1932) and others are based on colour (Marchi, 1929). In the first of these sources subtypes were ascribed to racing or trotting ('il cammelli corridori') and to pack types. Taxonomy on this basis was confined to Eritrea in line with the needs of the Italian military colonial power. Among the qualities of trotting camels were regularity of pace, endurance for a whole day and a steady speed of 8-10 km/hr. Tribal types are almost all referable to Sudanese ones. The Bisari ('Bisciari') were preferred for riding and both females and males were used. Another Sudanese breed, the Anafi, was not subject to as intense selection in northern Ethiopia as in Sudan and it became more a general purpose type than a pure fast riding camel. Additional tribal types described were Cabbaci, Beni Amer (said to be very strong with reasonable speed) and Sceraf. Colour descriptions overlap tribal types. The Anafi thus become the Tzedi (white) and the Beni Amer the Cajeh (red). One type identified along the Red Sea coast was called the Grain (sandy).

Ethnic groups owning camels are rarely confined by the national boundaries of Eritrea or Ethiopia. The nomadic way of life and family ties have led to considerable continuity of camel types across the various national frontiers (Wilson,1984). The camels of the northern lowlands of Gonder and Eritrea thus have much in common with those over the border in Sudan; Afar camels are found also in Djibouti and northern Somalia; Somali camels cross the whole length of the frontier with Somalia and with Kenya in the southeast and Borana camels extend into northern Kenya in the south-central area.

The most common camel types in Eritrea are the Bisharri, Arrir and Afar. There are also unidentified camels in the Gash-Barka and Anseba regions. The Arrir is the preferred type in the southwestern lowlands due to its high milk yield, good market price and high transport value (Gebrehiwet, 1998).

In Ethiopia several camel populations have been identified although there is inconsistency in terminology even within the same group of authors (Table 1, Table 2).

Products

The camel value chain includes milk, meat, hides, transport and medicines. The traditional pastoralist mode of production is not, however, one of commodities and is not primarily aimed at producing for the market. There is, nonetheless, limited – but probably rapidly increasing — commercial trade in milk and meat as well as in live animals. The standard outputs of milk and meat are mainly for home consumption. Herd accumulation is a vital economic function not only for cash but for traditional values in the context of the extended family (being able to loan out animals), as bride price and for prestige within the community. Camel owners therefore tend to be asset rich whilst remaining cash poor.

1. It was a study of camel-owning families!

Table 1. A succinct classification of Ethiopian camel types

Camel type	Location	Colour	Height (cm)		Function
			Male	Female	
Afar	Northern lowlands	Fawn, red	160	150	Milk, (transport)
Ogaden (Somali)	Southeastern lowlands	Fawn	210	190	Milk
Borana	Souther lowlands	Fawn	185	170	Milk, pack, draught
Anafi	Northwestern lowlands	Very pale	190	170	Fast riding

Source: Yosef Tadesse *et al,* 2014.

Table 2. Morphological features of some Ethiopian camel groups.

Group	Morphological features
Hoor	Wide belly, long legs, long body, narrow hip width
Gelleb and Liben	Prominent hump, broad chest and hip, long neck and tail
Jijiga	Short body, medium body size and barrel girth
Shinille	Long ears, light weight, small heart girth, short stature
Amibara and Mille (Afar)	Small size small heart girth, light weight, long tail

Source: Yosef Tadesse *et al,* 2015.

Camels are raised mainly for milk by the Afar in Eritrea and the Afar and Somali in eastern Ethiopia, for milk, transport and riding by the Beja in the western lowlands of Eritrea and for milk and transport by the Borana in the south of Ethiopia.

Milk and dairy products

Milking is done by hand direct into containers (most often plastic) by both men and women who, because of the size of the camel, are able to stand during the process (Fig 6). Camel calves are given access to their dams to start the let-down process. Some milk is sold outside the immediate and extended family. Camel milk, which is rich in Vitamin C, partially offsets the deficit in cow and small ruminant milk supplies in many areas of lowland Ethiopia and makes a major contribution to the protein and calorie intakes of nomadic populations. In Eritrea in the early 2000s total camel milk production was estimated at 5385 tonnes, equivalent to 1.5 per cent of all milk and providing an availability of 0.85 litres per person per year (Banerjee, 2006). Total milk production from the Ethiopian national camel herd was estimated at 23 500 tonnes in 2005 (CSA, 2006b) although other sources indicate much higher amounts of up to 75 000 tonnes (Felleke, 2003). Formal export of camel milk ranges from 1600 to 2500 litres per day at a price of USD 0.08 per litre although a large amount is informally exported for Somali consumption through the Jijiga/Togochalle border land route (Abebe Bereda *et al,* 2016).

In the Shinile and Jijiga Zones of Somali Regional State the daily milk yield of a camel varied from 1 to 10 litres with an average of 5.2±.2.2 litres: lactation length varied from 180 to 720 days and averaged 382.7±96.0 days (Eyassu Seifu, 2009).

In Gode township in Somali Region it has been found that average milk production in the traditional system is 2.43 litres per day in the dry season and 3.38 litres per day in the rains. Under a periurban/urban system dry season yield was raised to 3.9 litres of which 0.98 litres was consumed at home and the remainder sold in the market. In the wet season the yield was 6.25 litres of which 1.27 litres was used by the household and 4.52 litres was available for sale (Hussien *et al,* 2011).

Fresh and fermented camel milk products are often credited with therapeutic properties. These include prophlaxy or cures for gastritis, asthma, stomach discomforts, HIV, hamot (kar), tuberculosis, fever, urinary problems, hepatitis, jaundice, common cold, dearbeh ("diarrhoea"), daarta ("nausea") and diabetes (Asresie and Kurtu, 2014).

Fermented camel milk, known as 'dhanaan' is said to have a shelf life of about five months (Asresie and Kurtu, 2014). Butter and cheese are potentially important value-added products of camel milk but in Ethiopia, as elsewhere, it has been found difficult to process and when successful the yield is lower than cow milk due to the lower butterfat content and the distribution of milk proteins (Asresie *et al,* 2013; Eyassu Seifu, 2007; Adugna *et al,* 2013; Tesfamariam *et al,* 2013; 2017; s2018). In one trial a now naturalized but invasive weed (*Parthenium hysterrophorus,* congress grass) was used to help turn

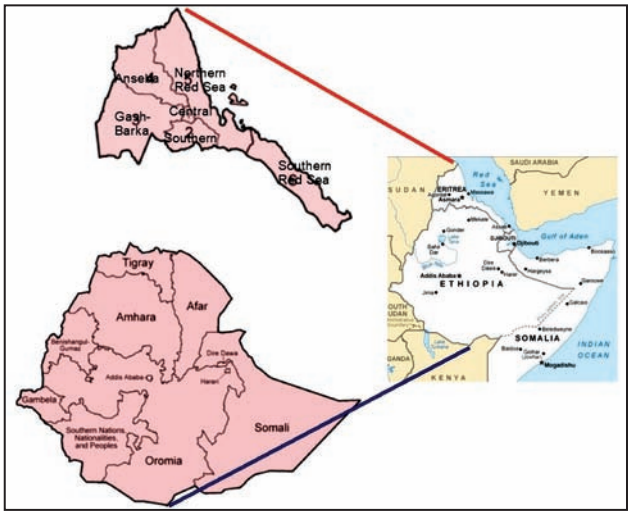

Fig 1. Location of Eritrea and Ethiopia in the Horn of Africa and country maps showing major administrative divisions (Source: compiled by the author from maps in the public domain).

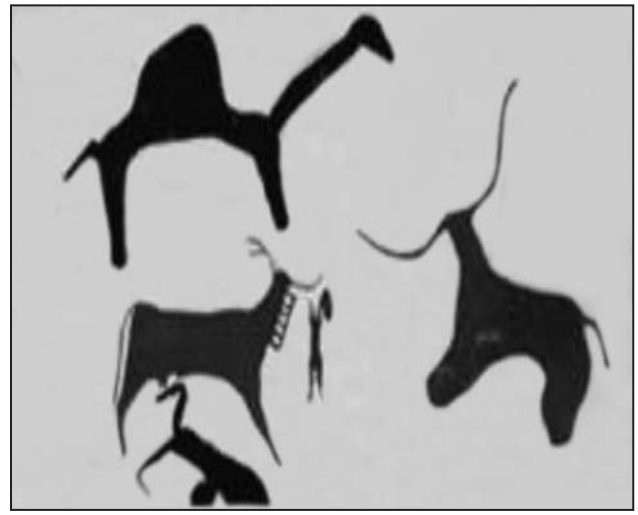

Fig 2. Rock paintings of camels in Laga Oda cave, southeast Ethiopia (Source: Cervicek, 1971).

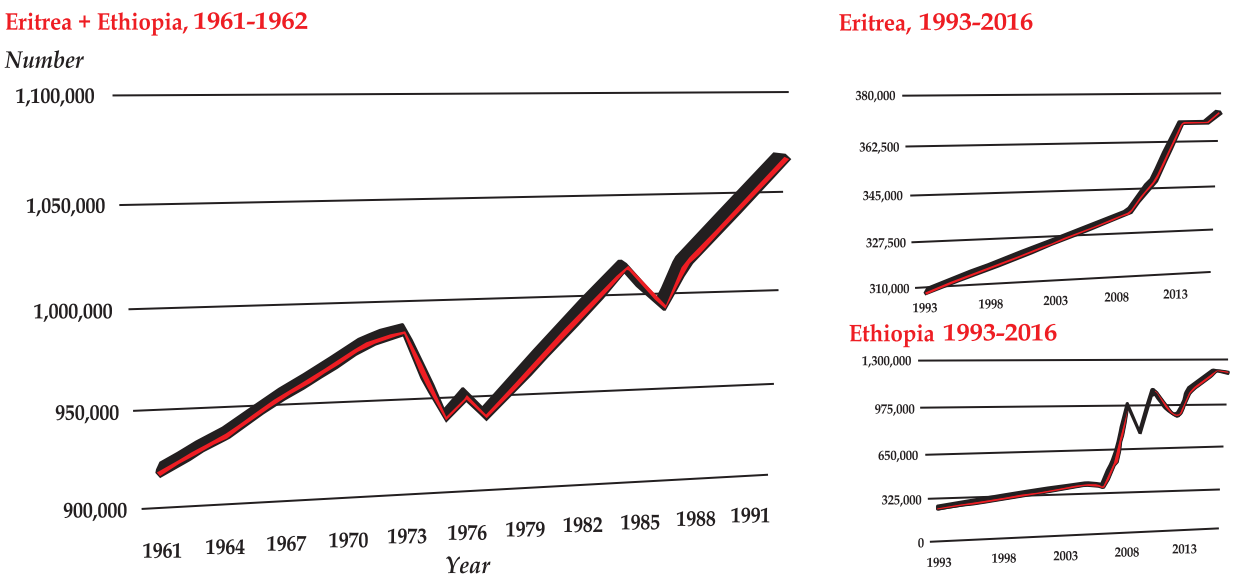

Fig 3. Camel numbers in Eritrea + Ethiopia 1961-1992, Eritrea 1993-2016 and Ethiopia 1993-2016: note different scales (Source: constructed from data in FAO, 2016).

the cream and produced a butter yield of 70 g from 3 litres of fresh milk.

Meat

In Eritrea in the early 2000s it was estimated that 48 614 slaughtered camels yielded 9722.8 tonnes, of meat and offal (i.e. a carcass weight of 200 kg) and that consumption of camel meat was 1.54 kg per person per year (Banerjee, 2006). In Ethiopia, camels will continue to serve a mainly niche as well as an emergency market for meat primarily in the lowlands but camel meat is not a preferred commodity among the Afar and Somali (Ayele Gebremariam, 1999). Total camel meat production in Ethiopia in 2005 was estimated at 4560 tonnes (CSA, 2006b). In 2010 camel meat was equivalent to 9 per cent of all meat produced whereas sheep and goats contributed 70 per cent and cattle 21 per cent (Abebe Bereda *et al,* 2016). In 2017-2018 some 6742 camels were slaughtered of which 4749 were males (CSA, 2018). Most camels are in poor condition (low body condition scores) at slaughter. Slaughter practices are often less than humane and even violate 'halal' (permitted) conditions including cutting the Achilles tendon of the hind legs, severing the neck with more than one stroke and sharpening knives and killing animals in

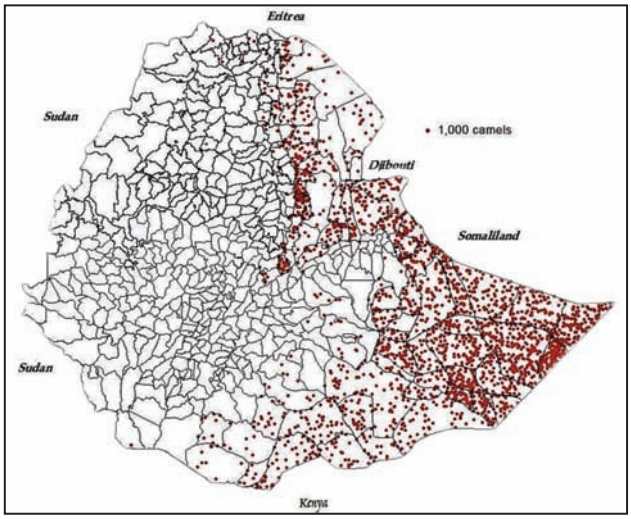

Fig 4. Gross distribution of camels in Ethiopia, 2004 (Source: generated from data in CSA 2006).

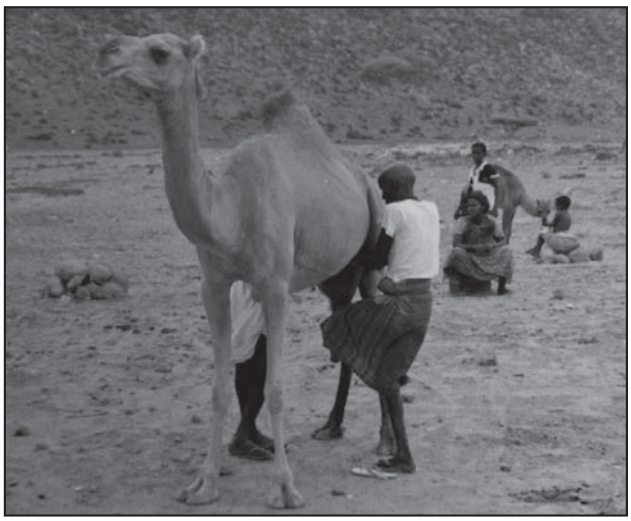

Fig 6. An Afar man milking a camel near Dire Dawa, 6 April 1986 (Source: photo by the Author).

Fig 8. Headquarters of the British Expeditionary force near Senafe (now in Eritrea) showing horses, mules and camels (Source: Engraving from the Illustrated London News, 8 February 1868).

Fig 5. Camels and mules loaded with salt from the Danakil at Wukro (3200 m altitude), 25 June 1974 (Source: photo by the Author).

Fig 7. Preservation of camel meat ('muremure') for longer storage life (Source: photos from Mitiku Eshetu Guya and Getachew Neme Tolesa, 2015).

Fig 9. Camels competing with rail transport in Italian Eritrea in the 1890s (Source: Gagliardi, 2016).

Fig 10. The Eritrean Camel Corps on parade in Asmara, 1920 (Source: Italian National Archives).

Fig 11. The Emblem of Eritrea honouring the camel for its transport role in the country's war of independence (Source: Public Domain).

Fig 12. A family of the Bilen clan of central Eritrea on a seasonal move of their dry season camp, 25 November 1993 (Source: photo by the Author).

Fig 13. Camels ploughing near Yabelo in Borana Zone, southern Ethiopia at 1800 metres altitude, 28 February 1987 (Source: photo by the Author).

Fig 14. Private small camel transport train carrying sacks of locally produced grain at Agula Pass (Tigray) at an altitude of 2000 metres, 6 November 2018 (Source: photo by the Author).

Fig 15. Pack camel in extremely poor condition and with severe (healed) saddle sores near Addy Abby, Tigray, 6 November 2018 (Source: photo by the Author).

Fig 16. Camel feed resources: desert conditions in Somali Regional state; lush vegetation on banks of River Awash in Oromia Regional State (Photos by the Author).

the presence of other camels awaiting slaughter (Seid *et al,* 2017).

In Jijiga and Harar towns in Somali Regional State in 1999 almost all slaughtered camels were adults and were predominantly male. Average live and carcass weights were 400 kg and 211 kg with males being significantly heavier and having higher dressing percentages than females. Carcasses comprised 76 per cent meat, 12 per cent fat and 20 per cent bone in both sexes (Mohammed Yusuf Kurtu, 2004). Issa camels at Dire Dawa slaughterhouse averaged 233.4 kg carcass weight and had a dressing

percentage of 52.7 per cent: dressing percentage was higher in camels of heavier live weight and there was more weight in the forequarters than the hindquarters (Abebe Wossene *et al,* 2002). Similar results were obtained in another study of Issa (Somali) camels (Seid *et al,* 2016).

In Somali Regional State camel meat is preserved by boiling to reduce the water content and to reduce water activity of the meat. Butter is added during boiling to enhance the flavour and eating quality of the product. The meat is then hung to dry and finally pelleted (Fig 7), The final product is known as 'mukmud' or 'muremure' and is said to have up to six months shelf life (Mitiku Eshetu Guya and Getachew Neme Tolesa, 2015).

It is claimed that camel meat is healthier than beef as it is high in protein (19 per cent), low in fat (1.17 per cent), contains most essentials amino acids, has low cholesterol (59.2 mg/100g) and low saturated fatty acids and is a rich source of vitamins and unsaturated fatty acids. Low levels of saturated fat in camel meat are important for avoiding atherosclerosis, for the control of obesity and hyper cholesterolaemia and decrease the risk of cancer because of their effect on plasma cholesterol levels. Camel meat, as for camel milk and perhaps equally as specious, is believed by Somalis to have remedial effects for as many as 13 diseases, including hyper acidity, hypertension, pneumonia and respiratory diseases and also to be an aphrodisiac. Camel meat in general is considered a functional food as a remedy for ailments that include seasonal fever, sciatica, shoulder pain, asthma, removing freckles and for improved performance. It is used as a cure for exhaustion and fatigue because it contains energy derived from sugar and not fat and also glycogen, a carbohydrate which is easily absorbed and metabolised in the body and converted to glucose which activates nerve as well as other cells (Hussein, 2018).

Transport and Draught

An early record of camels being used for transport relates to the British Expedition to "Abyssinia" in 1867-1868. During this exercise, undertaken to force the release of British hostages being held by Emperor Theodros II, up to 40 000 animals (elephants, mules, horses, donkeys, oxen and camels) were used to support the invading army (Fig 8). At various times throughout the expedition up to 10 000 camels were at work in the baggage trains (Holland and Hozier, 1870) although it is not known whence they came. Camels were not used only as baggagers, however, as they served to evacuate wounded personnel, either one each side on stretchers or in 2-person saddles on their backs for the less seriously wounded and generally sick or exhausted combatants.

Eritrea

The camel continued to be an important means of transport in Italian Eritrea at the end of the nineteenth century even after the advent of the railway with which in some respects it was in competition (Fig 9). The Italian administration of Eritrea as a colony wanted to be considered as a serious contender in the game of empire and as one expression of this — and like other colonial powers (Britain in Sudan and Somaliland, France in West Africa and Germany in Southwest Africa) — set up its very own Camel Corps (Fig 10). Camels were much used in the Eritrean war of liberation in the 1970s to early 1990s for transporting military equipment and weapons, a role for which they have been honoured by appearing on the Eritrean National Emblem (Fig 11). The Bilen people who are mostly located around Keren in central Eritrea use camels fitted with 'howdah' – possibly a relic of their historical links with the related Beja tribes and their origins in Sudan – when moving their temporary or seasonal camps for transporting family members and household goods (Fig 12).

Ethiopia

Before the establishment of Eritrea as an independent state estimates of camels used for draught were in the region of 130 000, of which two thirds were females (MOA, 1984): these were mostly found in Gonder in the northwest and Bale and Sidamo (Fig 13) in the south but the figures excluded any data for Eritrea and Tigray. Official estimates for Ethiopia for 2017-2018 were that 286 040 camels (about 20 per cent of the total camel population) were used for transport purposes of which 244 412 were males and 41 629 were females: draught camels numbered 26 584 all of which were males (CSA, 2018). About 330 000 households (1.85 per cent) of all households in Ethiopia) owned transport and draught camels. In the past camels were not normally hired out to other parties by the owners for transport or agricultural purposes. Years of drought have resulted in the deaths of many oxen, donkeys and mules and these are being replaced in the mid altitude areas by camels being kept by nonpastoral tribes who are more willing to rent out animals for both pack (Fig 14) and

draught to obtain income (Yacob Aklilu and Catley, 2011).

Hides and skins

Hides and skins and their value-added products are important items of internal commerce and international trade in Ethiopia. None of the numerous projects, reports and learned papers indicate, however, that camel skins are important in this respect. When flaying the hide is taken off by first making an incision along the back line (rather than the more common cut along the underline for other domestic animals) and then taken off in small pieces rather than as a whole ((Hussein, 2018).

Foreign exchange earnings

Livestock and their products are among the highest earners of foreign exchange for Ethiopia. Camels – whether legally or illegally exported – make a considerable contribution to the generation of foreign currency in support of Ethiopia's economy. In 2007-2010 some 62 per cent of an estimated illegal export of 6.8 million animals valued at USD 1.04 billion were camels (Abebe Bereda *et al,* 2016). Legal camel exports are mainly by sea through Djibouti and the Republic of Somaliland or by trekking overland.

In 2006 one company alone exported 20 000 camels to Egypt at a value of USD 6 million (Nazret.com, 2008). Some 79 000 camels were exported in 2010/2011 of which 15 000, valued at USD 6 124 800, crossed the border into Sudan through Humera on foot (Yakub Aklilu and Catley, 2011) with many having trekked 1500 km from Borana in southern Ethiopia: the other 64 000 were exported to or through Djibouti. During 2010/2011 Ethiopia exported 472 041 live animals of which 61 365 were camels. Camels thus contributed 13 per cent to the number of animals exported but their contribution to export revenue was 25 per cent (Sanitary & Phytosanitary Standards and Livestock & Meat Marketing Program, 2011). In the nine months prior to June 2017 a total of 11 527 camels valued at USD 6.57 million were exported (Ethiopian Herald, 2017).

Welfare and health

The Five Freedoms

Very few, if any, of the pastoralists or other groups who own and manage camels will have heard of the Five Freedoms (Brambell, 1965). If they are aware of them they are rarely put into practice.

Freedom from hunger occurs sporadically but for much of the year in many years the feed supply is less than the demand in terms of quantity and often inadequate in quality. Thirst is less of a problem for camels than for other domestic stock but even these water-efficient beasts have to contend with restricted water supplies at times.

Discomfort is ever present often in terms of searing heat, lack of shade, rough underfoot conditions and heavy and unstable loads.

Pain is often inflicted by owners in ignorance of what does not cause it rather than a desire to do harm. Animals are frequently injured through overloading and inappropriate harnessing (Fig 15). The nose peg is the preferred method of control rather than a halter: its insertion causes injury and it is painful in use. Disease might be considered the norm in camels including pathogens, parasites and skin ailments.

Camels are seen to express fear and be distressed when ill-treated by their handlers.

Perhaps the least noxious of the freedoms is the ability to express normal behaviour. Most camels are kept in herds and are able to associate with and interact with their fellow beings. Removal of the calf in order to provide milk for the family is, however, an intrusion on normal behaviour.

Diseases

Ethiopian camels suffer from a plethora of diseases caused by pathogens over a wide range of classes including viruses, bacteria, fungi and protozoa. In addition they are beset by internal and external parasites and an embarrassing array of mechanical injuries. Several diseases are zoonoses: of the five (rabies, anthrax, brucellosis, leptospirosis, and echinococcosis) given priority for a One Health approach in Ethiopia (Pieracci *et al,* 2016) brucellosis – mostly due *Brucella melitensis* – with 29 references in the bibliography and echinococcosis or hydatid disease with seven are common in camels. It is certain that the other three are present but have gone unreported. Among other major diseases reported as of concern by herd owners are trypanosomosis (20 references) and a complex of respiratory diseases (six references). Mastitis is also of concern (12 references in both traditional and "modernizing" herds. Skin problems include sarcoptic mange (Bekele Megersa *et al,* 2012; Nesibu *et al,* 2014), contagious ecthyma and tick infestations (Zeleke Mekuriaw and Tafesse Bekele, 2004).

Most of the early work on camel diseases was done by the Italian administration in Eritrea. A main interest was in the identification and control

of trypanosomosis (Pricolo and Ferraro, 1914; 1918; 1920; di Domizio, 1918; Frullini, 1938; Grassi, 1947; which is usually caused in camels by *Trypanosoma evansi* which is transmitted to them mechanically by biting flies of the Tabanidae family. There was also considerable interest in filarial worms (Pricolo, 1913a; 1913b; 1913c). At a later period the larval stages of *Taenia* species were causing concern (Angelotti, 1947; Pellegrini, 1947a; 1947 b; 1947c; 1947d; Batelli, 1949).

During the 1970s and 1980s the French Government financed a team of veterinarians from the Institut d'Elevage et de Médecine Vétérinaire des Pay Tropicaux (IEMVT) to assist the Ethiopians in disease identification and control. This team was also interested in trypanosomosis and carried out several trials to control it (Balis, 1977; Balis and Richard, 1977a; 1977b), helminths (Daynes and Richard, 1974) and bacterial diseases (Domenech, 1977; 1980; Domenech *et al,* 1977). The assistance was not wholly altruistic, however, as IEMVT personnel also benefited in doing research for doctoral degrees (Didier, 1975; Richard, 1975; 1979).

From the 1990s onwards most research on camel diseases has been undertaken by Ethiopian nationals. The French team can take some credit for this as they provided support to the Veterinary Faculty of Addis Ababa University — although veterinary education has also been supported by FAO and the British Government as early as 1969 (RTW, personal knowledge). In 2020 at least six Ethiopian universities provide courses in veterinary science and have awarded at least 44 postgraduate degrees (Addis Ababa Faculty at Debre Zeit, 33; Alemaya, 8; Haramaya and Hawassa, 1 each) on a variety of camel disease topics (see Annexe: Bibliography for full details).

Ethnoveterinary medicine

Only one direct study of the use of ethnoveterinary medicine in the treatment of camel ailments has been made (Tafesse Mesfin, 2000). In the Republic of Somaliland, bordering on Ethiopia's Somali and Afar Regional States and whose pastoralists are intimately related to their neighbours in Ethiopia ethnoveterinary practices have been documented since at least 1895 (Swayne, 1895). Camels showing stiffness were "fired", either by raising small blisters with a red-hot ramrod or spear or by striping with hoops of red-hot iron. Open sores had glowing stones strapped over them which was followed by an application of moist camel dung. When off feed a dose of melted sheep's tail was given. Thorns were removed from the foot with the 'biláwa' or dagger and camel dung was then applied. Sore backs caused by the chafing of a load was often bitten by the camel until it festered and became invaded by maggots, the treatment for which was a strip of calico, steeped in carbolic solution, tied over the wound to protect it from attack by omnivorous birds (Swayne, 1895). Some 30 years later a treatise on the camel provided additional information on ethnoveterinary medicine (Leese, 1927). Later work (Hunt,1951; Mares, 1951; 1954a; 1954b; Peck, 1939; 1940) included descriptions of plant remedies, traditional vaccination, cautery, use of broths and use of salt in the form of salt bushes, salty wells and salt-rich soils. Mares (1954a; 1954b) also provided an extensive list of Somali names for livestock diseases and parasites. In the 1990s participatory techniques were used to elicit information on indigenous practices (Catley, 1996; Catley and Ahmed Aden, 1996). More recent accounts of Somali ethnoveterinary practice show considerable agreement with the earlier work and even 40 years after the publication of Mares' work, herders in northern Somalia were still using soups, cautery and medicinal plants (Catley and Mohammed, 1995; 1996). A brief review of the literature indicates common terminology for some livestock diseases throughout Somali-occupied areas. For example, the words 'gendhi', 'dhukaan', 'caal', 'cadho' and 'cambaar' are very widely used by Somali herders from north-west Somalia through Ethiopia to northern Kenya.

There are other studies on ethnoveterinary medicine. Although not specifically targeted at the camel this animal has been part of the overall study. In one such study in a district with a camel population of only 244 head some 20 per cent of 51 plants identified as of ethnoveterinary value were used on camels. The most common ailments treated were diarrhoea, mange, ringworm, black quarter and bloat. *Allium sativum* was used against eight ailment types and *Croton macrostachyus* against seven (Ermias Lulekal *et al,* 2014). In Afar Regional State 12 species of plants were used in the treatment of camel ailments (Tafesse Mefin, 2000; Mirutse Giday and Tilahun Teklehaymanot, 2013).

Feeds and feeding

Feed is obtained from a wide range of habitats ranging from hyperarid deserts to succulent bushland (Fig 16) and such other resources as fallows and stubbles on agricultural land. Camels are predominantly browsers and because of their size are able to procure feed from heights of up to four

metres above the ground on resources that are not available to other domestic stock. They are eclectic in their tastes and feed on a broad spectrum of fodder plants that includes thorny trees and shrubs, halophytes and aromatic species that may be avoided by other domestic herbivores. At times, nonetheless, they compete with these latter for other types of feed including grasses and herbaceous legumes. Camels employ various feeding strategies depending on the season and the available resources, using the herbaceous layer of mainly annual species in the shorter rainy season and the browse layer of perennial plants in the longer dry season.

In Jijiga District camels mainly fed on browse species with all parts of the plants (leaves, twigs, seeds and pods) except the roots being eaten. More than 100 species of plants were identified of which 20 were commonly eaten. These species included *Acacia brevespica, Acacia bussei, Acacia etbaica, Acacia nilotica, Acacia senegal, Acacia seyal Acacia tortilis, Dichrostachys cinerea, Opuntia ficus-indica, Lantana camara, Blepharis persica, Grewia villosa, Ziziphus mauritiana, Euphorbia tirucalli, Heliotropium cinerascens, Commicarpus africanus, Rhus natalensis, Balanites glabra, Grewia ferruginea* and *Cadaba heterotricha*. Mean concentrations of Ca, Mg, K, Fe, Mn, Zn and Cu in forages were higher than the lower recommended levels in both the wet and dry seasons. The mean concentration of Na and P, however, were lower than the recommended levels for ruminants meaning that in the study district camels should be provided supplementary sodium and phosphorus from other sources (Tezera Getahun, 1998; Desalegn and Kurtu, 2012). This is well understood by pastoralists hence the frequent visits to salt pans or saline water sources.

In the Southern rangelands in Borana country the most important browse plants have been reported as *Acacia brevispica, Commiphora africana, Rhus natalensis, Grewia* spp., *Balanites* spp., *Boscia minimifolia, Cadaba glandulosa, Euphorbia* spp. and *Solanum tembensis* (Dessalegn, 1984).

Woody plants comprised 79 per cent of the diet in the dry season and 83 per cent in the wet season in the Errer Valley in eastern Ethiopia. The ten most preferred species were browsed for 87 per cent of the feeding time in the dry season whereas 80 per cent were browsed in the wet season. The highest ranked plant in the dry season was *Opuntia ficus-indica* in contrast to *Acacia brevispica* in the wet season. The range in composition of the ten most preferred species 88-228 g/kg dry matter for crude protein 1.3-3.3 for phosphorus, Ca 12- 48 for calcium, 29-216 for soluble tannins 9.4-129 for condensed tannins. *In vitro* dry matter digestibility (IVDMD) varied between 0.41 and 0.65 (Moges Dereje and Udén, 2005)

Attempts to improve nutritional status have not emphasized the natural feeding environment but have concentrated on niche feeding, mainly for dairy production. Milk yields were significantly increased, for example, from 7.6 kg per day to 12.9 kg when camels on natural grazing were provided with ground maize and a protein supplement: the percentage of butterfat also increased slightly (Moges Dereje and Udén, 2003). In another series of experiments lactating camels were studied in a cross-over trial in which they were watered once daily, every fourth day, every eighth day or every 16 days with a 5-day interval between treatments. When offered water every fourth or eighth day the camels drank enough to cover their needs for subsequent days but after 16

Table 3. SWOT analysis matrix.

Strengths	**Weaknesses**
Complement other domestic livestock species Multipurpose milk, meat, transport and draught animal Water conservation abilities resulting in long interval need for water Flexible management by owners (seasonal and long term) in search for and use of feed and water Adaptation to climate change	Extremely susceptible to respiratory infections and trypanosomosis Broad spectrum of other diseases constrain output Long reproductive intervals
Opportunities	**Threats**
Organisation into producer groups Better market information Improved market access Integration with agro-pastoral communities to provide draught power and transport Improve general management Improve disease control Improve nutrition	Inability to comply with World Trade Organisation Agreement on the Application of Sanitary and Phytosanitary Measures (SPS Agreement) Politically motivated bans on imports from Ethiopia National administration does not empathise with pastoralists Inadequate market infrastructure internally and for export Migration of owners (NOT camels) to urban environment Increasing numbers risk causing greater environmental damage

days of dehydration they did not drink enough to compensate the body weight loss. Rectal temperature fell at night and the camels searched shade during daytime minimising evaporative fluid losses. Contrary to general belief camels did not dilute their milk in response to water restriction ((Tafesse Bekele *et al,* 2011; 2013).

Discussion

Constraints

Ethiopia's pastoral communities have battled for centuries with adverse weather conditions, to say nothing of an often hostile political environment. In these circumstances they have been more successful in adapting to change than sedentary populations as they can be much more flexible in their daily, seasonal and annual cycles. There has, however, been widespread environmental damage from overgrazing in recent times. Some of this damage results from increases in both human and animal populations that themselves derive from better medical care and reduced mortality.

Over the years the main constraints to increased and more efficient production have been cited as disease, feed shortage, predators, water shortage, labour shortage and inadequate marketing channels and opportunities (Ayele, 2002).

Opportunities

Camels are better adapted to survival in areas with harsh climatic conditions than "conventional" domestic livestock species. As such the species supports the livelihoods and improves the resilience of the pastoral communities of the Ethiopian lowlands and are an extremely important source of food and of improved welfare for local pastoralists.

The camel is a major animal species in the lowland pastoral system but is assuming some importance in the mid-altitude mixed farming and agro-pastoral systems. This is due to its multipurpose role and the variety of products, both direct such as milk, meat and transport and indirect in its social and cultural importance. The camel is able to adapt to many aspects of climate change and continues to do just more than survive when confronted by shortages of feed and water. Demand for the camel and its products is increasing and will continue to do so at an accelerated rate that is in excess of the conventional domestic animal species (Seyoum Bediye *et al,* 2018). In view of the close relationship between feed and water, the latter should be used to direct access to the former with a reduction in environmental degradation being a principal aim of this (Wilson, 2007).

Lucrative export opportunities exist for both live animals and meat for transfers to, for example, Egypt, Libya, Saudi Arabia and the Gulf States (Tadele Mirkena *et al,* 2018). To capitalise on these, however, the value chain needs to become much better organised and potential problems with health and disease will need to be overcome.

The SWOT analysis (Table 3) provides a summary of the current situation with regard to strengths, weaknesses, opportunities and threats.

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ANNEX

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COMPARATIVE TRANSCRIPTOME ANALYSIS OF ADIPOSE TISSUES FROM BACTRIAN CAMEL

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ABSTRACT

The purpose of this study was to reveal the molecular mechanism that regulates fat metabolism in hump of Bactrian camel. The subcutaneous fat (SF), greater omentum (GOM) and hump fat (HF) of Bactrian camel were sequenced by RNA sequencing, and the differentially expressed genes of fat tissue in 3 different parts at transcriptome level were obtained by paired comparison. Genes related to fat metabolism were screened out, among which HF vs. GOM group had the largest number of different genes related to fat metabolism. Some genes such as SCD, FASN, ACACA, ADIPOQ, PLIN1, PLIN4, LPL and HSL were highly expressed in HF. These genes may play an important role in maintaining the energy homeostasis of Bactrian camels in the absence of food. Signaling pathway analysis revealed a number of pathways involved in fat metabolism including AMPK signaling pathway, PPAR signaling pathway, Adipocytokine signaling pathway, ECM-receptor interaction and regulation of lipolysis in adipocytes. These data suggests HF has a greater capacity to modulate the release and storage of triglycerides in adipocytes than GOM.

Key words: Adipose, Bactrion camel, RNA sequence, transcriptome

Fat is not only an important energy storage organ of the body, but also an important endocrine organ, which can secrete adipokines to regulate the energy balance of the body (Unamuno *et al,* 2018). According to the location of fat deposition, adipose tissue can be categorised into visceral adipose tissue (VAT) and subcutaneous adipose tissue (SAT). VAT has more cellular activity than SAT, has more abundant distribution of blood vessels and nerves, and contains more immune and inflammatory cells. VAT has higher metabolic activity and more lipolytic activity than SAT. VAT has a greater capacity to generate free fatty acids (FFA) and to absorb glucose than SAT, while SAT absorbs more circulating FFA and triglycerides (TG) (Ibrahim, 2010, Schoettl *et al,* 2018).

The reason why the camel can adapt to the extremely harsh living environment well is that its body structure and organ function have good adaptability to the desert ecological environment. Camels manage their stores of fat by having enough food. When food is abundant, the body needs to preserve fat and store energy, and when food is insufficient, the body needs to ensure that the camel can resist hunger, adapt to the barren vegetation in desert areas (Wu *et al,* 2014). Camels store fat mainly in hump, kidney, subcutaneous, abdomen, omentum and mesentery. The main component of hump is fat, and early studies have shown that hump and other fat storage sites are rich in fatty acids, phospholipids and TG (Kadim *et al,* 2002).

RNA-Seq approach has been applied to identify gene expression profiles of different types of adipose tissue in pigs, sheep and cattle (Cai *et al,* 2018; Kang *et al,* 2017; Xing *et al,* 2019) and many genes associated with fat metabolism have been identified. However, there has been no transcriptome study of adipose tissues in camel. We therefore, studied the differential gene expression in subcutaneous fat (SF), hump fat (HF) and greater omentum (GOM) by RNA-Seq technology, thus revealing mechanisms in adipose deposition and understanding of differences between adipose depots in Bactrian camel.

Materials and Methods

Ethic statement and tissue collection

All the procedures involving animals were approved by the Institutional Animal Care and Use Committee of the Inner Mongolia Agricultural University (IMAU) (License No. SYXK, Inner Mongolia, 2014–0008) with adherence to IMAU

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guidelines. The study was conducted on castrated Bactrian camel (aged approximately 10 years) obtained from Bayan hot in Inner Mongolia. A total of 3 healthy individuals were selected randomly. The 3 types of adipose tissues i.e., subcutaneous, visceral and hump were taken from the thoracic, greater omentum and hump regions, respectively. Each fat tissue was immediately submerged in liquid nitrogen, and stored at −80°C for RNA-seq analysis.

RNA sequencing

RNA sequencing was performed by Novogene Bioinformatics Technology Co., Ltd, Beijing, China. The main steps are described below. Total RNA was extracted from the sample. The sample quality was tested. The mRNA in the sample was purified with Oligo (dT) attached magnetic beads. The mRNA was then broken into short fragments and used as a template to synthesise double-stranded cDNA. The end was repaired, the A tail was added, and the adaptor was connected, the fragment size was selected. Then the cDNA library was enriched by PCR and library quality was assessed. Finally, High-throughput sequencing was performed on cDNA that passed the library inspection.

Bioinformatics analysis

First, the quality of the high-throughput raw data obtained by sequencing was evaluated, including sequencing error rate distribution check, GC content distribution check and sequencing data filtering. Hisat2 was then used to align to the reference genome. FKPM (fragments per kilo bases per million reads) was used to estimate gene expression in samples and padj < 0.005 was used as the threshold for screening differentially expressed genes (DEGs). The involved signaling pathways were analysed by comparison with KEGG.

Results and Discussion

Summary of transcriptome sequencing data

We sequenced 9 cDNA libraries from 3 adipose depots from Bactrian camel. Three replicates each of HF, SF and GOM depots, 59, 152, 350, 44, 336, 816 and 65, 552, 914 raw reads were obtained for HF; 57, 151, 528, 50, 973, 870 and 46, 332, 920 raw reads were obtained for SF; 55, 584, 566, 65, 647, 414 and 54, 536, 066 raw reads were obtained for GOM. The raw reads were filtered to obtain clean reads, which were then aligned to the camel reference genome using Hisat2. 88.52%~91.05% of the total sequenced fragments could be mapped to the reference genome (Table 1).

Identification of differentially expressed genes

We identified 723, 1, 460, 1, 757 genes showing differential expression between HF and SF, HF and GOM, SF and GOM, respectively, and were clustered by a visual heat map (Fig 1). Of 723 differentially expressed genes between HF and SF, 371 showed higher expression in the HF versus 352 genes showed higher expression in the SF. Of 1, 460 genes between HF and GOM, 502 expressed higher in the HF versus 958 expressed higher in the GOM. Of 1, 757 genes between SF and GOM, 570 expressed higher in the SF versus 1, 187 expressed higher in the GOM. HF vs. GOM and SF vs. GOM had greater differences than HF vs. SF at the transcriptional level. We found more than 20 genes involved in fat metabolism (Table 2).

Pathway analysis of differentially expressed genes

Pathway annotation of DEGs was performed using the KEGG database. The DEGs of HF vs. SF were significantly enriched in pathways including Cell adhesion molecules and ECM-receptor interaction. The up-regulated genes in HF vs. GOM were significantly 5 most enriched in pathways including AMPK signaling pathway,

Table 1. Statistics for filtering and mapping reads.

Sample	Raw reads	Clean reads	Total map	Clean bases	Error rate	Q20	Q30	GC(%)
HF1	59152350	58138948	52363415(90.07%)	8.72G	0.03	97.57	93.26	52.18
HF2	44336816	43320378	38969395(89.96%)	6.5G	0.03	97.03	92.19	50.5
HF3	65552914	63262250	56365092(89.1%)	9.49G	0.03	97.82	94.07	52.25
SF1	57151528	55824084	49754737(89.13%)	8.37G	0.03	96.75	91.6	49.54
SF2	50973870	49557888	44449340(89.69%)	7.43G	0.03	97.81	94.04	52.54
SF3	46332920	45363498	41304461(91.05%)	6.8G	0.03	97.09	92.15	51.17
GOM1	55584566	54503656	49061874(90.02%)	8.18G	0.03	96.91	91.88	51.04
GOM2	65647414	64780532	58769365(90.72%)	9.72G	0.03	97.78	93.73	52.29
GOM3	54536066	53184776	47077685(88.52%)	7.98G	0.03	96.88	91.85	50.94

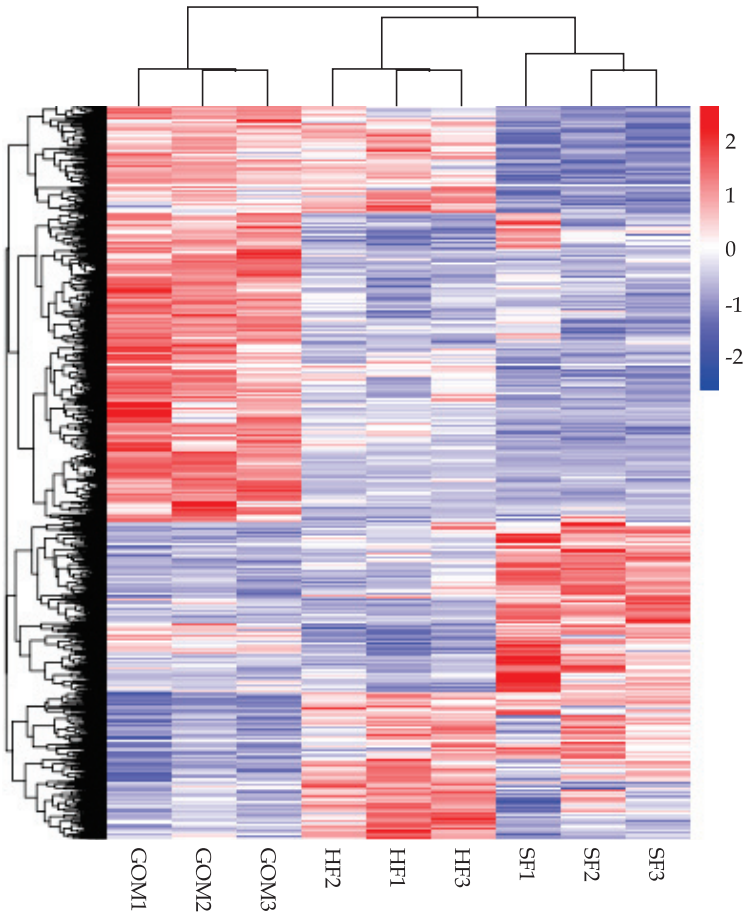

Fig 1. Clustering analysis of DEGs in three adipose depots.

PPAR signaling pathway, Adipocytokine signaling pathway, ECM-receptor interaction and carbon metabolism. The down-regulated genes in HF vs. GOM were significantly 5 most enriched in pathways including cytokine-cytokine receptor interaction, Hippo signaling pathway, Cell adhesion molecules (CAMs), Complement and coagulation cascades and Rap1 signaling pathway. The up-regulated genes in SF vs. GOM were significantly enriched in pathways including ECM-receptor interaction, Fatty acid metabolism, Biosynthesis of unsaturated fatty acids, Fatty acid elongation and glycosaminoglycan biosynthesis-keratan sulfate. The down-regulated genes in SF vs. GOM were significantly 5 most enriched in pathways including cytokine-cytokine receptor interaction, Cell adhesion molecules (CAMs), Chemokine signaling pathway, Epstein-Barr virus infection and Rap1 signaling pathway (Table 3).

In this study, SF, HF and GOM of Bactrian camel were sequenced by transcriptome sequencing technology, and the different genes of fat tissue in 3 different parts at transcriptome level were obtained by paired comparison. Among the differentially expressed genes of HF and GOM, the genes related to fat metabolism in HF were significantly up-regulated.

SCD is a rate-limiting enzyme that dehydrogenates saturated fatty acids to form monounsaturated fatty acids in mammals (Ntambi and Miyazaki, 2004). SCD activity is positively correlated with fat deposition and monounsaturated fatty acids (Jiang *et al,* 2008). The greater accumulation of fat in hump than in omentum may be related to the high expression of SCD. FASN and ACACA are rate-limiting enzymes for novo synthesis of long-chain fatty acids (Smith *et al,* 2003; Zu *et al,* 2013). Feeding carbohydrates to animals after a long period of fasting significantly increased FASN activity (Semenkovich, 1997) The high expression of FASN and ACACA in hump suggests that the Bactrian camel has an important role in the effective synthesis of fatty acids, and the energy storage is very important for Bactrian camel to adapt to the desert environment where food is scarce. ACSS, ACSM and ACSL are involved in the synthesis of fatty acid (van der Sluis and Erasmus, 2016). ELOVL5 and ELOVL6 are involved in the synthesis of monounsaturated fatty acids (Green *et al,* 2010). Monoacylglycerol (MAG) was converted to diacylglycerol (DAG) under the catalysis of MOGAT, and then to TG under the catalysis of DGAT (Liu *et al,* 2012). PPARG is mainly expressed in adipose tissue and liver and is the main regulator of adipogenesis, adipocyte differentiation, proliferation and lipid accumulation (He *et al,* 2013). THRSP plays an important role in fat deposition by regulating the expression of fat-synthesis-related genes such as FASN (Schering *et al,* 2017). The data from this study suggest that these genes involved in fat synthesis play an important role in fat deposition in hump. HSL and LPL mainly catalyse TG to release free fatty acids to participate in the oxidation and energy supply. LPL and HSL are key rate-limiting enzymes that hydrolyse TG in serum and adipocytes, respectively. MGLL hydrolysed MAG into glycerol and fatty acids (Lafontan and Langin, 2009). This suggests that the catabolism of hump is more active than that of omental fat. PLIN has a bidirectional regulation effect on lipolysis and is known as the "molecular switch" in the regulation of lipolysis. In the basal state, the

Table 2. The selected DEGs that were involved in fat metabolism.

Gene symbol	Gene name	FPKM		Log_2 fold change	*P* value	Padj
		HF	GOM			
MOGAT3	monoacylglycerol O-acyltransferase 3	15.38	0	6.45	2E-04	2E-03
SCD	stearoyl-CoA desaturase	6662.46	150.23	5.47	7E-11	6E-09
MOGAT1	monoacylglycerol O-acyltransferase 1	232.92	16.51	3.82	1E-05	3E-04
LEP	leptin	429.20	34.31	3.64	1E-05	3E-04
FASN	fatty acid synthase	31712.11	3398.73	3.22	7E-07	2E-05
PPARA	peroxisome proliferator-activated receptor α	382.72	48.18	2.99	4E-04	4E-03
ACACA	acetyl-CoA carboxylase α	16139.53	2580.54	2.64	1E-09	8E-08
ADIPOQ	adiponectin	106202.05	17555.19	2.60	5E-09	3E-07
PLIN1	perilipin 1	103254.92	19504.69	2.40	1E-06	4E-05
ACSL1	acyl-CoA synthetase long-chain family member 1	25172.84	5375.09	2.23	9E-05	1E-03
ACSS3	acyl-CoA synthetase short-chain family member 3	1204.72	262.14	2.20	2E-04	3E-03
MGLL	monoglyceride lipase	17906.87	4007.07	2.16	1E-06	4E-05
LPL	lipoprotein lipase	38504.93	8656.04	2.15	2E-06	5E-05
THRSP	thyroid hormone responsive	7338.78	1711.07	2.10	1E-06	4E-05
PLIN4	perilipin 4	233662.47	55569.29	2.07	1E-05	2E-04
ACSM1	acyl-CoA synthetase medium-chain family member 1	6736.44	1680.26	2.00	5E-05	9E-04
ADIPOR2	adiponectin receptor 2	11797.80	2963.55	1.99	2E-06	6E-05
ELOVL6	ELOVL fatty acid elongase 6	724.07	186.80	1.95	2E-06	6E-05
HSL(LIPE)	lipase hormone-sensitive	47290.33	12332.88	1.94	5E-05	9E-04
ACSS2	acyl-CoA synthetase short-chain family member 2	5690.35	1560.15	1.87	3E-07	1E-05
DGAT2	diacylglycerol O-acyltransferase 2	1377.71	414.99	1.73	2E-04	2E-03
PPARG	peroxisome proliferator-activated receptor γ	5185.60	1565.88	1.73	4E-06	1E-04
ELOVL5	ELOVL fatty acid elongase 5	5442.25	1664.24	1.71	9E-12	8E-10
Gene symbol	**Gene name**	**FPKM**		**Log_2 fold change**	***P* value**	**Padj**
		HF	**SF**			
ACACA	acetyl-CoA carboxylase α	13449.13	958.51	3.81	1E-17	2E-14
PLIN5	perilipin 5	1367.20	160.42	3.09	6E-06	3E-04
ACACB	acetyl-CoA carboxylase β	319.44	49.01	2.69	3E-05	1E-03
THRSP	thyroid hormone responsive	6137.50	1051.92	2.54	2E-06	1E-04
Gene symbol	**Gene name**	**FPKM**		**Log_2 fold change**	***P* value**	***P* value**
		SF	**GOM**			
SCD	stearoyl-CoA desaturase	24617.53	130.75	7.56	6E-16	1E-13
ELOVL3	ELOVL fatty acid elongase 3	47.71	0.67	5.95	5E-07	1E-05
ELOVL6	ELOVL fatty acid elongase 6	790.94	162.66	2.28	5E-10	3E-08
MGLL	monoglyceride lipase	13756.66	3484.99	1.98	2E-04	2E-03

PLIN package is placed on the surface of the lipid droplets, forming a molecular barrier that prevents lipase from contacting the TG in the lipid droplets, thus inhibiting lipolysis (Wolins *et al,* 2005). As energy demand increases, PLIN is phosphorylated by protein kinase A (PKA), and the barrier is modified to allow lipase to contact and break down TG (Lafontan and Langin, 2009). In this study, the high expression of PLIN1 and PLIN4 in the hump suggested that PLIN1 and PLIN4 play an important role in the fat deposition and lipolysis of the hump.

Adiponectin is one of the adipokines secreted by adipose tissue, which can regulate the energy homeostasis, glucose metabolism and fat metabolism of organisms. By binding to its receptors, AdipoR1 and AdipoR2, adiponectin can activate AMPK, PPAR and p38MAPK, promote the oxidation of fatty acids, and thus reduce the deposition of fat (Fang and Judd, 2018). The high expression of ADIPOQ and ADIPOR2 in hump in this study suggests that ADIPOQ and ADIPOR2 play an important role in maintaining the energy homeostasis of Bactrian camel in the absence of food. Leptin is mainly synthesised and secreted by white adipose tissue, which has the function of regulating fat storage and maintaining energy balance in the body. It can directly act on fat cells, inhibit the synthesis of fat, promote lipolysis, and avoid obesity (Triantafyllou *et al,* 2016). The study has reported that the plasma leptin concentration of camels decreased under the

Table 3. Pathway analysis of differentially expressed genes.

Pathway	**HF vs. SF**	**Gene name**	***P* value**	**Padj**
Cell adhesion molecules	Up	ICAM3/SELL/LOC105076320/CD8A/LOC105064235/SIGLEC1/CLDN15/CD6/CDH5/SELP	8E-05	0.016
ECM-receptor interaction	Down	COMP/FN1/CD44/THBS3/ITGA11/SDC4/THBS4/ITGA9/TNXB/TNC/SV2B	5E-07	9E-05
Pathway	**HF vs. GOM**	**Gene name**	***P* value**	**Padj**
AMPK signaling pathway	Up	SCD/ACACA/LOC105062383/ADIPOQ/HNF4A/FASN/SLC2A4/ADIPOR2/PPARG/LEP/LIPE/PPP2R1B/PCK1	6E-06	6E-04
PPAR signaling pathway	Up	SCD/LOC105062383/ADIPOQ/PLIN1/LPL/PPARG/SORBS1/PLIN4/AQP7/ACSL1/PCK1/PPARA	9E-08	2E-05
Adipocytokine signaling pathway	Up	LOC105062383/ADIPOQ/SLC2A4/ADIPOR2/MAPK10/LEP/SLC2A1/ACSL1/PCK1/PPARA	9E-06	6E-04
ECM-receptor interaction	Up	LOC105062383/LAMB1/COL4A3/COL4A1/COL4A2/HSPG2/TNC/COL1A1/LAMB3/LAMC1	3E-05	1E-03
Carbon metabolism	Up	ACSS2/CS/GPT2/HAO2/PCCA/SUCLA2/GPI/MCEE/PC	2E-03	0.033
Regulation of lipolysis in adipocytes	Up	PTGER3/MGLL/PLIN1/ADCY2/ADORA1/LIPE/AQP7/GNAI1	2E-05	1E-03
Glycerolipid metabolism	Up	GPAM/MGLL/LPL/PNPLA3/MOGAT1/LPIN2/MOGAT3/DGAT2	5E-05	1E-03
Cytokine-cytokine receptor interaction	Down	IL33/IL1RL1/TNFSF13/CXCL8/TGFB2/BMP7/IL1B/TGFB3/CD4/BMP4/IL18R1/IL36G/AMHR2/CLCF1/NGF/IL27RA/CX3CR1/EDAR/IL10RA/CCL21/TNFRSF18/IL1A/CCL24/CXCR3/LOC105064293/CD27/OSM	7E-05	2E-03
Hippo signaling pathway	Down	WNT2B/FZD10/WNT4/CRB2/TGFB2/WNT16/WWC1/WNT10A/BMP7/FZD2/TGFB3/BMP4/ID2/PRKCZ/TEAD3/LEF1/FZD9/ITGB2/WNT10B/CDH1/WNT6/AJUBA/LOC105075564/AMOT/DLG4	2E-07	1E-05
Cell adhesion molecules	Down	ICOSLG/CD4/SIGLEC1/CD8A/CLDN1/SELPLG/ITGA9/ITGB2/PDCD1LG2/CDH1/NTNG2/LOC105064249/LOC105064245/LOC105064248/MADCAM1/SELL/CLDN4/ITGAL/SPN/CADM1/CNTNAP2/LOC105064250	1E-06	7E-05
Complement and coagulation cascades	Down	C3/C4BPA/C4A/BDKRB1/F12/C1R/C1S/LOC105074040/SERPING1/CR2/LOC105062185/CFI/C7/CLU/PROS1/C1QB/ITGB2/VSIG4/C5AR1/F5/CD55	2E-11	5E-09
Rap1 signaling pathway	Down	RGS14/SKAP1/NGF/PRKCZ/PRKCB/ITGB2/PDGFRA/FGF16/RAC2/PDGFC/CDH1/PDGFA/FYB/TIAM1/ADCY7/LOC105079217/FGFR2/PLCB2/ITGAL/LOC105075564/PFN3	2E-03	0.021

Pathway	SF vs. GOM	Gene name	*P* value	Padj
ECM-receptor interaction	Up	COMP/TNC/THBS4/SDC4/COL9A1/COL6A3/CD44/LAMC2/CHAD/COL6A2	1E-04	0.017
Fatty acid metabolism	Up	SCD/ELOVL6/ACSL3/PTPLA/HSD17B12/ELOVL3/TECR	2E-04	0.019
Biosynthesis of unsaturated fatty acids	Up	SCD/ELOVL6/PTPLA/HSD17B12/ELOVL3/TECR	1E-04	0.017
Fatty acid elongation	Up	ELOVL6/PTPLA/HSD17B12/ELOVL3/TECR	9E-04	0.042
Glycosaminoglycan biosynthesis-keratan sulfate	Up	CHST2/LOC105078680/LOC105065926/B4GALT1	8E-04	0.042
Cytokine-cytokine receptor interaction	Down	IL1B/ACKR4/IL1RL1/IL33/AMHR2/LOC105064293/CX3CR1/IL2RG/CD27/LOC105064292/IL10RA/IL18R1/IL36G/CCL16/CD4/CXCR6/ACVRL1/BMP4/EDAR/OSM/TNFSF14/LEPR/CX3CL1/XCL1/NGF/CXCR3/CCR2/IL20RA/CXCL10	3E-04	5E-03
Cell adhesion molecules	Down	CD8A/SIGLEC1/SELL/LOC105064235/SPN/CLDN1/LOC105064249/CLDN15/CD6/LOC105064250/ITGB7/SELP/ITGB2/ITGAM/ICAM3/CDH1/CD4/CD2/LOC105064239/SELPLG/ITGAL/CLDN11/MADCAM1/LOC105064248/LOC105074084/PDCD1LG2	2E-07	1E-05
Chemokine signaling pathway	Down	JAK3/ARRB2/SHC2/RAC2/PLCB2/LOC105064293/CX3CR1/NCF1/RASGRP2/LOC105064292/CCL16/CXCR6/PIK3CG/HCK/WAS/PIK3R6/CX3CL1/ITK/XCL1/PRKCB/PTK2B/CXCR3/CCR2/GNB5/PIK3R5/CXCL10	3E-05	8E-04
Epstein-Barr virus infection	Down	JAK3/LOC105064235/CD3E/CCNA2/LOC105064249/BLNK/LOC105064250/CD19/CD3G/CCND3/BTK/MAPK12/LOC105064239/ITGAL/CD247/MAPK14/LOC105064248/LOC105074084/IKBKE/BAK1/ENTPD1/PLCG2/CXCL10	2E-03	0.023
Rap1 signaling pathway	Down	RAC2/SIPA1/PLCB2/RASGRP2/ITGB2/ITGAM/CDH1/TEK/LOC105079217/LCP2/RGS14/FYB/MAPK12/ITGAL/FLT4/ARAP3/PRKCB/MAPK14/NGF/PFN3/LOC105075564/CALML4/PRKD2	5E-03	0.043

condition of insufficient feed and increased under the condition of overfeeding. This suggests that leptin plays an important role in maintaining the energy balance of Bactrian camel.

In HF vs. GOM, genes with increased HF than GOM expression were enriched in several pathways involved in lipid metabolism. These proved that HF metabolism is more active than omental fat metabolism. In the Cytokine-cytokine receptor interaction, cytokine genes and their receptors were expressed significantly higher in GOM than HF and SF. These data suggests that GOM is more likely to cause inflammatory response than HF and SF.

In conclusion, we sequenced the transcriptome of the 3 sources of adipose tissues in Bactrian camel using Illumina Hiseq sequencing platform. DEGs were identified between subcutaneous, visceral and hump fat tissues. The KEGG enrichment were analysed. Our results provide new insight into understanding of differences between adipose depots and providing new insight into exploring the specific fat deposition in hump. Some genes related to fat metabolism in HF may play an important role in maintaining the energy homeostasis of Bactrian camels in the absence of food. These data suggests the hump fat has a greater capacity to modulate the release and storage of TG in adipocytes than omentum.

Acknowledgements

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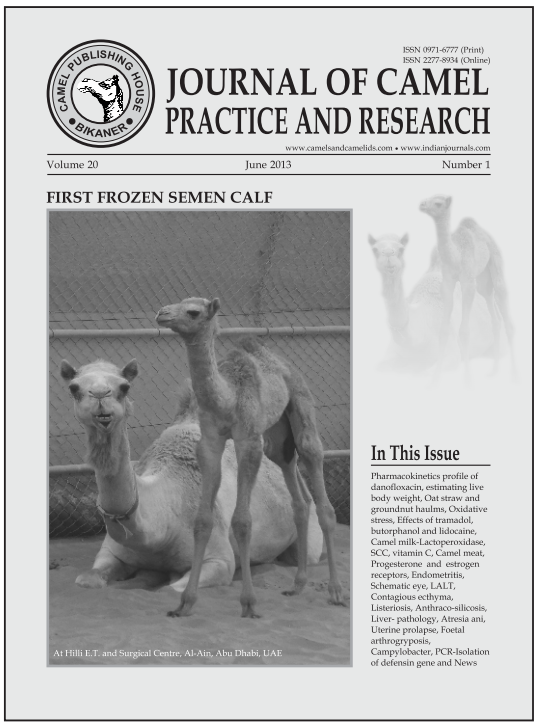

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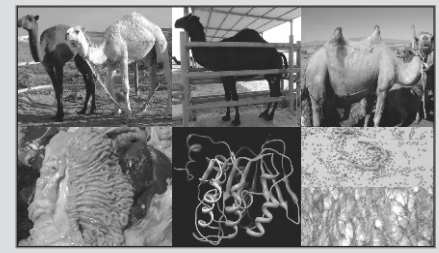

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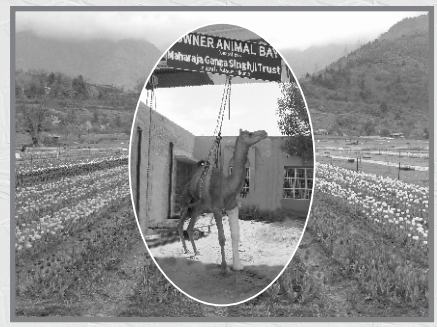

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IMPACT OF AGE AND PREGNANCY ON THE EXPRESSION LEVEL OF THE CORONAVIRUS (MERS-COV) RECEPTOR, DIPEPTIDYL PEPTIDASE 4 ON CAMEL BLOOD LEUKOCYTES

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ABSTRACT

In dromedary camels, young animals have shown higher susceptibility to MERS-CoV infection than adult camels. The current study, analysed the impact of animal age and pregnancy on the expression density of DPP4 on the main populations of camel leukocytes using flow cytometry.

Adult camels showed significantly higher expression levels of DPP4 on their monocytes in comparison to newborn calves. Newborn calves and adult camels showed comparable expression levels of DPP4 on their neutrophils and lymphocytes. The results of the current study argue against a role of different DPP4 expression levels on blood leukocytes in the higher susceptibility of camel calves than to MERS-CoV. The comparable expression levels of DPP4 on monocytes, neutrophils, and lymphocytes from pregnant and non-pregnant female camels indicate no significant impact of pregnancy on DPP4 expression on blood leukocytes of camels.

Key words: Camel, corona virus, dipeptidyl peptidase 4, MERS-CoV), middle east

Dipeptidyl peptidase 4 (DPP4), which is a type II transmembrane glycoprotein involved in cleavage of dipeptides and degradation of incretins (Lambeir *et al,* 2003), has been identified as a functional receptor for the Middle East respiratory syndrome coronavirus (MERS-CoV) (Ohnuma *et al,* 2013; Raj *et al,* 2013; van Doremalen *et al,* 2014). Differential expression of DPP4 in the respiratory tracts has been shown to be responsible for higher susceptibility of humans thandromedary camels for MERS-CoV infection (Widagdo *et al,* 2016). In opposite to human, where DPP4 is mainly found on human T lymphocytes rather than monocytes (Pierson *et al,* 2008), dromedary camels have shown the highest expression of DPP4 on their blood monocytes (Al-Mubarak, 2018; Haverkamp *et al,* 2018).

Age-related changes of several innate and adaptive cellular immune responses have been described for different species (Romanyukha and Yashin, 2003; Elghetany and Lacombe, 2004), including dromedary camel (Gaashan *et al,* 2020). In dromedary camels (Hussen *et al,* 2020) as well as in several other animals species (Leung *et al,* 2000; Oliveira *et al,* 2012; Spadaro *et al,* 2019), pregnancy is associated with modulations in several components of the immune system.

Expression pattern or density of DPP4 on blood leukocytes and its associated alterations by animal age or pregnancy has not been studied previously. The current study, therefore, comparatively analysed the expression of DPP4 on the main populations of peripheral blood leukocytes in newborn and adult camels and, during pregnant and non-pregnant states female camels.

Materials and Methods

Animals and blood sampling

For the comparison between newborn calves and adult camels regarding the expression of DPP4, blood samples were collected from 14 newborn camel calves (aged between 7 and 35 days) and 10 adult dromedary camels (*Camelus dromedarius*) aged between 7 and 9 years. For the evaluation of the impact of pregnancy on the expression level of DPP4 on blood leukocytes, blood samples were collected into vacutainer tubes containing EDTA from 12 pregnant and 9 non-pregnant apparently healthy she-camels. All camels were housed at the farm of

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the Camel Research Center, King Faisal University, Al-Ahsa, Saudi Arabia. All experimental procedures and management conditions used in this study were approved by the Ethics Committee at King Faisal University, Saudi Arabia (Permission number: KFU-REC/2020-09-25).

Isolation of leukocytes from camel blood

Separation of camel leukocytes was performed after hypotonic lysis of blood erythrocytes as described previously (Hussen *et al,* 2017). Briefly, blood was suspended in distilled water for 20 sec and double concentrated PBS was added to restore tonicity. This was repeated (usually twice) until complete erythrolysis. Separated cells were finally suspended in MIF buffer (PBS containing bovine serum albumin (5 g/L) and NaN3 (0.1 g/L)) at 5 x 10^6 cells/ml. Cell purity of separated leukocytes was assessed by flowcytometry according to their FCS/SSC properties and always exceeded 90 %. The mean viability of separated cells was evaluated by dye exclusion (propidium iodide; 2 µg/ml, Calbiochem, Germany) and it was above 90%.

Immunofluorescence and flow cytometry

After leukocyte separation, separated cells (1 x 10^6) were labeled with monoclonal antibodies against human DPP4 (goat IgG anti DPP4; R & D Systems) with cross-reactivity with camel DPP4 (Pierson *et al,* 2008; van Doremalen *et al,* 2014) diluted 1: 50 in PBS containing bovine serum albumin (5 g/l) and NaN3 (0.1 g/l). After incubation at 4°C for 20 minutes, the cells were washed twice and further incubated with a secondary antibody against goat IgG labeled with fluorochrome (Alexa Fluor 488a-labelled rabbit F (ab') 2-anti-goat IgG (H+L); Invitrogen). After incubation (20 minutes; 4°C), labelled cells were washed twice and analysed on the flow cytometer (Fig 1A). A Becton Dickinson FACS Calibur equipped with Cell Quest software (FACSCalibur; Becton Dickinson Biosciences, San Jose, California, USA) was used to collect the data. At least 100 000 cells were collected and analysed with the flowcytometric software FCS Express software Version 3 (De Novo Software, Thornton, Ontario).

Statistical Analyses

Statistical analysis was performed with Prism (GraphPad). Results are presented as means ± S.E. of the mean (SEM). Student *t test* was used for difference analysis between means. Differences were considered statistically significant at a p-value of less than 0.05.

Results and Discussion

In dromedary camels, young camels have shown higher susceptibility to MERS-CoV infection than adult camels (Khalafalla *et al,* 2015). Dipeptidyl peptidase 4 (DPP4) has been identified as a functional receptor for the MERS-CoV (Ohnuma *et al,* 2013; Raj *et al,* 2013; van Doremalen *et al,* 2014). The expression level of DPP4 on the surface of cells of respiratory tract has been found to be correlated positively with susceptibility to MERS-CoV infection in human (Cai *et al,* 2014; Meyerholz *et al,* 2016). In addition, the differential expression of DPP4 in the upper respiratory tracts has been shown to be responsible for higher susceptibility of humans thandromedary camels for MERS-CoV infection (Widagdo *et al,* 2016). Whether a different expression pattern or density of DPP4 on blood leukocytes contribute to this higher disease susceptibility in newborn camel calves than adult camels, it is unknown. The current study, therefore, analysed the impact of animal age on the expression density of DPP4 on the main populations of peripheral blood leukocytes of dromedary camels.

As shown in Fig 1B, monocytes from both newborn and adult camels expressed the highest levels of DPP4 when compared with neutrophils and lymphocytes (Fig 1B). The comparison between newborn calves and adult camels regarding the expression density of DPP4 bon blood leukocytes revealed significantly higher expression levels on monocytes from adult camels than monocytes from calves (Fig 1B). For lymphocytes and neutrophils, however, newborn and adult camels showed comparable expression levels of DPP4 on their cells. Although the finding of the current study are in line with previous studies, showing highest DPP4 expression levels on camel monocytes (Al-Mubarak, 2018; Haverkamp *et al,* 2018), the results, however, argue against a role of different DPP4 expression levels on blood leukocytes in the higher susceptibility of camel calves than adults to MERS-CoV, as DPP4 is higher expressed on adult monocytes than calf monocytes. The evaluation of clinical importance of the higher abundance of DPP4 on adult monocytes needs further investigation.

The expression density of DPP4 on the surface of blood leukocytes from newborn and adult dromedary camels.

Separated camel leukocytes were labeled with goat monoclonal antibodies to DPP4 and anti goat IgG secondary antibodies and labeled cells were analysed by flow cytometry. A)After gating on leukocytes,

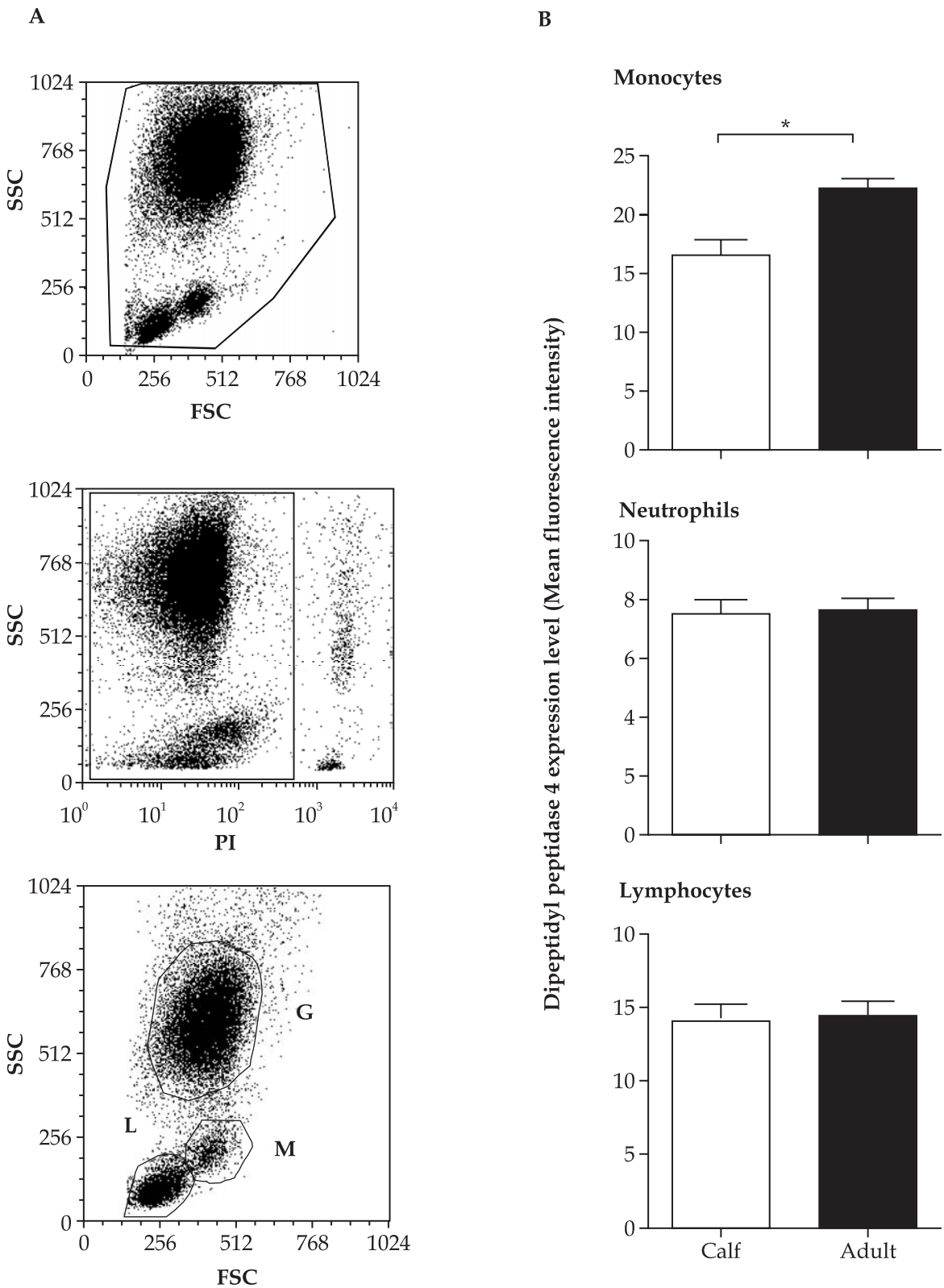

Fig 1. The expression density of DPP4 on the surface of blood leukocytes from newborn and adult dromedary camels. Separated camel leukocytes were labeled with goat monoclonal antibodies to DPP4 and anti goat IgG secondary antibodies and labeled cells were analysed by flow cytometry. **A)** After gating on leukocytes, dead cells were excluded from the analysis by gating on propidium iodide negative cells. Based on their FSC/SSC properties, camel neutrophilic granulocytes (G), monocytes (M), and lymphocytes (L) were gated and the mean fluorescence intensity of DPP4 was calculated for each cell type. **B)** Data for cells from newborn and adult camels were presented as mean +- SEM (* = $p<0.05$).

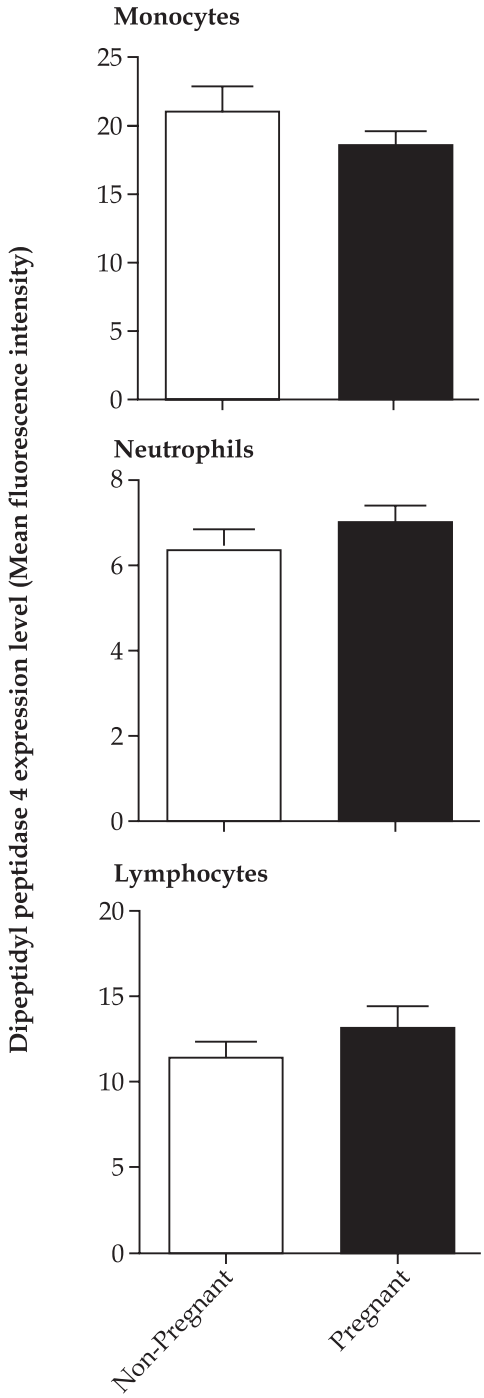

Fig 2. The impact of pregnancy on the expression density of DPP4 on the surface of camel blood leukocytes. Separated camel leukocytes were labeled with goat monoclonal antibodies to DPP4 and anti goat IgG secondary antibodies and labeled cells were analysed by flow cytometry. After gating on leukocytes, dead cells were excluded from the analysis by gating on propidium iodide negative cells. Based on their FSC/SSC properties, camel neutrophilic granulocytes, monocytes, and lymphocytes were gated and the mean fluorescence intensity of DPP4 was calculated for each cell type. Data for cells from pregnant and non-pregnant animals were presented as mean +- SEM (* = p<0.05).

dead cells were excluded from the analysis by gating on propidium iodide negative cells. Based on their FSC/SSC properties, camel neutrophilic granulocytes (G), monocytes (M), and lymphocytes (L) were gated and the mean fluorescence intensity of DPP4 was calculated for each cell type. B) Data for cells from newborn and adult camels were presented as mean ± SEM (* = p<0.05).

Pregnancy is a physiologic process with several changes in the immunophenotype of camel blood leukocytes (Hussen *et al*, 2019). It is unknown, whether the expression density of DPP4 on blood leukocytes is affected by pregnancy in the dromedary she-camel. In the present study, the comparison between pregnant and non-pregnant female dromedary camels regarding the expression levels of DPP4 on their monocytes, neutrophils, and lymphocytes revealed no significant differences between the two groups (Fig 2). Although this indicates no significant effect of pregnancy on DPP4 expression on blood leukocytes, a direct pregnancy-associated change in DPP4 expression in the lung tissues can not be excluded.

The impact of pregnancy on the expression density of DPP4 on the surface of camel blood leukocytes.

Separated camel leukocytes were labeled with goat monoclonal antibodies to DPP4 and anti goat IgG secondary antibodies and labeled cells were analysed by flow cytometry. After gating on leukocytes, dead cells were excluded from the analysis by gating on propidium iodide negative cells. Based on their FSC/SSC properties, camel neutrophilic granulocytes, monocytes, and lymphocytes were gated and the mean fluorescence intensity of DPP4 was calculated for each cell type. Data for cells from pregnant and non-pregnant animals were presented as mean ± SEM (* = p<0.05).

Conclusions

The comparison between newborn calves and adult camels regarding the expression density of DPP4 on blood leukocytes revealed significantly higher expression levels on monocytes from adult camels than monocytes from calves. Newborn calves and adult camels showed comparable expression levels of DPP4 on their neutrophils and lymphocytes. The results of the current study argue against a role of different DPP4 expression levels on blood leukocytes in the higher susceptibility of camel calves to MERS-CoV. The comparable expression levels of DPP4 on monocytes, neutrophils, and lymphocytes

from pregnant and non-pregnant female camels indicates no significant impact of pregnancy on DPP4 expression on blood leukocytes of camels. Further work is needed to explore the impact of animal age and pregnancy on the local expression of DPP4 in the respiratory system of dromedary camel.

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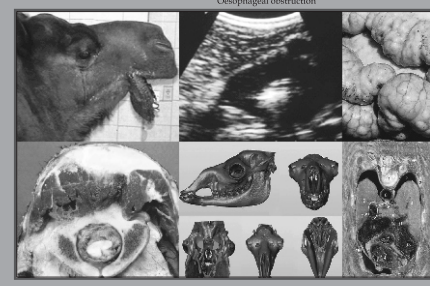

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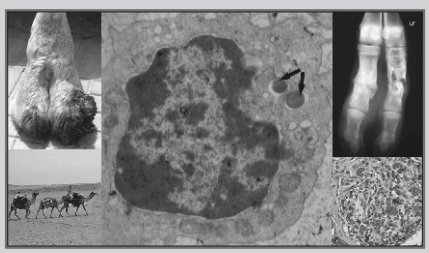

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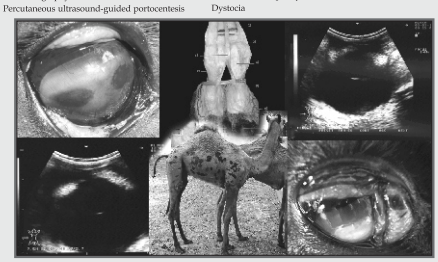

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OXIDATIVE STRESS, HISTOPATHOLOGICAL AND HAEMATO-BIOCHEMICAL FEATURES IN DROMEDARY CAMELS INTOXICATED WITH GLIOTOXIN

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ABSTRACT

The aim of the current study was to determine the effect of intravenous administration of gliotoxin on oxidative stress, histopathological and haemato-biochemical features in dromedary camels before injection (0h) and at 1, 24, 48 and 72 hours post injection. Five healthy adult female camels (8-10 years old having 300-350 kg b.wt.) were injected intravenously with gliotoxin (0.025µg/kg b.wt.). Blood samples were collected in plain and heparinised vacutainers at all time points (0, 1, 24, 48 and 72h). The whole blood in heparinised vacutainers was used for estimation of haematological parameters. Obtained sera stored frozen at -30°C until used for estimation of biochemical and oxidative stress biomarkers. Liver and heart tissues were collected from one camel died 24 hours post gliotoxin injection and subjected for histopathological examination. The findings revealed significant increase in the values of ALT, AST, urea, creatinine and neutrophils along with significant reduction in values of total leucocyte count (TLC), total erythrocyte count (TEC), hemoglobin (Hb), lymphocytes percentages, glucose, total proteins, albumin and globulin concentrations in gliotoxin treated camels 1, 24, 48 and 72 hour post gliotoxin injection. Gliotoxin induced significant increase in malondialdehyde (MDA) concentration accompanied with significant decrease in the activities of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPX), glutathione S-transferase (GST) and glutathione reductase (GR) at all time points post injection. Postmortem examination of the dead camel showed congestion and oedema in both lungs. Histopathological findings revealed presence of homogenous eosinophilic fluid within alveoli and congestion of interalveolar capillaries and within the myocardium. In conclusion, the injected dose of gliotoxin was acutely toxic to camels as reflected on disturbed liver and kidney functions, acceleration of oxidative stress and inhibition of antioxidants enzyme activities.

Key words: Camel, gliotoxin, mycotoxin oxidative stress, intoxicated

Fungi and their mycotoxins are widely distributed in the environment. After production, these are adsorbed onto airborne dusts and caused major public health problems (Eshetu *et al,* 2016). Gliotoxin is one of the most serious mycotoxin, as low concentration of this toxin is able to induce adverse effects on animal and human health (Bossou *et al,* 2017) and is produced by a number of fungi including *Aspergillus fumigatus* (Bauer, 1994; Eichner *et al,* 1988). Gliotoxin, a hydrophobic fungal metabolite of the epipolythiodioxopiperazine (ETP) group with a quinoid moiety and a disulfide bridge across the piperazine ring (Waring *et al,* 1988). Intravenous injection of gliotoxin (0.2 µg kg^{-1} b.wt.) once killed all the three camels tested in a pilot study of our previous work (Shathele, 2009). Therefore, half (0.1 µg kg^{-1} b.wt.) of the fatal dose of gliotoxin was injected intravenously once in five camels in the same study (Shathele, 2009). This dose (0.1 µg kg^{-1} b.wt.; Shathele, 2009) did not kill the tested camels but decreased the serum total protein and glucose concentrations accompanied with disturbed liver

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and renal function in dromedary camels 2 days post injection. Furthermore, half dose (0.05 µg kg^{-1} b.wt.) of gliotoxin was used in our previous work (Shathele, 2009) where it was injected intravenously for 3 days in five camels (Shathele, 2011). This dose (0.05 µg kg^{-1} b.wt.; Shathele, 2011) led to significant reduction in serum total protein, albumin, globulin, leukocytes, lymphocytes, neutrophils and lysosomal activity on day 7 post injection. The previous studies did not focus on the influence of gliotoxin injection on oxidative stress biomarkers and histopathology. Therefore, the present study was aimed to investigate the effect of intravenous administration of half dose (0.025µg/kg b.wt.) of the last examined dose of gliotoxin (Shathele, 2011) on oxidative stress, histopathological and haemato-biochemical examination before injection (0h) and at 1, 24, 48 and 72 hours post injection in dromedary camels.

Materials and Methods

Experimental Animals and gliotoxin injection

Five healthy adult female camels (8-10 years old, 300-350 kg body weight) were used in the present study. The animals were provided by camel research centre, King Faisal University, Saudi Arabia, kept in an open yard with free access to food and water. Animals were injected intravenously with Gliotoxin (0.025µg/kg b.wt; Sigma, UK). Blood samples were collected in plain and heparinised vacutainers before injection (0h) and at 1, 24, 48 and 72 hours post injection. The whole blood in heparinised vacutainers were used for estimation of haematological parameters. After centrifugation (3000rpm/10 minutes), the obtained sera samples were stored frozen at -30°C until used for estimation of biochemical and oxidative stress biomarkers. Liver and heart tissues were collected from one camel died 24 hours post gliotoxin injection and subjected for histopathological examination.

Haematological and biochemical analysis

Complete blood picture was determined by using electronic cell counter (VetScan HM5 Haematology system). Commercial diagnostic kits (United Diagnostic Industry, Dammam, Saudi Arabia) were used for determination of serum glucose (EP37L-660), total proteins (EP56-660) and albumin (EP03-570), ALT (EP07-500), AST (EP15-500), BUN (EP20-420), Uric acid (EP61-620) and Creatinine (EP33K-660) on ELIPSE full automated chemistry analyser (Rome, Italy). Concentration of the biochemical constituents were calculated according to the manufacturer's instruction.

Analysis of oxidative stress biomarkers

The ELISA kits of Cayman Chemical Company, USA were used for determination of all serum oxidative stress biomarkers. The concentration of MDA (catalogue #10010263) and the activities of total SOD (U/ gram tissue; catalogue #706002), CAT (nmol/min/gram tissue; catalogue #707002), GPX (nmol/min/ gram tissue; catalogue #703102), GST (nmol/min/ gram tissue; catalogue #703302) and GR (µM; catalogue #703202) were measured in the serum by using an ELISA reader (Absorbance Microplate Reader ELx 800TM BioTek®, USA). The results were calculated according to the manufacturer's instruction.

Histopathological analysis

Lung and heart tissues were cut into small pieces and immersed in neutral buffered formalin for 24 hours. The fixed tissues were processed routinely, embedded in paraffin, sectioned, deparaffinised and rehydrated using standard techniques (Bancroft and Gamble, 2002). The extent of gliotoxin toxicity was evaluated by assessing the morphological changes in the lung and sections stained with haematoxylin and eosin (H&E) using standard techniques (Bancroft and Gamble, 2002).

Statistical analysis

All data were presented as the mean ± standard error of the mean (SEM) using analysis of variance (ANOVA). All tests were performed using a statistical analysis system program (SAS, 2002).

Results and Discussion

The effect of intravenous administration of Gliotoxin (0.025µg/kg b.wt.) on haematological parameters before (0 h) and at 1, 24, 48 and 72 hours post injection are presented in table 1. Total erythrocyte counts, total leucocyte counts, hemoglobin and haematocrit percentage were significantly decreased at 1, 24, 48 and 72 hours post injection as compared to its value before injection (0 hour). The values of these parameters at 24 hours post injection and onwards were significantly lower than that at 1-hour post injection. The current findings were similar to that observed earlier in dromedary camels (Shathele, 2011), rams (Dönmez *et al,* 2012) and broilers (Mohamed and Mohamed, 2009). The reductions of RBC and WBC were observed previously (Chattopadhyay *et al,* 2013) in all *Fusarium* mycotoxins treated rats. Data presented in table 2 showed a significant decrease in lymphocyte percentage accompanied

Table 1. Effect of intravenous administration of Gliotoxin (0.025µg/kg b.wt.) on haematological parameters before (0 h) and at 1, 24, 48 and 72 hours post injection.

Variables	0 h	1 h	24 h	48	72
TEC (10^{12}/L)	8.9 ± 0.3[a]	7.4 ± 0.2[b]	5.6 ± 0.2[c]	5.9 ± 0.2[c]	6.0 ± 0.2[c]
Hb (g/dl)	11.1 ± 0.3[a]	9.1 ± 0.3[b]	7.1 ± 0.4[c]	7.4 ± 0.4[c]	7.2 ± 0.4[c]
Hematocrit (%)	28.4 ± 0.9[a]	25.1 ± 0.5[b]	21.8 ± 0.3[c]	21.4 ± 0.3[c]	21.6 ± 0.3[c]
TLC (10^9/L)	15.6 ± 0.2[a]	10.6 ± 0.3[b]	8.1 ± 0.3[c]	8.3 ± 0.3[c]	8.0 ± 0.3[c]
Neutrophils (%)	66.0 ± 1.1[a]	53.0 ± 0.9[b]	49.0 ± 1.1[c]	49.0 ± 1.2[c]	49.0 ± 1.1[c]
Lymphocytes (%)	29.1 ± 1.6[a]	42.1 ± 2.0[b]	46.1 ± 0.9[b]	46.1 ± 1.1[c]	46.1 ± 1.1[c]
Monocytes (%)	2.1 ± 1.2	2.3 ± 1.2	2.3 ± 1.2	2.3 ± 1.2	2.3 ± 1.2
Basophils (%)	0.6 ± 0.1	0.6 ± 0.1	0.6 ± 0.1	0.6 ± 0.1	0.6 ± 0.1
Eosinophils (%)	1.5 ± 0.5	2.0 ± 0.5	2.1 ± 0.5	2.1 ± 0.5	1.9 ± 0.5

Values are mean ± SEM of 5 camels

[a-c]Within the same raw with different superscripts differ significantly (P <0.05).

TEC: total erythrocyte count; Hb: hemoglobin; TLC: total leucocyte count.

Table 2. Effect of intravenous administration of gliotoxin (0.025µg/kg b.wt.) on selected serum biochemical parameters before (0 h) and at 1, 24, 48 and 72 hours post injection.

Parameters	0 h	1 h	24 h	48 h	72 h
Glucose (mg/dl)	83.33 ± 1.0[a]	72 ± 1.9[b]	65 ± 2.1[c]	66 ± 2.5[c]	68 ± 2.4[c]
Total Protein (g/l)	8.0 ± 0.3[a]	6.0 ± 0.3[b]	4.9 ± 0.3[c]	5.0 ± 0.4[c]	5.1 ± 0.4[c]
Albumin (g/l)	6.0 ± 0.1[a]	5.5 ± 0.1[b]	4.7 ± 0.1[c]	4.8 ± 0.1[c]	4.8 ± 0.1[c]
Globulin (g/l)	2.0 ± 0.1[a]	1.5 ± 0.0[b]	0.2 ± 0.0[c]	0. 2 ± 0.0[c]	0.2 ± 0.0[c]
ALT (U/l)	22.44 ± 1.1[a]	34.7 ± 1.1[b]	40.7 ± 1.1[c]	40.7 ± 1.0[c]	38.7 ± 1.0[c]
AST (U/l)	45.21 ± 2.4[a]	70.70 ± 3.1[b]	86.70 ± 2.5[c]	87.70 ± 1.8[c]	85.70 ± 2.8[c]
BUN (mg/dl)	18.70 ± 1.6[a]	25.5 ± 1.4[b]	35.5 ± 1.6[c]	34.5 ± 1.3[c]	37.5 ± 1.9[c]
Creatinine (mg/dl)	1.0 ± 0.1[a]	1.4 ± 0.1[b]	1.8 ± 0.1[c]	1.8 ± 0.1[c]	1.8 ± 0.1[c]

Values are mean ± SEM of 5 camels

[a-c]Within the same raw with different superscripts differ significantly (P < 0.05).

ALT: alanine transaminase; AST: aspartate transaminase; BUN: blood urea nitrogen

by significant increase in neutrophil percentage at 1, 24, 48 and 72 hours post injection as compared to its value before injection (0 hour). The observed lymphocytopenia and neutrophilia at 24 hours post injection and onwards were augmented significantly as compared to their values at 1 hour post injection. Observed lymphocytopaenia may be attributed to the toxic effects of gliotoxin on cells in the peripheral circulation or suppression of bone marrow and lymphoid organs function as indicated earlier for aflatoxicosis (Valchev *et al,* 2018). The mycotoxins lead to haematopoietic suppression and anaemia by reduction of red blood cells and hemoglobin (Reddy and Waliyar, 2012). Reduced haematocrit, haemoglobin, erythrocytes and lymphocyte percentage was observed in broiler chickens with experimental aflatoxicosis, probably due to the inhibitory effect of aflatoxins on haematopoietic organs (Mohamed and Mohamed, 2009). This decrease in red blood parameters may result from inhibition of protein biosynthesis (Kubena *et al,* 1993; Abdel-Wahhab, *et al,* 2002) or the faster degradation of erythrocytes in the spleen (Mokif and Muiz, 2015). Increase in the neutrophils percentages might indicate a tendency of the organism to compensate for the decrease of its resistance (Sova *et al,* 1991). The effect of intravenous administration of gliotoxin (0.025µg/kg b.wt.) on selected biochemical parameters before (0 h) and at 1, 24, 48 and 72 hours post injection were presented (Table 2). Total proteins, albumin, and globulin were decreased significantly at 1, 24, 48 and 72 hours post injection compared to its value before injection (0 hour). The values of these parameters at 24 hours and onwards post injection were significantly lower than that at 1-hour post injection. The current findings were similar to that observed earlier in dromedary camels (Shathele, 2009; 2011) and fish (Kaur and Saxena, 2018). This inhibition in protein biosynthesis (total proteins, albumin and globulin) supports the reduction of haematological

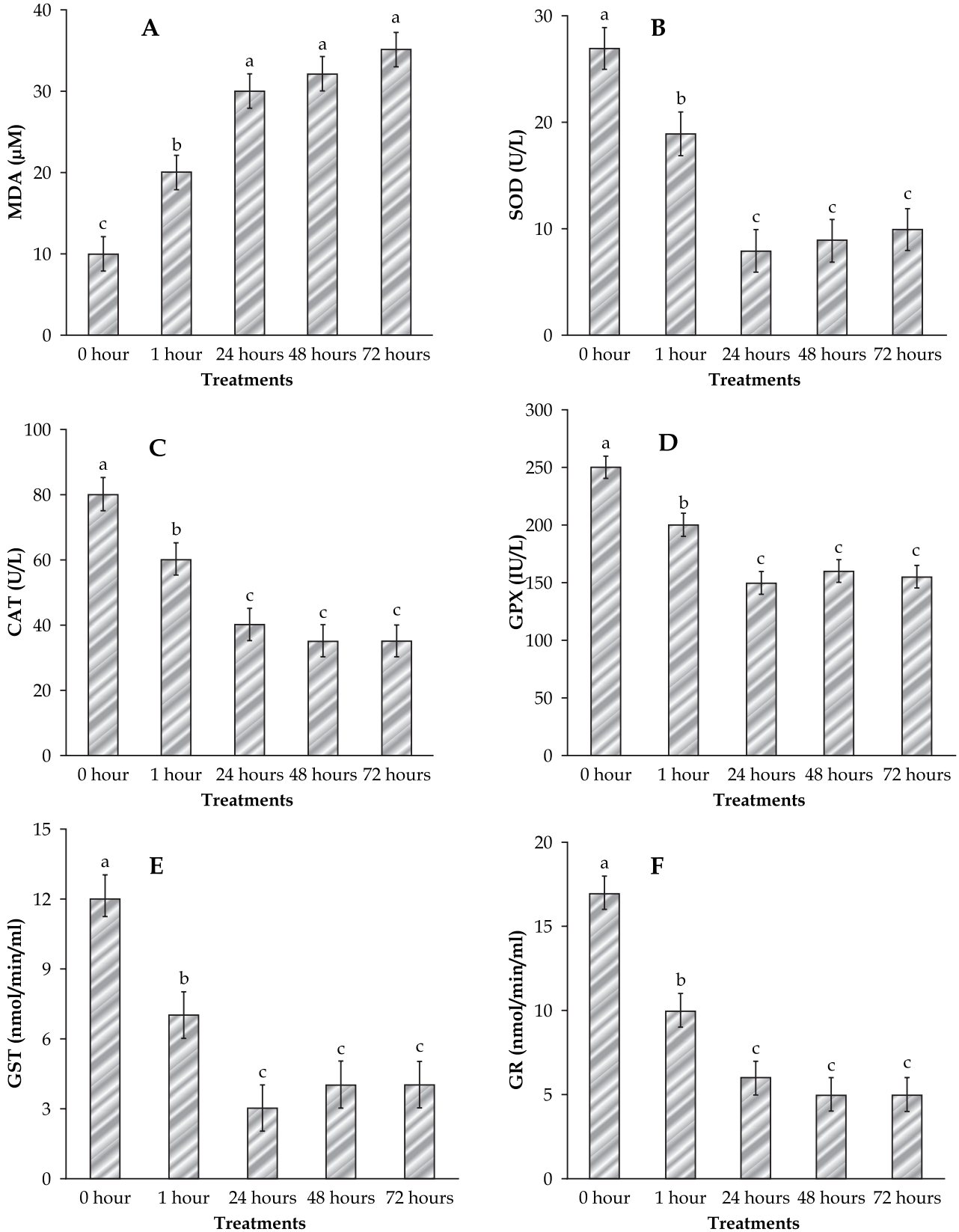

Fig 1. Values of serum oxidative stress biomarkers **(A)** MDA, **(B)** SOD, **(C)** CAT, **(D)** GPX, **(E)** GST and **(F)** RG before injection (0h) and at 1h, 24h, 48h, and 72 hours post intravenous administration of Gliotoxin (0.025µg/kg b.wt.) in dromedary camels. MDA; malonaldhyde; SOD: superoxide dismutase; CAT: catalase; GPX: glutathione peroxidase; GST: glutathione-s-transferase; GR: glutathione reductase.

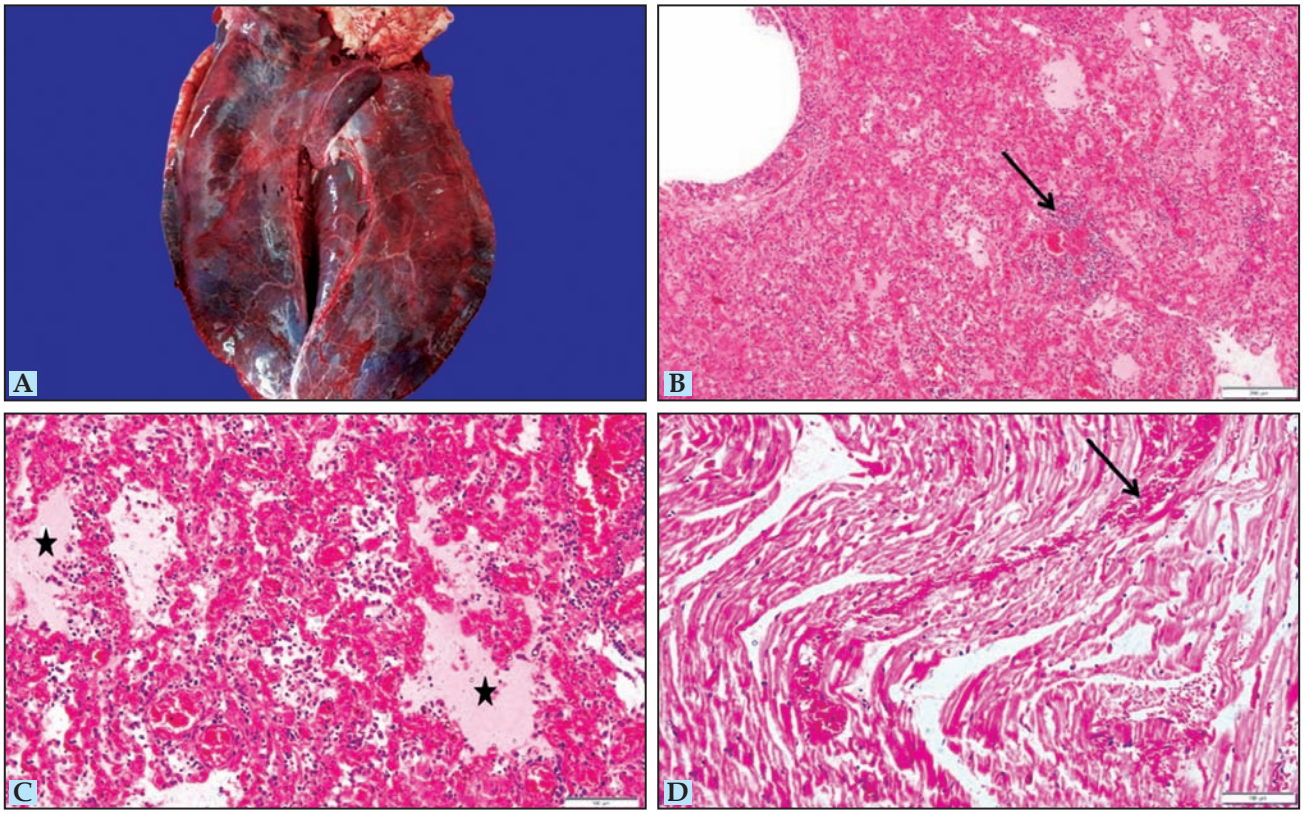

Fig 2. Images from camels injected with gliotoxin (0.025µg/kg b.wt.). **(A)** The whole lungs were congested and oedematous. **(B)** Loss of normal architecture of the pulmonary tissue as well as perivascular cuffing with lymphocytes (arrow). **(C)** Severe congestion of the interalveolar capillaries and homogenous eosinophilic material filing alveoli (asterisk). **(D)** Congestion of the capillaries of cardiac myofibres.

parameters of present study. The significant decrease in globulin supports the immune-depressant effect of gliotoxin in mice (Hussain *et al,* 2020). The AST and ALT are well-known biomarkers of hepatic damage (Recknagel *et al,* 1989). In this study, intravenous administration of gliotoxin (0.025µg/kg b.wt.) caused an increase in ALT and AST activities in camel liver at 1, 24, 48 and 72 hours post injection compared to its value before injection (0 hour). The values of these parameters at 24 hours post injection and onwards were significantly higher than that at 1-hour post injection. This finding was consistent with the findings of previous studies (Shathele, 2009, 2011) in camels intoxicated with gliotoxin (0.05µg/kg b.wt. and 0.1µg/kg b.wt., respectively). As indicated in Fig 1A, the intravenous administration of gliotoxin induced significant elevation in serum MDA levels at 1, 24, 48 and 72 hours post injection compared to its value before injection (0 hour). The MDA values from 24 hours post injection and onwards were significantly higher than its value at 1-hour post injection. These findings indicated that injected camels were subjected to a state of oxidative stress or lipid peroxidation due to gliotoxin injection. This state of oxidative stress reached its maximum 24 hours post injection. This finding was consistent with previous studies in the livers of rats (El-Bahr, 2015) intoxicated with AFB1. The superoxide radicals are converted to H_2O_2 by SOD. Furthermore, either CAT or GPX converts H_2O_2 to molecular oxygen and H_2O. Moreover, GPX can reduce lipid peroxides and other organic hydroperoxides that are highly cytotoxic products (Mujahid *et al,* 2005; Lu *et al,* 2010). Thus, SOD, CAT, GPX and GST constitute the principal components of the antioxidant system (Ahmad *et al,* 2012). In the present study, the activities of antioxidant enzymes such as total SOD (Fig 1B), CAT (Fig 1C), GPX (Fig 1D), and GST (Fig 1E) and GR (Fig 1F) in camels intoxicated with gliotoxin decreased significantly at 1, 24, 48 and 72 hours post injection compared to its value before injection (0 hour). The maximum decline of activities of these enzymes observed at 24, 48 and 72 hours post injection. The observed increase in lipid peroxidation was followed by decrease in the activities of enzymatic antioxidants (SOD, CAT, GPX, GST and GR). Thus, the significant increase in lipid peroxidation (MDA) could be due to a significant reduction in the activities of enzymatic

antioxidants (SOD, CAT, GPX, GST and GR). The same finding were demonstrated in liver tissue of rats intoxicated with AFB1-intoxicated (El-Bahr, 2015). Gross examination of died camel revealed that both lungs were diffusely congested, oedematous and oozing foamy exudate in cut section with presence of large focally extensive area of consolidation in the cranioventral part. The mucosa of the nasal passages and trachea were moderately congested. Small amount of clear straw yellow fluid was seen within the abdominal and thoracic cavities. Microscopically, the main histopathologic lesions were seen in the lungs. The inter-alveolar capillaries as well as peribronchiolar arterioles were greatly dilated and filled with blood. Approximately, 75% of the alveoli were filled with homogenous eosinophilic fluid and some foamy macrophages. Lymphocytes were noticed infiltrating the lung tissue and surrounding hyperemic capillaries and arterioles. Many desquamated and necrotic epithelial cells were seen within the bronchiolar and alveolar lumen. Some alveoli were lined with pneumocyte type II. The cardiac capillaries between myofibers were congested in association with multifocal area of hemorrhages. The observed lung histopathological changes were similar to that observed in lung of mice injected with gliotoxin (Hussain *et al,* 2020). In conclusion, the injected dose of gliotoxin was acutely toxic to camels as reflected on disturbed liver and kidney function, acceleration of oxidative stress and inhibition of antioxidants enzyme activities.

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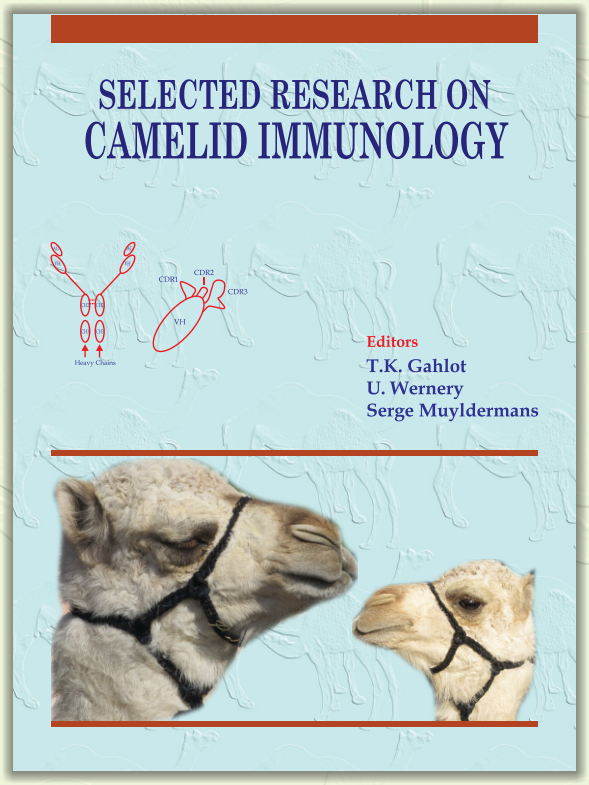
SELECTED RESEARCH ON CAMELID IMMUNOLOGY

(Hard Bound, 392 pages, few figs coloured, Edition 2016)

In 1989 a group of biologists led by Raymond Hamers at the Free University Brussels investigated the immune system of dromedaries. This discovery was published in Nature in 1993. Based on their structure, these peculiar camelid antibodies have been named Heavy Chain Antibodies (HCAb), as they are composed of heavy chains only and are devoid of light chains. Sera of camelids contain both conventional heterotetrameric antibodies and unique functional heavy (H)-chain antibodies (HCAbs). The smaller size and monomeric single domain nature make these antibodies easier to transform into bacterial cells for bulk production, making them ideal for research purposes. Camelid scientists world over were greatly fascinated by a new field of research called "Camelid Immunology". Significant research has been done on camelid immunology in recent decade. In order to benefit future camelid immunology researchers, this book was planned in the series of "Selected Topics" by Camel Publishing House with a title- "Selected Research on Camelid Immunology" edited by T.K. Gahlot, U. Wernery and Serge Muyldermans. This book is a unique compilation of research papers based on "Camelid Immunology" and published in Journal of Camel Practice and Research between 1994-2015. Research on this subject was done in 93 laboratories or institutions of 30 countries involving about 248 scientists. In terms of number of published papers in JCPR on the immunology the following countries remain in order of merit (in parenthesis), i.e. Iran (1), India and UAE (2), China and Saudi Arabia (3), Sudan (4), Kenya and Belgium (5), USA (6), Germany (7) and so on. The book contains 11 sections and is spread in 384 pages. The diverse sections are named as overview of camel immune system; determinates of innate immunity, cells, organs and tissues of immune system; antibodies; immunomodulation; histocompatibility; seroprevalence, diagnosis and immunity against bacteria, viruses, parasites and combination of other infections; application of camel immunoglobulins and applications of immune mechanisms in physiological processes. The camelid immunology has to go a long way in its future research, therefore, this reference book may prove quite useful for those interested in this subject. Book can be seen on www.camelsandcamelids.com.



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Schistosoma indicum, A RARELY REPORTED FINDING IN A DROMEDARY - A CASE REPORT

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ABSTRACT

A carcass of an emaciated female dromedary imported from Pakistan to the United Arab Emirates was sent for necropsy. The main pathological alterations were liver amyloidosis and thickening of the stomachal wall (compartment 3). Two pairs of schistosomes were discovered in the mesenteric veins of the small and large intestines. Morphological features matched those of *Schistosoma indicum*.

Key words: *Camelus dromedarius*, dromedary, Pakistan, *Schistosoma indicum*, United Arab Emirates

Members of the Family Schistosomatidae occur on all continents, except Antarctica. The range of species infecting mammals and human species, however, is restricted to tropical and subtropical latitudes. It was believed that *Schistosoma mansoni*, *S. haematobium* and *S. japonicum* infect humans and primates, and a wider species variety can be found in farm animals as well as in wild mammals. Recent molecular research showed that human and mammalian schistosome species may crossbreed. All schistosomes are biohelminthes with water snails as intermediate hosts, and transcutaneous infection occurs when final hosts have contact to cercariae of the parasite while wading or swimming in infested waters; final hosts can also be infected via drinking. While textbooks (Skrjabin, 1951; Eckert *et al,* 2005; Taylor *et al,* 2008) or review papers (Dakkak and Ouheli, 1987; Farhati *et al,* 1995; Erdenebileg, 2001; Parasani *et al,* 2008; Sazmand *et al,* 2019) mention camels as final hosts for schistosomes, original reports on camel schistosomosis are rare. We report here findings of *Schistosoma indicum* in a dromedary camel imported from Pakistan.

Case report

A recently imported adult female dromedary from Pakistan was sent for necropsy to the Central Veterinary Research Laboratory, in Dubai, in October 2020. The animal came to Dubai in March 2020 and displayed a poor general condition at necropsy with a history of chronic emaciation with progressive inappetence, recumbency and CNS signs. The sender suspected of camel paratuberculosis. The main pathological alterations of the 260 kg chronically emaciated carcass were seen in the liver and the stomach. The compartment 1 of the stomach showed severe calcification of the folds and massive thickened greyish, firm wall (2-4 cm thick layer). Histopathology of C3 showed massive pyogranulative inflammation with severe fibrosis, many necrosis with bacteria-colonies surrounded by eosinophilic Splendore-Hoeppli material. Histology of the swollen, firm liver revealed severe amyloidosis with 80% of the tissue occupied by amyloid-deposits as well as marked interlobular fibrosis.

In a histological section, a strange parasite like structure in a mesentery vein of the ileum consisting of several fragments with a diameter between 130 to 460 µm caught our interest. The largest fragment was 2080 µm long and consisted of an oval shaped smaller portion trapped in a bigger part (Fig 1). The outer surface was covered with tubercles and spines (Fig 2). A careful inspection of the remaining mesentery revealed a white worm like structure in a vein of the proximal colon (Fig 3). A closer look on the flushed out content revealed two morphologically different structures (Fig 4). The shorter (17 mm) and broader (0.43 mm) helminth with oral and ventral suckers at the anterior end and a long gynaecophoric canal commencing prior to the ventral sucker was a male. The subterminal oral and pedunculated ventral suckers measured 330 and 360 µm in diameter, respectively. There were eight testes posterior to the ventral sucker.

The longer (24.5 mm) and thinner (0.25 mm) with uterine eggs in the filiform anterior half and with a dark coloured posterior end turned out to be

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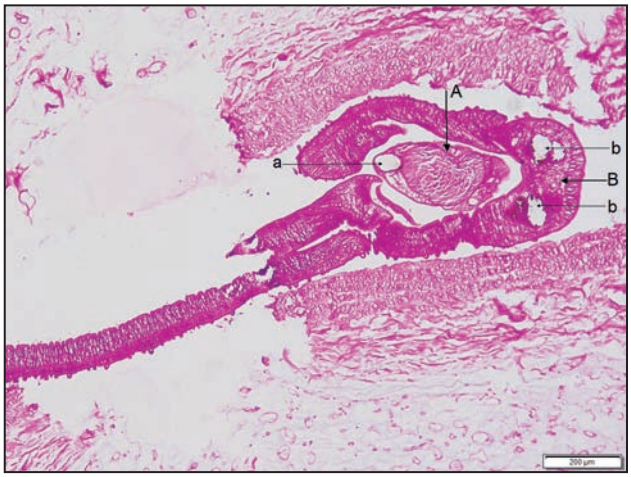

Fig 1. A pair of *S. indicum* in a histological section of a mesenteric vein. A: female in the gynaecophoric canal of the male, cut at the level of the ovary. B: male. a, b: intestinal branches.

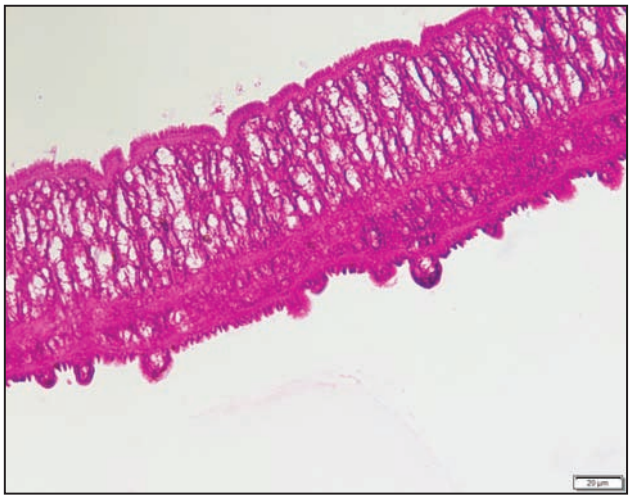

Fig 2. The tegument of the male is covered with tubercles and spines.

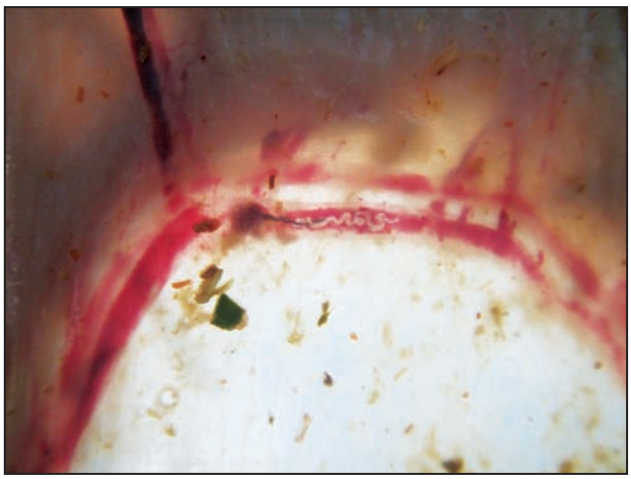

Fig 3. Schistosomes in the mesenteric vein of a camel.

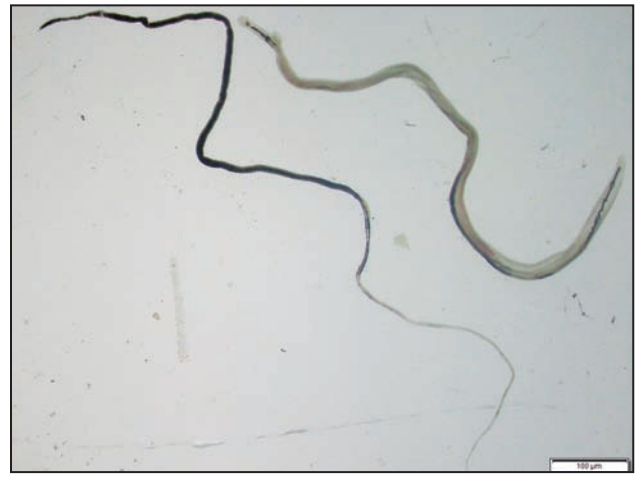

Fig 4. *S. indicum* isolated from the mesenteric vein. The shorter male can be recognised by the distinct presence of two suckers at the anterior end. The longer female has a filiform anterior end. The posterior end appears dark coloured due to the content of the intestine.

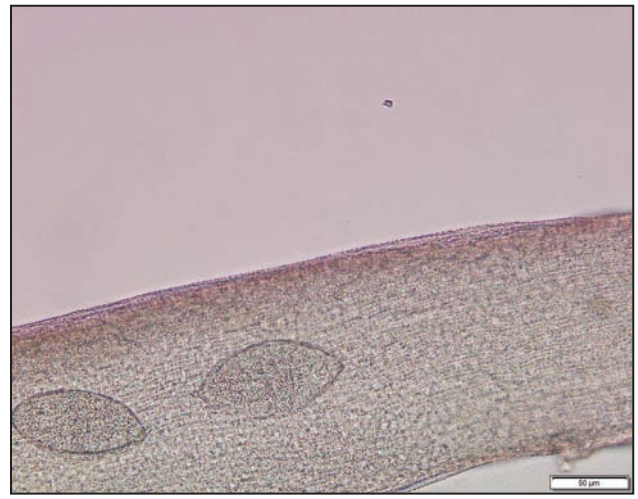

Fig 5. *S. indicum.* Uterine egg.

a female schistosome. The fusiform eggs (97x50 µm) had a spike on one end (Fig 5).

Considering the morphometrical characteristics of male and female worms and the origin of the animal we concluded dealing with *Schistosoma indicum*.

Discussion

S. indicum was first described by Montgomery (1906), in Uttarakhand, in North East India. It infects a wide range of farm animals, including buffalos, cattle, sheep, goats, horses and donkeys and the Ramshorn snail, *Indoplanorbis exustus,* serves as intermediate host. Predilection sites of *S. indicum* are venous blood vessels of the mesentery, pancreas and pelvis as well as the portal system of the liver.

Other mammalian blood fluke species occurring on the Indian subcontinent are *S. spindale*, *S. bomfordi*, *S. incognitum, S. nasale, S. nairi, Orientobilharzia dattai, O. turkestanicum* and *O. harinasutai* (Agrawal and Rao, 2011).

While schistosomosis is well investigated in India (Kali, 2015), little is known about species that occur in Pakistan. Human cases were reported from travelers returning from Africa (Khalid and Mahmood, 2001; Subhani *et al,* 2014) whose clinical signs and the shape of the parasitic eggs suggested *S. haematobium* as the pathogen. Anwar and Gill (1990) identified *S. indicum* and *S. bovis* in cattle and buffaloes. However, *S. bovis* is an African species and requires bulinid snails as intermediate hosts, which were not found in the investigations.

Niaz *et al* (2010) established the prevalence of Schistosoma infection in cows and in water buffaloes at different sites in Punjab, Pakistan, and found an overall prevalence of 15.0 and 15.1%, respectively, with the highest prevalence of 19% in cows and the lowest of 10.33% in buffaloes in Sialkot. In another study, in four districts of Punjab, Arshad *et al* (2011) established the extensity of *Schistosoma* in Buffaloes and found prevalences varying between 13.66 and 17.0%. Both studies were based on egg findings in coproscopical examinations and the Schistosoma species was not mentioned. Niaz *et al* (2013) collected more than 10,000 freshwater snails in Punjab and examined them for trematode development stages. Only *Indoplanorbis* species were found to excrete *Schistosome cercariae*. Our finding proves that *S. indicum* is one of the species that occur in Pakistan.

Montgomery (1906) mentioned small bead like nodules in the enlarged liver of a horse from which he described *S. indicum* and Datta (1933) reported alterations in the liver, lungs, small and large colon and kidneys caused by *S. indicum* in detail. A recent paper by Singh *et al* (2012) from Rajasthan (India) described hepatic schistosomosis based on egg findings in histological sections in two out of 137 examined dromedary livers.

In our case, a burden of only two pairs of *S. indicum* were found. Since there are no freshwater snails at the dairy farm in Dubai where the camel was kept, the infection must have happened in the place of origin of the animal, in Pakistan. The emaciation of the animal and liver alterations were attributed to amyloidosis of the liver.

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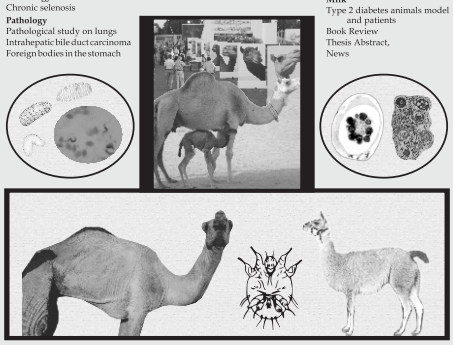

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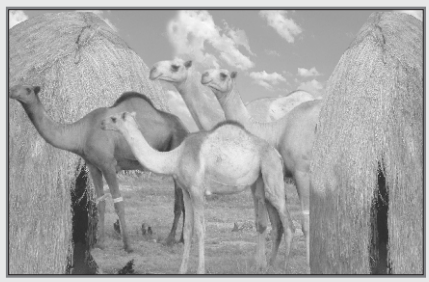

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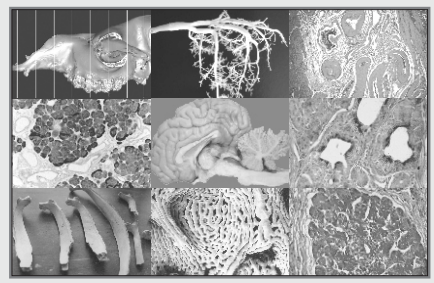

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SOCIO-ECONOMIC DIMENSIONS OF CAMEL BREEDING AND CAMEL WRESTLING IN TURKEY

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ABSTRACT

This study was conducted to examine the socio-economic dimensions of the camel breeding done for camel wrestling which has been on-going as a family tradition from the periods when nomadism was performed widely to present times. Data were obtained from the survey study conducted in 2020 with 56 camel breeders who were selected by random sampling. Most of the participants dealt with camel breeding because of the tradition/habit handled down from the family (78.6%) and 21.4% of them were involved in camel wrestling. Breeders fed their camels an average of 11 kg of roughage and 5 kg of concentrate feed daily and the average annual feed cost was $ 924. About 23.2% of the participants fed their camels with additional feeds 7 days before wrestling events. Those breeders having more camels incurred an extra average cost of $ 430 labour (savran) per month. Camel wrestling, led to soft tissue injuries in the face and in some body parts, leading the foot problems, and breeders had to spend average annual cost of $ 125 for veterinary care and medicine. The results of the survey were evaluated in terms of the health of camels and it was determined that there is a shortage of veterinarians specialised in camel breeding and in the treatment of common diseases and injuries in camels.

Key words: Breeding, camel, Turkey, wrestling

Camel is a multi-purpose animal and is bred for many purposes, such as milk, meat, wool production, cargo transportation, tourism, agricultural activities and races (Koyuncu and Yılmaz, 2019; Yılmaz *et al*, 2011). Camels are also bred for races in such countries as Arab Emirates, Tunisia, Sudan, Kenya, India and Egypt, for fights in Afghanistan and Pakistan, and for the camel wrestling in particularly the western regions of Turkey (Atasoy and Ozbaser, 2014). Although, the start date of camel wrestling in Anatolia is not precisely known, it is believed that it started in the Aegean Region in the 1800s (Atasoy and Ozbaser, 2014; Calıskan, 2013). Camel-wrestling has been organised and bear a cultural and touristic importance. Unlike other sports, it is performed in a festival together with music, folk dances and with special rituals according to traditions and customs. Thus camel wrestling in considered as a socio-cultural activity (Culha, 2008).

The aim of this study was therefore to determine the socio-economic status of the breeders raising camels for camel wrestling.

Materials and Methods

The data about camel breeding and the economic dimension of camel wrestling was collected from surveys with 57 camel breeders in camel wrestling events organised in the Aegean Region (in Aydın, Afyonkarahisar, İzmir, Denizli, Kutahya, Manisa, Mugla and Usak) between September-2019 and February-2020. SPSS 16 package program was used to analyse the obtained data. In the study, 'chi-square' and 't test' were also used to reveal the relationship between categorical variables.

Results and Discussion

The education level of the breeders was primary school (41.1%), middle school (12.5%), high school (35.7%), and university level (10.71%). In the study, 35.7% of the participants were determined to be the members of a union or cooperative and while 10% of them are members of a cooperative/ union not related with camel breeding, 35% of them were members of the Federation of Camel Breeding Culture and Camel Wrestling, and remaining were members of Camel Breeders' Associations

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(Table 1). There was no statistical relation between the education levels of the breeders and their membership to any organisation; however, the higher the education level, the more membership to any organisation (p>0.05) was seen.

Table 1. The relation of the education levels of the participant camel breeders to their membership to any cooperative/ union.

Education levels	Membership to any cooperative/union		Total
	Yes	No	
Primary school	6	17	23 (41.1%)
Middle school	2	5	7 (12.5%)
High school	9	11	20 (35.7%)
University	3	3	6 (10.7%)
Total	20 (35.7%)	36 (64.3%)	56 (10.71%)

X^2 = 2,369 p>0,05

It was revealed that out of 56 breeders only 48.2% (27) bred camels and the rest 51.8% bred cattle.

It was reveled that they had this occupation since 20.65 years as an average (min1, max 64) and that they breed 3.86 head camels (min 1, max 17); however, they only participated into wrestling with only 2.57 head of them (min 1, max 10) (Table 2). Breeders having a membership to a cooperative were younger than those not having a membership and that they breed approximately 1 camel more. It was found that 78.6% of the participants did camel breeding as a family tradition, while 21.4% were doing that for camel wrestling.

Approximately 45% of the breeders were of the opinion that camel breeding was not difficult. However, around 25% feel that camels are too hard to many when females are in their oestrus periods.

It was found that they gave an average of 11 kg of roughage per camel and 5 kg of concentrate (Table 3). Camel breeders give an average of 5.5 tons of feed per year to a single camel. It was seen that camel breeders generally prefer straw and crushed barley in the first place and then they prefer cereals mixed with alfalfa, vetch, barley or oat. About 23.2% of camel breeders also feed their camels before camel wrestling. It was determined that breeders give an average of 5 kg of additional feed (usually barley) to per animal daily, with an average of 7 days (min: 2, Max: 10) before wrestling. The camel breeders generally prefer straw and crushed barley in the first place. In a study, Koyuncu and Yilmaz (2019) reported that camels converted low-quality feeds into higher yields than other farm animals due to their digestive physiology, and thus being more economical.

As a result of the evaluations made with the t test in line with the data obtained, although there was no statistically significant difference between the income earned by the producers from wrestling and additional feeding before wrestling (p = 0.115 p> 0.05),

Table 2. Some information about the camel breeders.

Some information	Member of the cooperative	Mean	Std. Error	P Value
Experience time in camel breeding	Evet	16.50	2.401	P=0.09
	Hayır	22.03	2.659	p>0.05
The number of camels in farm	Evet	4.25	0.914	P=0.39
	Hayır	3.64	0.510	p>0.05
Number of wrestling camels in farm	Evet	2.95	0.394	P=0.89
	Hayır	2.36	0.319	p>0.05
Experience time in camel wrestling	Evet	15.75	2.585	P=0.99
	Hayır	19.61	2.109	p>0.05

Table 3. Preference of dry grass and concentrate feeds.

Dry grass	n	Order of preference	Concentrate feeds	n	Order of preference	Additional Feeding	n	Per cent
Hay	50	1	Barley meal	47	1	Barley	4	30.8
Clover	48	2	Mix cereal	15	2	Barley Vetch	3	23.1
Vetch	43	3	Wheat meal	14	3	Barley Vetch Oat	4	30.8
Barley Oat	35	4	Factory feed	8	4	Barley Vetch Wheat	2	15.3
Meadow grass	4	5	Diğer	7	5	Total	13	100

Table 4. Economic dimension of camel wrestling ($).

Income / Expense Items ($)*	Minimum	Maximum	Mean	Std. Deviation
Income from wrestling	75,0	1.800	400,3	335,9
The sale price of a wrestler camel	8.555	30.000	11.763,8	5.656,6
Annual feed cost	250,0	1.667,03	924,7	432,1
Monthly labour cost	250,0	833,3	430,0	241,2
Participation costs	83,3	83,3	83,3	0,00
Transport cost	17,0	150,0	40,0	21,5
Veterinary medicine cost	20,0	330,0	125,0	114,9

• 1 $=6,00 TL

the average income of those who feed the camel before wrestling with additional feed was $ 574. 94 ± 191.64, and those who did not feed with additional feed had an average income of $ 391.06 ± 62.62 (Table 4).

The breeders earned an average of $ 400 if their camels won wrestling. However, if the camel is a champion, the price of the camel increases and beomes important income.

The most important cost item in camel breeding is feed and this expense is determined to be an average of $ 924 per camel per year. The study revealed that those who fed 5 or more camels (17.85%) with hired workers, it incurs an average of $ 430 per month of labour (Savran) expenses. The participation, transportation and accommodation expenses of champion camels in particular were paid by the Wrestling Organising Committee (WOC). In the interviews, it was determined that 32.1% of those participating in camel wrestling were paid by the same committee in addition, 4 people were given an average of $ 1.167 (min: 850, max: 2,000) decoration costs.

That although, it varies according to the injuries, there is an average of $ 125 per year veterinary medicine and medication expenses (Table 5).

In the study, some injuries were seen in the camels of 28.6% of the participants. The most frequent injuries in wrestling are given in table 5.

Table 5. Types of injuries to wrestling.

Injuries	Frequency*	Per cent
Foot problems (nail dislocation, lameness etc.)	10	41.7
Injuries in the head area (lip splits, nosebleeds etc.)	8	33.3
Other soft tissue injuries	6	25.0
Total	24	100.0

* It is stated that the grower can make more than 1 choice.

According to (Turkish Statistical Institute) (TUİK, 2020) data, the presence of camels in Turkey, which was 865 heads in 2004, increased over the years and reached to 1.651 heads in 2019. In 2019, 29.68% of the camel presence in Turkey was in Aydın province, 16.05% of it was in Izmir, 14,72% in Canakkale, 10.36% in Antalya and 9.87% in Mugla. According to TUİK data, a great majority of the camels bred in totally 17 provinces in Turkey are bred so as to use them as cargo animals and in camel wrestling events and for tourism (Yılmaz *et al*, 2018).

With the increase in the number of camels, in the number of camel breeders and in camel wrestling, two federations were established in 2012 in Aydın and İzmir in Turkey. About 36% of the breeders were members of any cooperative or association and the participants were generally members of the Camel Breeders Association.

The participants have been participating in camel wrestling for 21 years on average and 78.6% were doing camel breeding as a tradition of family. Calıskan (2013) also found that a significant majority of wrestler camel owners (61.6%) did camel breeding as a family tradition.

In the study, it was determined that the participants carried out camel wrestling as socio-cultural activity, tradition, and advertisement-recognition. Similarly to the result of present study, Sanlı (2019) also reported the similar reasons for participation into the wrestling. Calıskan (2013) reported that the breeders participated in camel wrestling as family tradition and hobby purposes.

Approximately 40% of the breeders reported that camel care was not difficult and 25% found et difficult to manage especially during oestrus periods. However, the difficulties of the transportation process and the expenses to participate in a camel wrestling were also expressed. Delayed sexual maturity period, late in terms of milk production and the inability to determine oestrus period were also expressed as other difficulties by the breeders.

Almost all of the breeders were from the countryside and had financial constraints and left that their participation in camel wrestling was mandatory and incurred a significant cost. The maintenance-feeding of the camels, the worker (saver) fee, the transportation fee to participate into the wrestling, the equipment-accessory costs, and the accommodation during the wrestling involve additional cost (Yılmaz and Ertugrul, 2015).

In present study, it was opined that there were breeders participating into the wrestling to sustain their tradition in a socio-cultural activity and for hobby purposes even though their financial means were not sufficient to meet out the participation fees, the transportation and accommodation expenses. Only the 32.1% of them were paid by the associations and/or the municipalities organising the wrestling. Other breeders were found to have participation and transportation costs between $ 100- $ 200. The saddle dressing and decoration called "Camel's Packsaddle" consisting of colourful accessories made of sackcloth on the back of the camel, costs an average of $ 1.167 (min: 850, max: 2.000) and 4 participants of the survey felt that saddle dressing is also another expense item. Similarly, Culha (2008) reported that the money spent for these accessories varied between 1.000 TL and 10.000 TL.

Although, the transportation costs arising from the transport of camels in wrestling vary, it was determined in this study that they ranged between $ 17 and $ 150. In line with this study finding, Yılmaz *et al* (2018) reported that the transportation cost varies depending on the camel's reputation and the distance to be travelled. It was also determined that the transportation and participation fees of champion camels were paid by the Wrestling Organising Committee (WOC) (Yılmaz *et al,* 2018).

Camels are generally cared for by family members. However, some of the breeders (17.9%) having high number of camels pay some caregivers called as "savran" who are experts in the care and education of camels. Although, the amount varies depending on the number of camels, the pay is usually around $ 430 per month. In this regard, the findings of the study are in parallel with the findings of Calıskan (2009).

In the study, it was determined that the breeders earned an average of $ 400 income, depending on the size of the organisation, if their camels won wrestling. This income was insufficient in meeting out the annual expense of the breeders. However, it was determined that the price of the camel rises to $ 12.000 in average if the camel becomes the champion and provided a significant income for the breeder. As a matter of fact, although camel breeding and participation in wrestling were very expensive, the breeders attended these organisations especially, for the purpose of recognition and advertisement in the community. However, it was determined that many camel breeders could not participate in many organisations due to the insufficient financial situation. Therefore, if the wrestling organisers do not receive transportation and participation fees from these breeders, it can be said that the number of camels participating in camel wrestling will increase. In general, an average of 50 or 55 pairs of camels wrestle in camel wrestling festivals depending on the size of the organisations (Culha, 2008).

Camel wrestling is not only important organisations for camel breeders. On the contrary, it is intertwined with many subsidiary sectors such as camel feed, equipment, various accessories and ornaments, food and beverage and accommodation. Together with these elements, it should be seen as an important sector in the revival of the economy in the region and creating employment in the winter months (Yılmaz and Ertugrul, 2015). The wrestling events, organised particularly in November and March, are conducted by municipalities and the income provided is spent for some charities like school, mosque and bridge constructions. The awards are great in the organisations held in December and February and are organised by the Federation of Camel Breeding Culture and Camel Wrestling (DEGUF) and Camel Breeders' Associations. It was stated that state support was insufficient in camel wrestling. Calıskan (2009) reported that wrestling could hardly be financed in many regions and wrestling organisations could not perform wrestling events regularly every year due to financial problems. It was also stated in the same study that this problem seriously threatened the future of camel wrestling, so the state should support these organisations. Sanlı (2019), in a study stated that the nomadic lifestyle is appreciated in desert festival (desert camel races) held in the United Arab Emirates. He also added that the region's economy was contributed greatly with the participation of thousands of camels for high money awards.

Another problematic result that occurs in camel wrestling is the injuries that occurred during the wrestling of animals. Yılmaz and Ertugrul (2015)

reported in their studies that foot problems occurred and camels limped, especially when camels older than 20 years old are wrestled. In the wrestling events organised in Turkey, injuries of the camels are reduced by putting on camel saddle (havut) and net muzzle on mouth. Injuries were generally related with soft tissue injuries. The camel breeders have to spend $ 125 towards veterinary-drug costs. In line with the findings of Yilmaz and Ertugrul (2015), it was concluded that there is a shortage of specialised veterinarians in camel breeding, disease prevention and treatment.

Camel wrestling held in western Anatolia should be considered as an important opportunity in terms of job opportunities and local economy and rural development for the people in the region.

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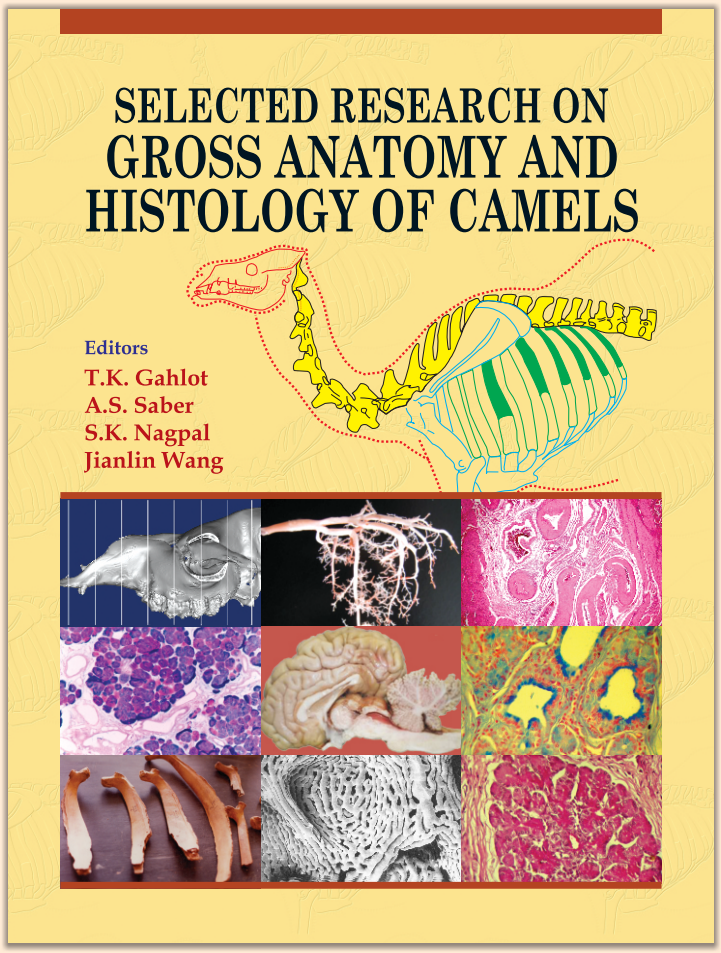

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EFFECT OF GENERAL ANAESTHESIA WITH HALOTHANE OR ISOFLURANE ON SERUM CONCENTRATIONS OF INFLAMMATION AND BONE BIOMARKERS IN DROMEDARY CAMELS

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ABSTRACT

This study was carried out to determine the effect of halothane or isoflurane general anaesthesia on the serum concentrations of the inflammation biomarkers C-reactive protein (CRP) and haptoglobin (Hp), and on the bone biomarkers osteocalcin (OC), bone-specific alkaline phosphatase (b-ALP) and pyridinoline cross-links (PYD) biomarkers. Six healthy female camels were premedicated with xylazine and anaesthesia was induced with ketamine and maintained with either isoflurane (isoflurane group, *n*=6) or halothane (halothane group, *n*=6). A washout period of 2 weeks was allowed between the two anaesthetic protocols. Three blood samples were obtained from each camel; immediately before anaesthesia (T0), 80 min of recovery (T1), and 48 h after recovery from anaesthesia (T2). In the halothane group, the CRP values decreased significantly 48 h after anaesthesia compared to preanaesthetic values (P=0.04). In the isoflurane group, the serum concentration of CRP has increased significantly 80 min after recovery compared to preanaesthetic values (P=0.0009), but decreased significantly 48 h after anaesthesia compared to 80 min of recovery (P=0.0005). The most important finding in the halothane and isoflurane groups in the current study was the sharp increase in the serum concentration of Hp where it dramatically increased from 0.2±0.04 mg/L preanaesthetic (T0) in both groups to 43.4 mg/L and 20.8±4.6 mg/L (T1), respectively. The bone formation (OC, b-ALP) and bone resorption (PYD) biomarkers serum levels in this investigation did not show any significant changes following either halothane or isoflurane general anaesthesia compared to preanaesthetic values at any test point. In conclusion, isoflurane is superior to halothane as an inhalation anaesthetic in dromedary camels as acute phase reaction occurred sharply in halothane compared to isoflurane anesthetised camels.

Key words: Anaesthesia, bone biomarkers, camels, halothane, inflammation biomarkers, isoflurane

The inflammation biomarkers or acute phase proteins (APPs) are group proteins whose concentrations decrease or increase in sera in animals subjected to external or internal challenges (Eckersall and Bell, 2010). The acute-phase response (APR) is a rapid, nonspecific, systemic response occurring secondary to many types of tissue injury and might be a physiological protective mechanism during inflammatory events (Tharwat, 2020a).

The common biomarkers of bone formation include osteocalcin (OC), bone-specific alkaline phosphatase (b-ALP) and amino and carboxy propeptides of collagen type I. The most common biomarkers of bone resorption include pyridinoline cross-links (PYD), deoxypyridinoline enzyme tartrate resistant acid phosphatase and amino and carboxy telopeptides of collagen type I (Tharwat and Al-Sobayil, 2020).

The use of inhalation anaesthetics such as halothane, isoflurane, and sevoflurane in large animals is increasing, especially for prolonged procedures (Ahmed *et al,* 2015; Al-Sobayil *et al,* 2016). The aim of the present study was to determine the effect of halothane or isoflurane general anaesthesia after xylazine and ketamine administration on the serum concentrations of the inflammation biomarkers CRP and Hp, and bone biomarkers OC, b-ALP and PYD.

Materials and Methods

Camels, anaesthesia and blood sampling

Six adult female dromedary camels (*Camelus dromedarius*) weighing 321-503 kg and aged 5-12 y

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were used. The experiment was divided into two parts, two weeks apart, each using six camels. The camels were premedicated with xylazine HCl (0.2 mg/kg, IV, Bomazine 10%, BOMAC Laboratories Ltd., New Zealand). Anaesthesia was induced 20 min later with Ketamine (2 mg/kg, IV, Ketamine 10%, Alfasan, Woerden, Holland) and was maintained with either isoflurane (Floran, HIKMA Pharmaceuticals, Amman, Jordan) or halothane (Anestane®, HIKMA Pharmaceuticals, Amman, Jordan) using 100% oxygen at a flow rate of 6 L/min. The animals were first subjected to isoflurane and then to halothane anaesthesia, with a washout period of 2 weeks between the two anaesthetic protocols. The anesthetised camel was moved onto a padded operating table, positioned in right lateral recumbency and was connected *via* the endotracheal tube with a semiclosed-circle rebreathing anaesthetic machine (SurgiVet Foal Circuit Set, Smith Medical North America, Waukesha, WI, USA). Anaesthesia was discontinued after 1 h and the camels received supplemental oxygen (6 L/min) through the endotracheal tube. After tracheal extubation, oxygen was insufflated through a nasal tube until sternal recumbency was achieved. Three blood samples were obtained from each camel for serum analyses of inflammation and bone metabolism biomarkers. The first blood sample was collected immediately before anaesthesia (T0), the second (T1) was collected 80 min of recovery, and the third was collected 48 h after recovery from anaesthesia (T2).

Assays of inflammation and bone metabolism biomarkers

The serum concentration of CRP was determined using a commercially available human kit (Minineph Hunan CRP kit, Binding Site Group Ltd.), according to the manufacturer's instructions. The determination of soluble antigen concentration by nephelometric method involves a reaction with the antibody bound to a latex particle to form insoluble complexes. When light is passed through the suspension, a portion of the light is scattered by a photodiode. The amount of light scattered is directly proportional to the CRP concentration in the serum samples. The analytical sensitivity of the assay was 0.44 mg/L, and intra- and inter-assay CVs were 3.6-6.6% and 3.6-6.0%, respectively. The serum concentration of Hp was determined in the sera using a colorimetric assay (Haptoglobin kit, second generation, Tridelta Ltd., Ireland) as reported recently (Tharwat and Al-Sobayil, 2015a; Tharwat and Al-Sobayil, 2015b; Tharwat and Al-Sobayil, 2018a; Tharwat, 2020a; Tharwat, 2020b). The analytical sensitivity of the assay was 0.0005 mg/mL, and intra- and inter-assay CVs were 5-6% and 4-6%, respectively.

The serum concentrations of the bone metabolism biomarkers OC, b-ALP and PYD were determined using commercial human immunoassay kits (Metra Biosystems Inc., a division of Quidel Corp.) as reported recently (Tharwat and Al-Sobayil, 2015b; Tharwat and Al-Sobayil, 2018a; Tharwat and Al-Sobayil, 2018b; Tharwat and Al-Sobayil, 2020). The limit of quantification of OC ranged from 2 to 32 ng/mL, and precision CVs within and between runs were 5-10%. The dynamic range of BAP was 2-140 U/L, and precision CVs within and between runs were 4-6% and 5-8%, respectively. The dynamic range of PYD was 15-750 nmol/L, and precision CVs within and between runs were 6-10% and 3-11%, respectively.

Statistical method

Data are presented as means ± SD, and were analysed statistically using the SPSS statistical package (SPSS, Statistical Package for Social Sciences, SPSS Inc., Chicago, IL, USA, Copyright© for Windows, version 18.0, 2009). A repeated measures analysis of variance was employed as the statistical model to evaluate the differences over time in the isoflurane and halothane anesthetised camels. The Duncan test was used to calculate multiple comparisons. Student t test was used to evaluate differences between isoflurane and halothane anesthetised animals. Results were considered significant at $P<0.05$.

Results and Discussion

The serum concentrations of CRP in 130 and isoflurane anaesthetic camels are illustrated in Fig 1A. In the halothane group, although the serum concentration of CRP decreased 80 min after recovery (T1) (1.7±0.2 mg/L compared to 2.7±1.8 mg/L preanaesthesia); this decrease was insignificant (P=0.1). Forty eight hours after anaesthesia (T2), the CRP values (1.2±0.2 mg/L) decreased significantly compared to preanaesthetic values (T0) (P=0.04) and compared to 80 min of recovery (P=0.0002). In the isoflurane group, the serum concentration of CRP was increased significantly 80 min after recovery (5.1±2.0 mg/L) compared to preanaesthetic values (1.8±0.4 mg/L) (P=0.0009). At 48 h after anaesthesia, the CRP value (1.6±0.3 mg/L) were decreased significantly compared to 80 min of recovery (P=0.0005), but not significantly when compared to preanaesthetic values (P=0.2).

The serum concentrations of Hp in halothane or isoflurane anaesthetic camels are illustrated in Fig 1B. In the halothane group, the serum concentration of Hp has increased sharply 80 min after recovery (T1) (43.4 mg/L vs 0.2±0.04 mg/L preanaesthetic; p=0.0005). Forty eight hours after anaesthesia (T2), the Hp value decreased to 14.3±2.7 mg/L; a significant difference when compared to preanaesthetic value (T0) (P=0.0001) and when compared to 80 min of recovery (P=0.01). In a similar pattern, in the isoflurane group, the serum concentration of Hp has increased dramatically 80 min after recovery (20.8±4.6 mg/L vs 0.2±0.05 mg/L preanaesthetic; p=0.0001). Forty eight hours after anaesthesia, the Hp value decreased to 3.7±2.6 mg/L; a significant difference when compared to preanaesthetic value (P=0.002) and when compared to 80 min of recovery (P=0.0001).

Fig 2A illustrates the serum concentrations of OC in halothane or isoflurane anaesthetic camels. In the halothane group, the serum OC values were 26.6±18.4 ng/mL preanaesthesia (T0), 23.1±5.1 ng/mL 80 min of recovery (T1) and 28.3±20.1 ng/mL 48 h after anaesthesia (T2) with no significant difference among the 3 values (P>0.05). In the isoflurane group, the serum OC values were 25.3±9.2 ng/mL preanaesthesia, 24.2±20.5 ng/mL 80 min of recovery and 26.8±23.8 ng/mL 48 h after anaesthesia with no significant difference among the 3 values (P>0.05).

The serum concentrations of b-ALP in halothane or isoflurane anaesthetic camels are illustrated in Fig 2B. In the halothane group, the serum OC values were 14.8±4.0 U/L preanaesthesia (T0), 16.1±5.4 U/L 80 min of recovery (T1) and 16.9±6.71 U/L 48 h after anaesthesia (T2) with no significant difference among the 3 values (P>0.05). In the isoflurane group, the serum OC values were 12.3±3.3 U/L preanaesthesia, 12.0±4.7 U/L 80 min of recovery and 13.0±6.2 U/L 48 h after anaesthesia with no significant difference among the 3 values (P>0.05).

Fig 2C illustrates the serum concentrations of PYD in halothane or isoflurane anaesthetic camels. In the halothane group, the serum OC values were 7.6±1.7 nmol/L preanaesthesia (T0), 5.8±0.4 nmol/L 80 min of recovery (T1) and 8.9±3.8 nmol/L 48 h after anaesthesia (T2) with no significant difference among the 3 values (P>0.05). In the isoflurane group, the serum OC values were 8.9±4.5 nmol/L preanaesthesia, 8.0±2.8 nmol/L 80 min of recovery and 6.8±3.5 nmol/L 48 h after anaesthesia with no significant difference among the 3 values (P>0.05).

To the authors' knowledge, this is the first study in dromedary camels evaluating the effect of general anaesthesia by either isoflurane or halothane on the serum concentrations of inflammation (CRP and Hp) and bone metabolism (OC, b-ALP and PYD) biomarkers.

The APR is a rapid, nonspecific, systemic response occurring secondary to many types of tissue injury and might be a physiological protective mechanism (Yazwinski *et al*, 2013). This response is induced by the pro-inflammatory cytokines IL-1, TNF-α and especially IL-6. These cytokines activate receptors on various target cells and promote

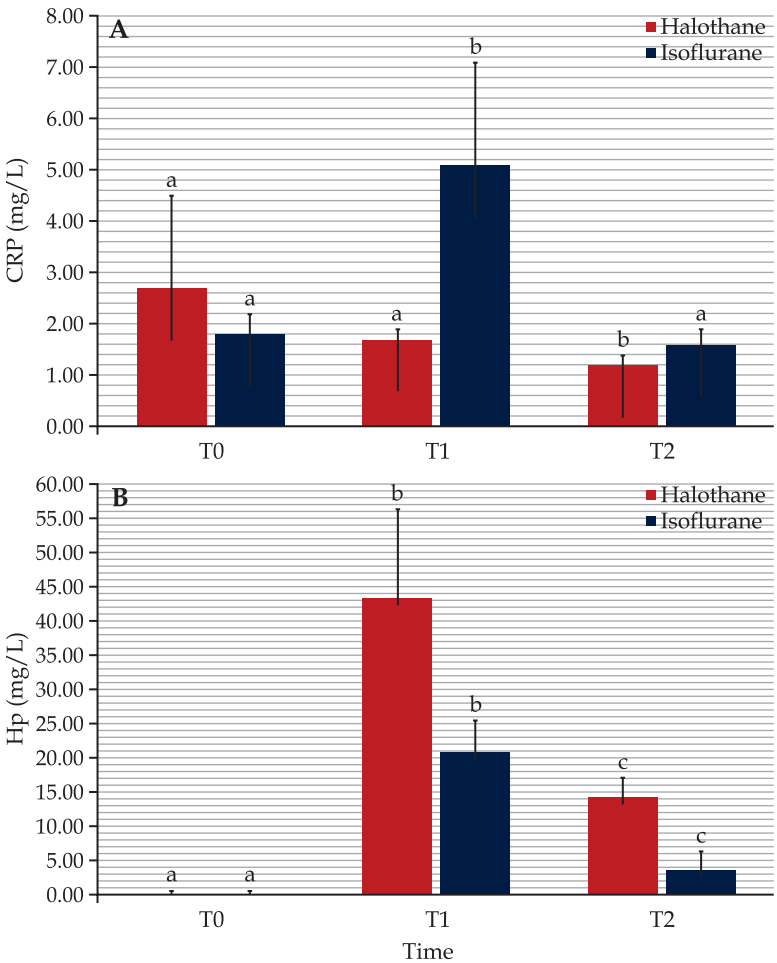

Fig 1. Serum concentrations of acute phase protein C-reactive protein (CRP) (**A**) and haptoglobin (Hp) (**B**) in camels (n = 6) undergoing isoflurane and halothane anaesthesia. T0, immediately before anaesthesia; T1, 80 min of recovery; T2, 48 h after anaesthesia. Different letters differ significantly at P<0.05.

hormonal and metabolic changes leading to local and systemic effects, including APP synthesis in the liver (Petersen *et al,* 2004; Tizard, 2009). The APPs can be used in diagnosis, prognosis and in monitoring response to therapy, as well as in general health screening and animal welfare (Eckersall, 2000; Eckersall and Bell, 2010). The APPs have received attention as biomarkers for APR due to its low physiological levels, a fast incline, marked rise in concentration during APR that eases detection and a fast decline after cessation of a stimulus (Murata *et al,* 2004; Murata, 2007; Tharwat, 2020a).

In the present study, the serum concentration of the acute phase protein CRP decreased insignificantly in the halothane group 80 min after recovery from anaesthesia (T1) and significantly 48 h after anaesthesia (T2). On the contrary, the serum concentration of CRP in the isoflurane group increased significantly 80 min after recovery compared to preanaesthetic values (T0). However, 48 h later, the CRP value in the same group decreased significantly compared to 80 min of recovery. In human medicine, CRP was used as a marker of the surgical stress reduction within an enhanced recovery after surgery protocol. It was found that, the presence of complications post operation was independently associated with an increase in CRP values (Olivares *et al,* 2018). Furthermore, the CRP blood level in humans was estimated to depend on the method of anaesthesia. It was found that the smallest increase in plasma CRP was found in patients operated on under regional anaesthesia compared with those operated on under general anaesthesia (Kolomachenko, 2018). In dogs with anaesthetic protocols using sevoflurane or a combination of fentanyl, midazolam, and sevoflurane, a significant difference in serum CRP concentrations was not detected post-operatively between groups at any time point. However, serum CRP concentrations in the same study were significantly increased post-anaesthetic induction in both groups. These significant increases were attributed to surgical trauma (Saunders *et al,* 2009). Following ovariohysterectomy in female dogs due to naturally occurring pyometra, the serum concentrations of CRP in two different anaesthesia and analgesic protocols

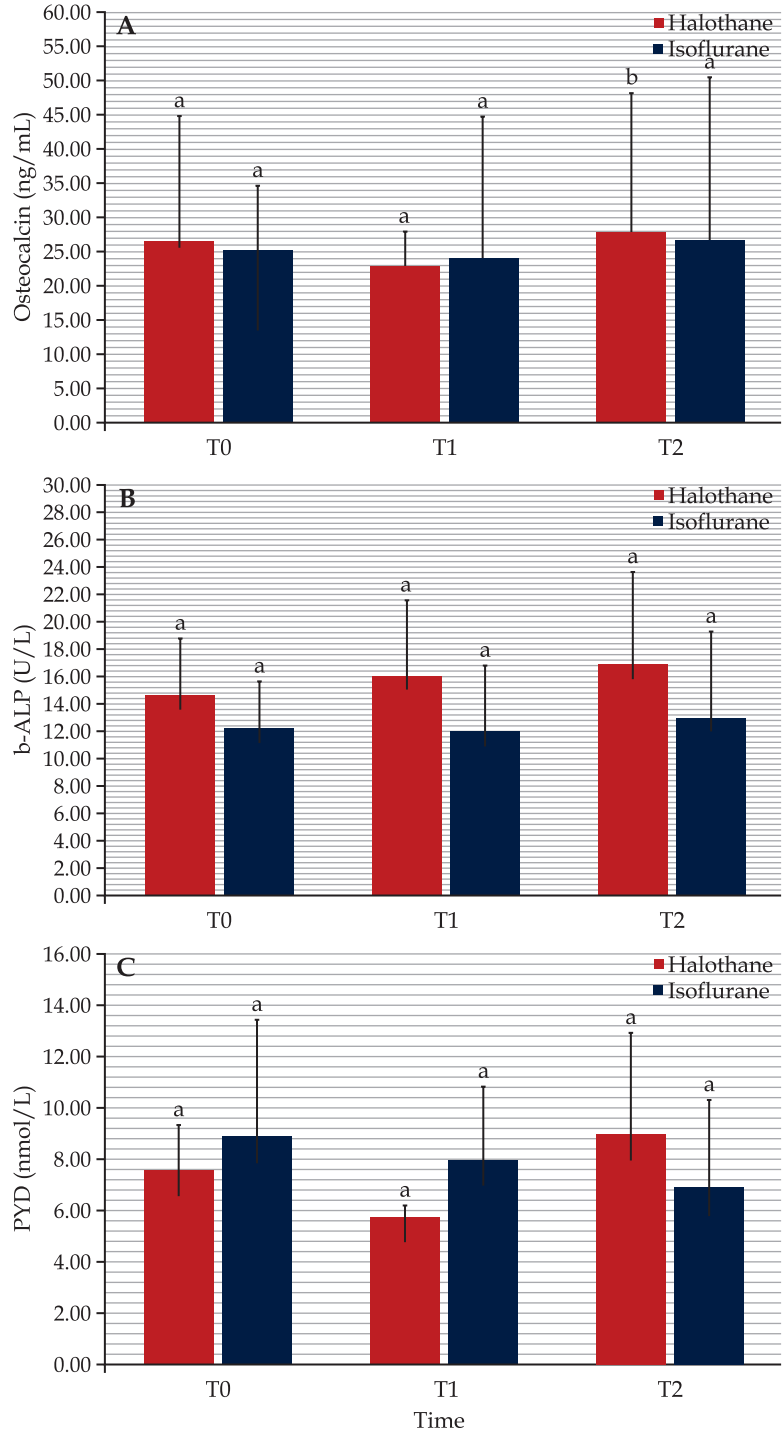

Fig 2. Serum concentrations of bone biomarkers osteocalcin (A), bone-specific alkaline phosphatase (b-ALP) (B) and pyridinoline cross-links (PYD) (C), in camels (n = 6) undergoing isoflurane and halothane anaesthesia. T0, immediately before anaesthesia; T1, 80 min of recovery; T2, 48 h after anaesthesia. Same letters did not differ significantly at P>0.05.

with and without low-dose ketamine were essentially the same before surgery, but significantly increased in the control group and decreased in ketamine group at 48 hours after surgery. It was concluded from that study that low-dose ketamine attenuated the postoperative concentration of serum CRP in dogs with pyometra compared with dogs that did not receive ketamine in the perioperative period (Liao *et al,* 2014).

The most important finding in the halothane and isoflurane groups in the present study was the sharp increase in the serum concentration of the acute phase protein Hp where it dramatically increased from 0.2±0.04 mg/L preanaesthetic (T0) in both groups to 43.4 mg/L and 20.8±4.6 mg/L (T1), respectively. Further, the Hp values in both groups declined at 48 h after anaesthesia (T2), but still significant when compared to preanaesthetic values. Postoperative changes of serum concentrations of Hp were studied in women undergoing elective hysterectomy during either general anaesthesia or epidural analgesia. Results showed that Hp was reduced during the first postoperative day, followed by a gradual, significant increase of haptoglobin to 140% above preoperative levels on day 7 after surgery (Rem *et al,* 1980).

The OC, a product of the osteoblasts, is regarded as a sensitive indicator of bone formation (Pullig *et al,* 2000). In addition to its increase that accompany skeletal growth, weight-bearing exercise induces changes in serum concentrations of OC (Eliakim *et al,* 1997). The b-ALP, a glycoprotein found on the surface of osteoblasts, has also been shown to be a sensitive and reliable indicator of bone metabolism. Although, OC and b-ALP are considered bone formation biomarkers, their correlation in the serum of camels was reported to be weak (Al-Sobayil, 2010). The lack of a strong correlation between the two biomarkers has been attributed to the fact that each of them reflects different stages of osteoblast function (Delmas *et al,* 1990). The PYD cross-links, indicators of type I collagen resorption, are found in the mature collagen of bone. Increased concentrations of PYD in the blood or urine are most commonly considered as indicators of bone resorption (Thompson *et al,* 1992; Tharwat and Al-Sobayil, 2020).

The bone formation (OC, b-ALP) and bone resorption (PYD) biomarkers serum levels in this investigation did not showed any significant changes following either halothane or isoflurane general anaesthesia compared to preanaesthetic values at any test point. In response to isoflurane-induced anaesthesia in young female guinea pigs the bone metabolism biomarkers OC, but not deoxypyridinoline, increased (Tabatabaei *et al,* 2015). In another study conducted in cynomolgus monkeys, the anaesthetic isoflurane decreases ionised calcium and increased the OC secondary to the decrease in ionised calcium (Hotchkiss *et al,* 1998). In a study carried out in 36 female patients undergoing elective total hip replacement, OC as well as b-ALP concentrations decreased significantly until 72 h post-surgery. Increased cortisol secretion and other hormonal and inflammatory components of the peri-operative stress response may play a role in mediating this response (Nicholson *et al,* 2002). In another study conducted in humans scheduled for general anaesthesia due to hip fracture, serum concentrations of the bone formation biomarkers OC and b-ALP did not change significantly until 12 weeks postoperative (Biricik *et al,* 2019). It was concluded from the results of this study that isoflurane is superior to halothane as an inhalation anaesthetic in dromedary camels. Acute phase reaction occurred sharply in halothane anesthetised camels as indicated by a remarkable increase in the serum concentration of Hp (two fold increase when compared with isoflurane anesthetised camels). The bone formation and resorption biomarkers did not change significantly by either halothane or isoflurane general anaesthesia.

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TRANSCRIPTOME ALTERATIONS IN HIGH GLUCOSE-INDUCED RENAL TUBULAR CELLS OF BACTRIAN CAMEL

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ABSTRACT

The transcriptome alterations in renal tubular cells of Bactrian camel treated with high glucose were investigated. Primary tubular epithelial cells, obtained from kidney of Bactrian camel were treated with low glucose (LG) or high glucose (HG). The transcriptome gene expression in primary tubular epithelial cells of Bactrian camel was examined using RNA sequencing technology. 1322 genes from HG and LG were significantly altered. The up-regulated genes in the HG group include CAMK2B, CRYAA and VNN1. Several pathways associated with diabetic nephropathy including PI3K-Akt signaling pathway, AGE-RAGE signaling pathway, ECM-receptor interaction and PPAR signaling pathway were activated by HG stimulation. This study provides scientific basis for understanding the mechanism of renal tolerance to hyperglycemia of Bactrian camel.

Key words: Bactrian camel; high glucose; transcriptome; tubular epithelial cells

The Bactrian camel lives in the Gobi Desert or semi-desert regions of the northwest China. Bactrian camel has evolved many distinct abilities to adapt to such environments (Chen *et al,* 2009). The levels of blood glucose in camels are much higher than in sheep and ponies (Elmahdi *et al,* 1997). However, Bactrian camel do not develop diabetes and hypertension (Wu *et al,* 2014). It indicates that Bactrian camel has a unique hyperglycemia tolerance mechanism. High-glucose concentrations trigger various metabolic and cellular dysfunctions, which in the kidney affect many types of cells including mesangial cells and renal tubular cells (Vallon and Komers, 2011). The transcriptome alterations in high glucose-induced mesangial cell model which represents diabetic nephropathy (DN) *in vitro* have been investigated (Li *et al,* 2019). However, there is no reported study on the transcriptome changes in high glucose (HG)-induced renal cells of Bactrian camel. This paper will report our work on transcriptome alterations in HG-induced renal tubular cells of Bactrian camel using RNA-seq approach. This study provides scientific basis for understanding the mechanism of renal tolerance to hyperglycemia of Bactrian camel.

Materials and Methods

All the procedures involving animals were approved by the Institutional Animal Care and Use Committee of the Inner Mongolia Agricultural University (12150000460029509N). Primary tubular epithelial cells (PTECs) were isolated from the kidney cortex segment of healthy Bactrian camel. The cortical fragments were incubated for 30 min at 37°C in a buffer containing 0.1% collagenase type I. The preparation was washed and filtered through a cell strainer. The obtained cells were treated with medium containing 1g/L D-glucose (low glucose, LG) and 10% foetal bovine serum (FBS). Cells were incubated in 5% CO_2 atmosphere at 37°C and grown to confluence. In certain plates, the medium was changed. Cells were treated with medium containing 4.5 g/L D-glucose (HG) and 10% foetal bovine serum (FBS).

After 48 h exposure to HG, Total RNA was extracted from PTECs from LG and HG groups

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using TRIzol reagent. The quality and quantity of the extracted RNA samples were determined with the NanoPhotometer spectrophotometer and the Bioanalyser 2100 system. Pooled RNA from samples was used for library preparation. The mRNA sequencing libraries were constructed by the IlluminaTruSeq RNA preparation kit. The samples were sequenced on the Illumina Hiseq 2500 platform (Novogene, Beijing, China) with125 bp/150 bp paired-end reads. Data quality was checked using the fastq software. The reads were compared with Bactrian camel genome and treated by Hisat2. Differential gene expression analysis was performed by the DESeq2 R package. For statistical analysis, two comparisons (HG vs. LG) were analysed by *p* value and false discovery rate (*q* value). The statistical enrichment of differentially expressed genes (DEGs) in KEGG pathways was tested by clusterProfiler R package. KEGG pathway with corrected p-value and q value less than 0.05 were significantly enriched by DEGs.

Results

To measure the gene expression profiles exposure to HG, we performed RNA-Seq of renal tubular cells of Bactrian camel treated with LG (control) or HG. The genes whose expression differed in the two groups were identified and filtered for corrected p values < 0.05 and $|\log_2$ fold change$|$ ≥0.3. A total of 1, 322 DEGs were identified, of which 758 genes were up-regulated and 564 genes were down-regulated in the HG group. Top 10 genes with the highest $\log_2$ fold change in the HG group were listed in Table 1 and these genes included calcium/calmodulin-dependent protein kinase II beta (CAMK2B), crystallin alpha A (CRYAA), vanin 1 (VNN1), NLR family 2C pyrin domain containing 13 (NLRP13) and bone morphogenetic protein 8a (BMP8A). Analysis of DEGs through KEGG showed that 758 up-regulated genes in the HG group could be significantly enriched in 24 pathways including PI3K-Akt signaling pathway, Cytokine-cytokine receptor interaction, AGE-RAGE signaling pathway in diabetic complications, ECM-receptor interaction and PPAR signaling pathway (Table 2).

Discussion

HG can induce cell injury, formation of Reactive oxygen species (ROS) has been considered as one of the principal mechanisms of glucose-induced cell toxicity (Vallon and Komers, 2011). Our results showed that HG can change some gene expression level in renal tubular cells of Bactrian camel. CaMKII was reported to as a pathological mediator of ER stress, oxidative stress and mitochondrial dysfunction in a murine model of nephronophthisis. Experiments *in vitro* and *in vivo* demonstrated that CaMKII inhibition relieved endoplasmic reticulum stress and oxidative damage and improved mitochondrial integrity and membrane potential (Bracken *et al,* 2016). The elevated expression of CAMK2B gene indicated that the renal tubular cells of Bactrian camel were damaged by HG stimulation. Vanin-1 (VNN1), another elevated gene which is associated with oxidative stress, was found in the HG group. Studies have showed that urinary vanin-1 could be a useful biomarker for the detection of drug-induced acute tubular necrosis focusing on oxidative stress (Hosohata, 2016). Recent studies have shown that crystallins play an instrumental role in diabetes and its complications. Crystallins specifically α-crystallins had been damonstrated to possess antioxidant, antiinflammatory, antiapoptotic and antiaggregation (chaperone) functions. Crystallins could be exploited as therapeutic targets for diabetic complications (Reddy and Reddy, 2016). The elevated expression of CRYAA in renal tubular cells of Bactrian camel may be one of the reasons why its kidney can tolerate HG.

In the PI3K-Akt signaling pathway, fibroblast growth factor 1 (FGF1), fibroblast growth factor receptor 1 (FGFR1), phosphatidylinositol 3-kinase (PI3K) and protein kinase B (Akt) were upregulated

Table 1. Top 10 upregulated genes in the HG group.

Gene symbol	Gene name	Log_2 fold change	*p* value
CAMK2B	calcium/calmodulin-dependent protein kinase II beta	4.568	0.027
CRYAA	crystallin alpha A	4.382	0.019
LOC105079214	putative serine protease 47	4.023	0.009
VNN1	vanin 1	3.599	0.031
NLRP13	NLR family pyrin domain containing 13	2.888	0.048
BMP8A	bone morphogenetic protein 8a	2.854	0.037
INHBC	inhibin beta C	2.546	0.020
LOC105075982	E3 ubiquitin-protein ligase Midline-1-like	2.514	0.018
PCDH11X	protocadherin 11 X-linked transcript variant X2	2.441	0.007
SPRY3	sprouty homolog 3	2.427	0.036

Table 2. Significantly enriched KEGG pathways of upregulated genes in the HG group.

Pathways	*p* value	*q* value
PI3K-Akt signaling pathway	2E-07	4E-05
Human cytomegalovirus infection	4E-04	0.031
Proteoglycans in cancer	9E-04	0.044
Cytokine-cytokine receptor interaction	3E-03	0.044
Focal adhesion	2E-03	0.044
cAMP signaling pathway	3E-03	0.044
Hippo signaling pathway	4E-03	0.049
Adrenergic signaling in cardiomyocytes	4E-03	0.049
Small cell lung cancer	3E-04	0.031
AGE-RAGE signaling pathway in diabetic complications	5E-04	0.031
Thyroid hormone signaling pathway	3E-03	0.044
FoxO signaling pathway	3E-03	0.044
Prostate cancer	2E-03	0.044
Parathyroid hormone synthesis, secretion and action	2E-03	0.044
Cholinergic synapse	3E-03	0.044
ECM-receptor interaction	2E-03	0.044
Longevity regulating pathway	2E-03	0.044
Pancreatic cancer	2E-03	0.044
Insulin secretion	2E-03	0.044
Non-small cell lung cancer	2E-03	0.044
PPAR signaling pathway	3E-03	0.044
Sphingolipid metabolism	1E-03	0.044
Cholesterol metabolism	4E-03	0.044
Glycosaminoglycan biosynthesis	3E-03	0.044

in the HG group. FGF1 has been proved to ameliorate chronic kidney disease via PI3K/AKT mediated suppression of oxidative stress and inflammation (Wang *et al*, 2019). In this study, the elevated expression of FGF1 may contribute to the upregulation of PI3K-Akt signaling pathway. Upon glucose entry into renal cells, there were a number of intracellular events that occurred in the presence of high-glucose ambience. The advanced glycation endproducts (AGEs) were formed. The intracellular AGEs could modify the protein functions and activate PKC, MAPK and transcription factor-like NF-κB and thus modulating the expression of various growth factors, cytokines and consequentially the ECM proteins (Kanwar *et al*, 2005). We identified some pathways that involved in the cellular pathobiology of diabetic kidney disease in the HG group, such as AGE-RAGE signaling pathway in diabetic complications, Cytokine-cytokine receptor interaction and ECM-receptor interaction. These results indicated that HG could cause injury to the renal cells of Bactrian camel. Experimental evidence suggested that PPARα activation attenuates or inhibited diabetic microvascular damage, including lipotoxicity, inflammation, reactive oxygen species generation, endothelial dysfunction and thus might influence the development and pathogenesis of diabetic microvascular complications (Hiukka *et al*, 2010). PPARα may become an important therapeutic target for treating diabetic renal complications (Chung and Park, 2011). Some up-regulated genes in the HG group were enriched in the PPAR signaling pathway, such as peroxisome proliferator-activated receptor α (PPARα) and retinoic acid receptor (RXR) (another nuclear receptor that can heterodimerise with PPARs). These results indicated that elevated expression of PPARα in renal cells of Bactrian camel induced by HG may have protective effects on the cells. This study provided scientific basis for understanding the mechanism of renal tolerance to hyperglycemia of Bactrian camel.

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Trypanosoma evansi ABORTION IN A DROMEDARY CAMEL HERD IN THE UAE - PART I

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ABSTRACT

A dromedary camel breeding herd consisting of 16 female and 2 male camels experienced an abortion storm over a period of 2 years. Fifteen (94%) of the 16 female camels aborted during the last stage of pregnancy. *Trypanosoma evansi* was directly identified by the buffy coat technique in 7 (39%) of the camels and in 15 (83%) with the indirect in-house antibody ELISA developed at the Central Veterinary Research Laboratory (CVRL). One bull and two newly introduced female camels were negative in both tests.

The whole herd was treated with 20ml of Cymelarsan. Blood samples were retested 6 weeks after treatment. All 18 dromedary camels did not clear the parasite from the blood and four dromedaries were still positive in the buffy coat technique and the level of antibodies had further increased. In Part II of our investigations we will report if another *Trypanosoma*l drug had cured the herd.

Key words: Antibody ELISA, buffy coat technique, camel abortion, *Trypanosoma evansi*

All trypanosomes infecting camels belong to the Salivaria group and are transmitted by bites of blood sucking flies. Surra caused by *Trypanosoma* (*T.*) *evansi* is the most important parasitic disease in dromedaries. The literature of surra in Bactrian camels is scarce. A good overview of trypanosomiasis in camels has been compiled by Wernery *et al* (2014) and OIE (2018). In the OIE chapter 3.1.21 multiple diagnostic techniques are explained in detail including agent identification, animal inoculation, polymerase chain reaction (PCR) and serological tests. At CVRL in the UAE, surra is diagnosed serologically with an indirect in-house antibody ELISA (i-ELISA) and with the buffy coat technique (BCT) (OIE, 2018). In camels, the *T. evansi* prevalence rate in the UAE reached 25% in 2019 (CVRL Annual Report, 2019). The increase of trypanosomiasis cases in camels is most probably due to advanced landscaping and imported dromedary camels. Surra has been diagnosed in dromedaries in many Middle Eastern countries (Wernery *et al,* 2014), but literature of abortions is rare. We report here an abortion storm in a camel herd in the UAE caused by *T. evansi*.

Materials and Methods

Dromedary camels

A breeding herd of 18 dromedary camels was affected which consisted of 16 females and 2 males. The herd roamed 40 km north-east of Dubai in a desert area near a village with ponds, agriculture greeneries and farms. Fifteen of the 16 female dromedary camels aborted during the breeding season 2018-19 of which 4 became again pregnant during the breeding season 2019-20.

Test methods

Jugular venipuncture was performed to obtain blood samples from all 18 dromedaries. The EDTA blood was tested for the presence of the parasite with the BCT and the sera were examined for *T. evansi* antibodies with a CVRL indirect ELISA (i-ELISA). Both methods are described by the OIE (2018). The results are shown in Table 1. The blood was also tested for haematological parameters and iron levels with analysers from Sysmex XN for haematology and Cobas C311 for iron. The values were then compared between the 15 infected dromedary camels and 3 non-infected camels as a mean and the significant difference calculated before and after treatment. The results are shown in Table 2.

Treatment

All camels including the 3 negative ones were treated with 20 ml of Cymelarsan (Melarsomine) intramuscularly (im) divided into 2 injections 10 ml each. This is the recommended dosage for a 400 kg camel.

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Table 1. Results of *T. evansi* infection diagnosed with BCT and CVRL i-ELISA.

S.No.	Camel ID	Gender	Before treatment		6 weeks after treatment with Cymelarsan	
			BCT	i-ELISAxx	BCT	i-ELISAxx
1	Shaheen Ajeeb	F	-	2.0	-	2.4
2	Shaheen Abbady	F Pregnant	++*	1.1	-	2.2
3	Shaheen Al Muhairi	F	-	2.3	-	2.5
4	Bint Mukhles	F	+	2.2	-	2.4
5	Al Khawara	F	-	-	-	-
6	Shaheen Al Arti	F	-	2.1	-	2.5
7	Shaheen Hamroor	F Pregnant	-	2.0	++++	1.2
8	Bint Dhabyan	F Pregnant	-	1.7	-	2.3
9	Sudaniyah	F Pregnant	++	2.3	-	2.6
10	Bint Al Qaher	F	+	2.2	++++	2.4
11	Al Naseem	F	-	2.0	++++	2.3
12	Ghourob	F	+	1.5	-	2.2
13	Bin Mashaal	F	-	2.2	-	2.5
14	Faraha	F	+	1.6	-	1.7
15	Shaheen Weld Al Nuaimiyeh	F	-	0.4	-	-
16	Al Hathrah	F	-	-	-	-
17	Nassi	M	++++	0.78	++	1.1
18	Moshawesh	M	-	-	-	-

* = number of parasites seen in BCT + = 1 or 2 in a slide ++ = very few in a slide +++ = 1 in view field
++++ = more than 1 in view field xx = Optical Density (OD) - = negative

Results

Table 1 shows the results of *T. evansi* infection before and after 6 weeks of treatment. Table 2 shows the haematological and iron values compared between 2 groups, the infected and non-infected dromedaries as a mean before and after treatment. The results show that one bull of the herd and two pregnant female camels which were recently introduced into the herd remained negative for *T. evansi*. However, 15 camels (94%) aborted during the last stage of pregnancy during the breeding season 2018-2019 (Fig 1). *T. evansi* was directly identified by the buffy coat technique (BCT) in 7 (39%) camels and in 15 (83%) of them serologically. One bull and two newly introduced pregnant females were negative in both tests. Six weeks after the Cymelarsan treatment, *T. evansi* was still detected in the blood of 4 camels by BCT and the i-ELISA antibody levels had further increased (Table 1). The comparison of haematological and iron values of the 2 groups, which is shown in Table 2 revealed significant differences in most of the values. Red blood cell count (RBC), haemoglobin (HB), packed cell volume (PCV), platelets (PLT) and iron values were significantly decreased in comparison to the non-infected camels, but remained within the reference values; outside the reference values were only White Blood Cell (WBC) and the lymphocyte counts.

Table 2. Comparison of mean blood parameters and iron values of 15 dromedaries with no *T. evansi* in their blood and 3 infected camels.

Parameters	SI units	Reference values*	Before treatment Mean Results 4 weeks later	
			3 Non-infected	15 infected
Haematology				
RBC	10^{12}/L	7.00 – 10.50	10.1	8.3
HB	g/dl	10.5 – 14.5	13.9	11.1
PCV	L/L	0.23 – 0.30	0.30	0.25
PLT	10^{9}/L	270 – 600	424	381
IRON	µmol/l	15 - 27	22	16
WBC	10^{9}/L	8.0 – 15.0	13.9	23.6
NEU	%	40 – 60	43.8	42.5
LYM	%	25 - 45	36.2	50.3
MONO	%	3.0 – 6.0	3.8	3.6
EOS	%	0.0 – 8.0	4.2	2.5

* Wernery *et al* (1999)

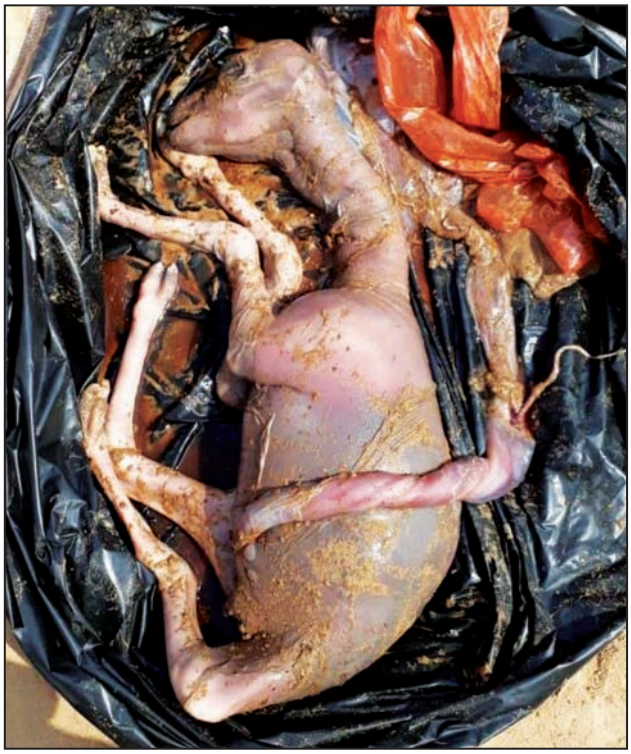

Fig 1. Aborted approximately 7 month-old foetus caused by *T. evansi*.

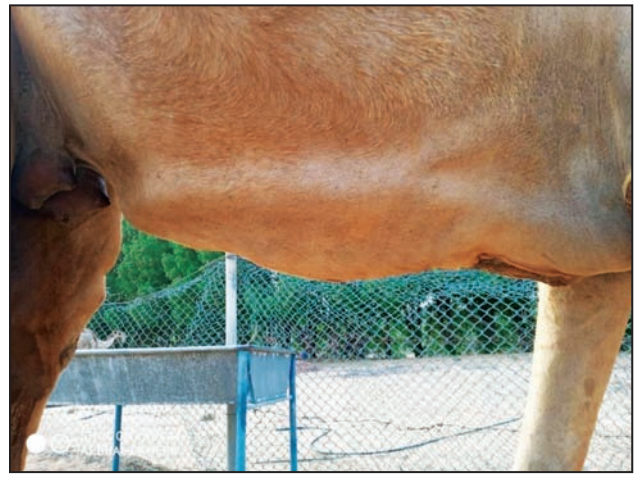

Fig 2. Oedema under the belly of a dromedary, pregnant with a 7 month-old foetus, caused by *T. evansi*.

Fig 3. Two horse Tabanid flies under the belly of a dromedary camel.

Discussion

Surra is an arthropod borne parasitic disease of camels and horses, but also all domestic animals are susceptible. Several species of haematophagus flies like Tabanids and Stomoxys transfer the parasite from host to host, acting as mechanical vectors. The disease can be fatal, particularly in camels, horses and dogs, but in other animal species it appears to be non-pathogenic and these species serve as reservoirs for the parasite. Also a few wild animals are susceptible to infection and may also serve as reservoirs. Surra occurs in North Africa, the Middle East, Asia, the Far East and Central and South America (Wernery *et al*, 2014). The distribution of *T. evansi* in Africa extends into the Tsetse belt areas, where differentiation from *T. brucei* is difficult. Surra is transmitted by biting flies, probably resulting from interrupted feeding. The parasite does not undergo a development in the flies. A large number of horse fly species act as mechanical vectors for *T. evansi* (Wernery *et al*, 2014). Ticks, mosquitos and Culicoides do not play a role in the transmission of the parasite. The clinical signs of surra in dromedaries are multifaceted and may vary widely depending on the infection phase. Therefore, it is important to differentiate between acute and chronic cases. The acute form occurs mostly in horses and camels (Van den Bossche *et al*, 2009). In a classical surra case, the disease, if not treated, develops into a chronic course with weight loss, weakness, loss of condition, rough coat and oedema at different locations but mainly under the belly (Fig 2). Two of the infected camels developed oedema at the ventral part of the abdomen and hind limbs. This clinical sign develops also during the acute stage. The animal develops anaemia, mucous membranes are pale and changes in the haematological parameters are often significant with low red blood cell count (RBC), low haemoglobin (HB), low packed cell volume (PCV) and decreased platelets. Anaemia is often a reliable indicator of a chronic trypanosome infection, but it is not pathognomonic as mild subclinical and acute infections often show no evidence of anaemia.

Although there was a significant difference in haematological and iron levels of the 15 infected dromedaries in comparison to the 3 non-infected camels, most of the values were still in the normal reference range as shown in Table 2. This shows

that it is nearly impossible to diagnose a *T. evansi* infection on haematological grounds. However, the WBC and the lymphocyte counts were significantly increased and lay outside the reference values. It is worthwhile to mention that an elevation of eosinophil count is common in a parasitic infection. However, the eosinophil count was not elevated in the *T. evansi* infected dromedaries.

One of the most important features of surra is abortion and it occurs in all stages of pregnancy (Gutierrez *et al*, 2005). So far it is not known why abortions occur and this would be an interesting research field in future to examine the aborted foetus and afterbirth material thoroughly for any alteration.

In 2018/2019 an abortion storm caused by *T. evansi* infection occurred in a dromedary camel herd consisting of 16 female and 2 male dromedaries. The parasite was directly identified by the buffy coat technique (BCT) in 7 (39%) of the camels and by the indirect ELISA in (83%) of them. It is a well-known fact that *T. evansi* is intermittently excreted into the circulating system of a camel and therefore an infection can be easily missed when only parasite detection techniques are applied for the diagnosis of surra. Recurring episodes of parasitaemia occur regularly during the course of the disease. Identification of the agent and a serological test was therefore used in this camel herd. The indirect antibody ELISA identified more than 50% of the infected dromedaries compared to BCT: 7 with BCT, 15 with i-ELISA. According to the OIE (2018) several tools should be used to diagnose a *T. evansi* infection which include serological tests as well as tests for the identification of the parasite. At CVRL, the BCT and i-ELISA are regularly used in parallel as they give the most reliable results for the diagnosis of surra.

Using the recommended doses by the producing company, Cymelarsan had no effect on the eradication of the *T. evansi* infection of this dromedary herd. Blood test after the treatment showed that the antibody levels had further increased as shown by the OD values and that in 4 camels the parasite was still detectable. In a Part II we will report about the outcome of the infection when another *Trypanosomal* drug is used.

Conclusion

Typanosoma evansi abortion in the dromedary camel is common and the diagnosis of surra cannot be made on haematological grounds. Recurring episodes of parasitaemia occur and therefore only direct and indirect tests can identify all infected camels. At CVRL, the buffy coat technique (BCT) and an indirect antibody ELISA are routinely used to diagnose surra in dromedary camels.

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VITAMIN B1, B2, B6 AND B12 LEVELS IN SERUM AND CEREBROSPINAL FLUID OF DROMEDARY CAMELS (*Camelus dromedarius*) AFFECTED WITH NEUROLOGICAL SIGNS

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ABSTRACT

This study was designed to evaluate and correlate CSF chemical analysis specially Vitamin B1, B2, B6 and B12 in the diagnosis of neurological manifestations in 13 camels (aging 2-19 years). The studied vitamins in affected animals were compared to healthy. Additionally, the current results of vitamin concentrations of Vitamins B1, B2, B6 and B12 in cerebrospinal fluid and serum samples were determined using HPLC assay. The results revealed that the concentration of those vitamins in the CSF was significantly less than in serum.

Key words: Biochemical, camel, CSF, UPLC–MS/MS, neurological, serum

The vitamin status of the camel is not well known, it needs more studies and the number of references is low, which little work has been done on vitamins in camel specially B vitamins (Faye and Bengoumi, 2018). Usually, B vitamins are synthesised in ruminants by ruminal flora (Mohamed, 2006). There were several methods for the determination of vitamins B including the spectrophotometric method of determination of vitamin B1 in a pharmaceutical formulation was simple, accrued, and rapid but depends on the colorimetric reaction mechanism (Shaopu Liu *et al,* 2002), method of high-performance liquid chromatography (HPLC) (Moreno and Salvado, 2000) and method of liquid chromatography/mass spectrometry (LC-MS/MS) which is an efficient, accurate and stable in the analysis of vitamins, it has a unique advantage of sensitivity, specificity and also the ability to be applied to antibiotics, drugs or metabolite analysis in any biological samples (Geng *et al,* 2017; Roelofsen *et al,* 2017; Nicholas *et al,* 2016).

The determination of normal chemical values of cerebrospinal fluid in different animal species has been documented including cattle, sheep, horses, dogs, cats, and some laboratory animals (Roberta and Simon, 2009; Achaaban *et al,* 2009; Naziji and Maleki, 1998; Arneri and Mousavian, 2007; Stocker *et al*, 2002 and Welles *et al,* 1992). Generally, there is a lack of information on normal CSF constituents and their normal values in camels. Recently Shawaf *et al* (2018) reported some values of CSF constituents from healthy camels in Saudi Arabia. Cerebrospinal fluid may provide a wide range of valuable biochemical and cellular information in the diagnosis of neurological disorders, that helps in the evaluation of the nervous system health of animals (Al-Sagair *et al,* 2005; Frosini *et al,* 2000; Welles *et al,* 1992).

Dubduba syndrome in an emerging viral neurological disease of camels (Al-Dubaib *et al,* 2008). Al-Swailem *et al* (2010) diagnosed cerebral listeriosis in a she camel with neurological signs, i.e. lock of coordination, Parkinson's like tremors of head and lower lip paralysis. Babelhadj *et al* (2018) detected prion disease in camels of Algeria. Authors detected pathgnomonic neurodegeneration and disease specific prion protein (PrP^{sc}) in brain tissues of camels. It is also known as camel spongiform encephalopathy or mad camel disease.

Wernery *et al* (2004) described lock jaw and stiff gait as predominant neurological sign in case of tetanus in a camel.

Neurological disorders present a significant sanitary and economic risk to the animal production industry worldwide (Lecollinet *et al,* 2019). Vitamin B1 (Abbas *et al,* 2008), B6 (Ahmad *et al,* 2013), and B12 (Nijst *et al,* 1990) are closely associated with

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neurological functions. However, Vitamin B1 deficiency has been defined in camels as responsible for polioencephalomalacia (PEM), which is called cerebrocortical necrosis and irritating severe nervous signs (Faye and Bengoumi, 2018). Mohamed (2006) reported data for vitamin B12 in the plasma of a healthy camel. It is important to recognise the relation between levels of vitamin B and neurological disorders in camel because there are indications that some of the diseased camels with neurological disorders may improve after receiving doses of vitamin B (Bhandare, 2009). This study was designed to evaluate the levels of vitamins B1, B2, B6 and B12 in serum and CSF of affected camels by using a unique and accurate technique (ultra-performance liquid chromatography coupled with mass spectrometry) UPLC/MSMS system to help in the diagnosis of neurological manifestations in camels.

Materials and Methods

2.1 Ethical Approval

All experimental procedures and management conditions used in this study regarding blood and CSF sampling in camels were approved by the Ethics Committee at King Faisal University, Saudi Arabia (Permission number KFU-REC/2020-10-06).

2.2 Animals and sampling

Thirteen dromedary camels (aging 2-19 years) with a history of neurological signs were presented to the Veterinary Teaching Hospital, King Faisal University were investigated in this study. The main neurological signs shown by these animals were shivering, tremor, staggering, rotation of the head, slight vision impairment, and progressive worsening general condition. It excluded other diseases through haematological and chemical analysis. Five healthy camels were used for comparison. The animals were examined clinically, and then blood samples were obtained from the jugular vein for serum preparation. After sedation and aseptically preparation of the caudal part of the neck, CFS samples were taken from the Atlanto-occipital articulation according to Shawaf *et al* (2018).

2.3 Vitamins determination

2.3.1 Materials and Reagents

Vitamins B1, B2, B6, and B12 were purchased from ACROS ORGANICS (New Jersey, USA, 1-800-ACROS-01, Geel, Belgium). Pyridoxine hydrochloride (B6, 99%; extra pure, HPLC assay, lot A0310405, code: 150770500). Thiamine hydrochloride (B1, 98.5-101%; extra pure HPLC assay, lot: A0309985, code: 148990100. Riboflavin (B2, 98% extra pure, HPLC assay, lot: A0305365, code: 132350250). Cyanocobalamin (B12, 96% extra pure, HPLC assay, lot: A0304024, code: 405920010). Ethyl acetate, water, ammonium acetate, caffeine, and methanol HPLC grade were purchased from Sigma Aldrich (USA).

2.3.2 Sample Preparation

Serum samples 250 µL, 25 µL (0.5 mM) NaOH and 25 µL Caffeine (Internal standard, analytical grade) (1µg mL^{-1}) were vortexed for a half min, then 2 mL of ethyl acetate solvent (EA) was added and vortexed again for 2 min, followed by refrigerated centrifuge at 4000 rpm for 6 min at 4°C. The supernatant (upper organic layer) was separated and then transferred into another ppt (polypropylene tube), it evaporated under a gentle flow of nitrogen gas to dryness at 37°C. The residue of supernatant was reconstituted with a 0.1 ml mobile phase, and 10µL was injected for analysis according to Geng *et al* (2017).

2.3.3 The procedure of UPLC–MS/MS Conditions

Ultra-performance liquid chromatography coupled with mass spectrometry (UPLC–MS/MS) analysis was done according to Geng *et al* (2017). An ultra-performance liquid chromatography (UPLCTM) system Acquity (Waters, Milford, MA, USA) was interfaced with a triple quadruple mass spectrometer (UPLC/MSMS) (TQDTM, Waters Micro mass, Manchester, UK) using an electrospray interface. Vitamins B1, B2, B6, and B12 and IS in serum were separated, using an Acquity UPLC, C18 analytical column (150 × 4.6 mm, particle size 3 µm, 100A) (Waters). The eluted with the mobile phase of methanol: ammonium acetate 5 mM (60:40, v/v) with a flow rate of 0.5 mL min^{-1} and the temperature of the oven was 30°C. Multiple reaction monitoring (MRM) mode was used for the detection of vitamin B1, B2, B6, B12, and internal standard (IS), with ion transition of m/z 265 → 122.1, 377 → 244, 170 → 152, 678.6 → 359.0, 195.1 → 137.8, respectively. Calibration curves were prepared for serum sample extracts (after reconstitution in mobile phase) were spiked with different aliquots of Vitamins B standard solution to give final concentrations. The calibration samples consisted of six different levels of vitamin B1 (1, 5, 10, 20, 50, 100 ng mL^{-1}), B2 (5, 10, 20, 50,100, 200ng mL^{-1}), B6 (1, 5, 10, 20, 40, 80 ng mL^{-1}) and B12 (5, 10, 20, 30, 40, 50 ng mL^{-1}). Quality control samples of LOD (limit of detection), for vitamin B1 were 1, 2, 50 and 160 ng mL^{-1}, for B2: 5, 10, 20, 35 ng mL^{-1},

for B6: 1, 2, 20, 75 ng mL^{-1}, and for B12: 5, 10, 20, 40, 50 ng mL^{-1}, respectively.The amount of vitamin was calculated as the calibration line y = ax + b. Quality control and quantification purposes were according to Geng *et al* (2017).

2.4 Statistical analysis

Data were recorded in Excel spreadsheets and imported into Stata version 14 (Stata Corp., TX, USA) for further analyses. Descriptive statistics (mean, SEM) were calculated for each parameter. Variation within each parameter was evaluated using the coefficient of variation (CV). The effects were considered significant at $P < 0.05$.

Results

Figures 1, 2, 3, 4 and Table 1 showed serum and CSF levels of Thiamine (B1), Riboflavin (B2), Pyridoxine (B6), and Cobalamin (B12) in healthy and camels with neurological signs. Thiamine (B1) levels showed a significant decrease in the serum of affected camels than healthy ones as well as CSF of affected and healthy. Riboflavin (B2) levels showed a significant decrease in the serum of affected camels than healthy ones but there was no significant difference in CSF of affected and healthy animals; on the other hand, there was a highly significant difference in the serum compared to CSF in healthy animals but with the same significant difference in affected animals. Pyridoxine (B6) levels showed a significant decrease in the serum of affected camels than healthy ones as well as CSF of affected and healthy, Cobalamin (B12) levels showed a significant decrease in the serum of affected camels than healthy ones as well as CSF of affected and healthy, on the other hand, there was a highly significant difference in the serum compared to CSF in healthy animals.

Discussion

Using LC-MS/MS which has higher sensitivity and specificity, is an accurate alternative. However, Electrospray ionisation (ESI) is an effective means of ionising the B vitamins, but it is sensitive to matrix effects which change the relative response between samples and standards due to changes in ionisation efficiency (Geng *et al,* 2017; Roelofsen *et al,* 2017; Nicholas *et al,* 2016). Vitamins are vital for health status, and their lack could affect the animals (Faye and Bengoumi, 2018). Vitamin B1 or thiamine is a coenzyme, which is a needed phase in the synthesis of fatty acids, nucleic acids, aromatic amino acids and steroids, and the precursors to neurotransmitters and bioactive compounds vital for neurological tract function (Kerns *et al,* 2015). Thiamine deficiency has been defined in camels as responsible for polioencephalomalacia (PEM), which is called cerebrocortical necrosis, irritating severe nervous signs Faye and Bengoumi (2018). However, Kiupel *et al* (2005) and Oliveira *et al* (1996) reported that sulfur toxicity is a cause for thiamine deficiency in lama and ruminants, respectively. Mohamed (2006) and Wernery *et al* (2009) reported more levels for thiamine in the serum of healthy animals than in the present study. However, Abbas *et al* (2008) stated that the concentration of thiamin in the serum of a healthy camel calf was less than that reported in the present study. The different results of thiamine in the serum of healthy camels in the present study as compared to previous studies could be attributed to training and feeding type, which affecting microflora by produce thiamin. The results of decreased thiamine in the serum of affected camels in the present study are in agreement with most previous studies (Brent and Bartley, 1984; Faye and Bengoumi, 2018; Kiupel *et al,* 2005; Milad and Ridha, 2009; Nema *et al,* 2014; Rachid *et al,* 2011; Ramos *et al,* 2005). In agreement with our results, Wernery *et al* (2009) reported similar results for thiamine in the serum of healthy camels. However, Abbas *et al* (2008) reported lower results for thiamine (21±10 µg/l) in the serum of affected camels with neurological signs than those reported in the present study. To our best knowledge, there is a lack of information on the concentration of thiamine in cerebrospinal fluid in animals. The decreased thiamine in CSF of

Table 1. The mean concentrations of vitamin B1, B2, B6 and B12 in serum and CSF samples healthy and affected obtained LC–MS/MS methods (mean ± SEM).

	Serum Healthy		Serum Affected		CSF Healthy		CSF Affected	
	Mean ± SEM	Range	Mean ± SEM	Range	Mean ± SEM	Range	Mean ± SEM	Range
B1 (mg/L)	53.56 ± 2.64	40.45-70.56	36.49 ± 1.23	30.37-41.41	48.53 ± 2.92	40.14-70.03	36.9 ± 1.05	30.5-45.13
B2 (mg/L)	350.57 ± 55.05	50.96-650.05	137.01 ± 6.02	100.09-165	55.71 ± 17.64	3.01-330.12	27.61 ± 5.88	3.76-140
B6 (mg/L)	12.63 ± 2.1	5.2-32.2	5.76 ± 0.34	4.9-7.8	4.68 ± 0.28	2.8-6.6	3.28 ± 0.2	2.2-4.9
B12 (ng/100ml)	44.8 ± 1.77	34-59	30.84 ± 0.92	19.6-37	14.21 ± 0.48	9.9-14.1	8.6 ± -55	4.9-13.2

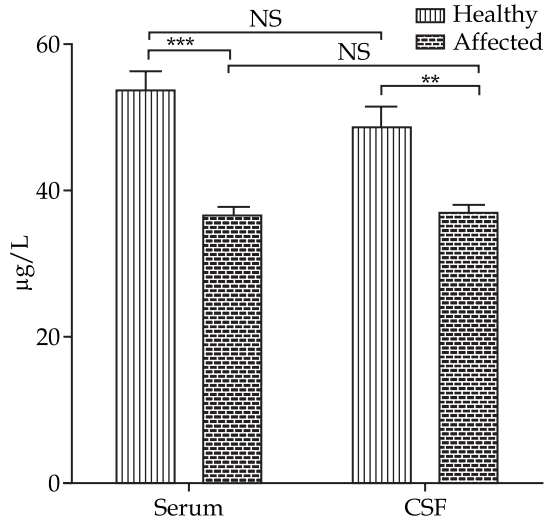

Fig 1. Thiamine (B1) levels (µg/l) in serum and cerebrospinal fluid (CSF) of healthy and camel with neurological disorders. NS>0.05, **<0.01, ***<0.001.

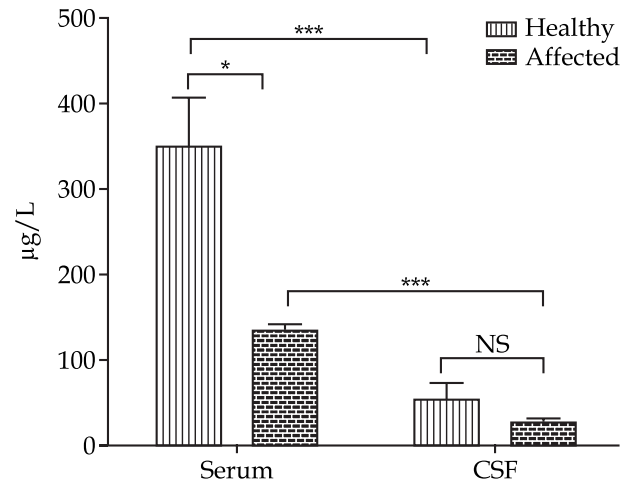

Fig 2. Riboflavin (B2) levels (µg/l) in serum and cerebrospinal fluid (CSF) of healthy and camel with neurological disorders. NS>0.05, *>0.05, **<0.01, ***<0.001.

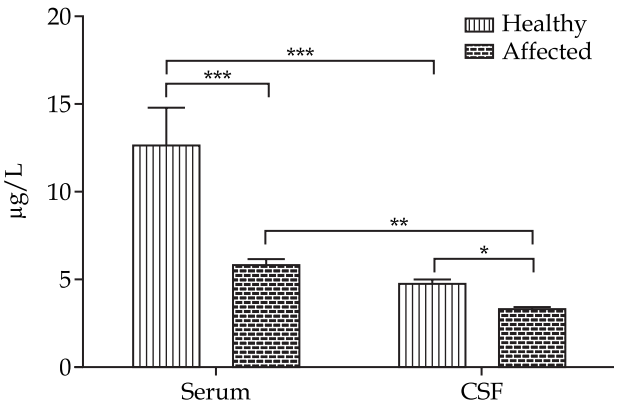

Fig 3. Pyridoxine (B6) levels (µg/l) in serum and cerebrospinal fluid (CSF) of healthy and camel with neurological disorders. *>0.05, **<0.01, ***<0.001.

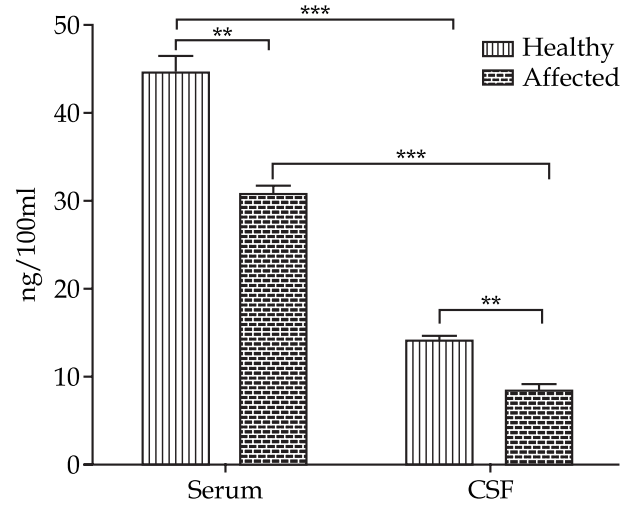

Fig 4. Cobalamin (B12) levels (ng/100ml) in serum and cerebrospinal fluid (CSF) of healthy and camel with neurological disorders. **<0.01, ***<0.001.

healthy camels comparing to its concentration in serum agreed with Spector and Johanson (2007). In agreement with our results, Pedraza and Botez (1992) reported lower levels of thiamin in CSF than those in blood in people affected with ataxia. Similar to our results for thiamine concentration in CSF of healthy and affected animals Jimenez-Jimenez *et al* (1999), found no difference for thiamine levels between healthy and affected people.

There is no data on plasma (Faye and Bengoumi, 2018) or CSF riboflavin concentration in camels. However, it is stated that there are riboflavin entered and transported to CSF at the blood-brain barrier, which can control the riboflavin in the CSF (Spector and Johanson, 2014). The treatments using vitamin B injection including riboflavin in camel affected by hematuria appeared effective (Bhandare, 2009). The deficiency of riboflavin in the current study in affected animals may be attributed to the fact that the diseased animals with neurological symptoms suffered from loss of appetite or difficulty in eating, or there was a decrease in the formation of riboflavin in the digestive system, as a result, some digestive tract disorder leads to neurological symptoms.

Vitamin B6 is closely associated with the function of the nervous system (Ahmad *et al*, 2013). There are no previous studies that reported vitamin B6 concentration in the blood of healthy or diseased

camels (Faye and Bengoumi, 2018). The results of vitamin B6 in the serum of healthy camels in the present study were higher than those reported in people (Spinneker *et al,* 2007). On the other hand Bisp *et al,* (2002); Jungert *et al* (2020) reported lower results for Vitamin B6 in plasma than those reported in the present study. The variation in our results among previous studies in human may be due to the different nature of nutrition between ruminants and humans, as the camel eats plant-based foods while their digestive system synthesises this vitamin (Wernery *et al,* 2009), while humans depend on their animal-based foods with has a higher bioavailability to absorb vitamin B6 (Reynolds, 1988). On the other hand, the method of analysing this compound in the blood, as there are many associated forms of it (Spinneker *et al,* 2007), could affect the variation of its levels among studies (Zhang *et al,* 2018). There is a relationship between B6 levels in serum and CSF (Albersen *et al,* 2015). Spector and Johanson (2007) reported higher results for vitamin B6 in CSF of a rabbit than that reported in the current study. However, Albersen *et al* (2014) reported details for parts of vitamin B6, who stated that pyridoxal phosphate and pyridoxic acid were higher in plasma than in CSF, while pyridoxamine and pyridoxal were higher in CSF than plasma.

Vitamin B12 deficiency was not clinically reported in the camel (Faye and Bengoumi, 2018). Mohamed (2006) reported lower results for cobalamin concentration in plasma (around 25 ng/100 ml) of a healthy camel than that reported in the present study (44.8 ng/100 ml). In contrast to our results for the levels of cobalamin for healthy camels, lower levels were reported in the plasma of healthy sheep (Clark *et al,* 1989). In agreement with the levels of cobalamin in the serum of healthy camels in the present study, Kather *et al* (2020) reported a similar range in dogs. Indeed, higher results for vitamin B12 levels in the serum of healthy people and people affected with neurological diseases were reported comparing to the present results in healthy and diseased camels (Nijst *et al,* 1990). However, Christine *et al,* (2020); Regland *et al* (1992) reported a strong correlation in Vitamin B12 levels between serum and CSF. Higher results were reported for vitamin B12 in CSF of healthy people compared to results in the current study (Nijst *et al,* 1990). Similar studies for higher results of vitamin B12 for serum and CSF in both healthy and affected people were reported (Christine *et al,* 2020; Simpson 1964). The discrepancy of results for B12 in serum and CSF of healthy and affected camels in the current study may be due to several factors, including the pathogenesis of the neurological disorders in camels, which are still not well studied.

Conclusions

We concluded that the affected camel with neurological signs had a deficiency in vitamins B1, B2, B6 and B12.

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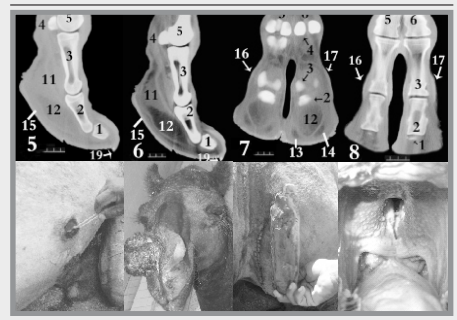

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EVALUATION OF TOTAL MIX RATION WITH DIFFERENT PROPORTIONS OF ROUGHAGES IN DRAUGHT CAMELS

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ABSTRACT

An experiment was conducted using 3 dromedary camels (498-531 kg b.wt) aged 8-10 years in latin square design to evaluate the effect of feeding different proportions of roughages on nutrient utilisation, physiological responses and draught performance under sustained working. Animals were fed total mix ration (TMR) containing roughages and concentrate mixture in the ratio of 70:30. In roughages, soybean straw (*Glycine max* L.) and gram straw (*Cicer arietinum* L.) were mixed in one of the three ratios 75:25, 50:50 and 25:75 which were designated as T_1, T_2 and T_3, respectively. The DCP and TDN contents were higher in camels fed on total mix ration containing 50:50 proportion of soybean straw (*Glycine max* L.) and gram straw (*Cicer arietinum* L.) as compared to other treatment groups. The nutrient intake was higher in T_2 while, T_1 and T_3 were at par with each other. The difference for DDMI and DOMI was also significant between the treatment groups. The CPI, DCPI and TDNI ($g/kgW^{0.75}$) were significantly higher in T_2 whereas, T_1 and T_3 exhibited non-significance difference. A four wheel cart was used as a loading device for applying load cell between the body of cart and the beam for measuring the draught capacity. There was non-significant difference between the treatments for initial body weight, final body weight, body weight gain, average daily gain and draught (kgf). However, the speed of operation and power (hp) was significantly higher in T_2 as compared to T_1 and T_3. The results may conclude that feeding of 50:50 proportions of soybean and gram straw in TMR has positive effect on nutrient utilisation and draught performance in dromedary camels without any detrimental effect on physiological responses under sustained working.

Key words: Camels, draught, feeding, nutrient utilisation, physiological responses, TMR

Camels with poor feeding methods and conditions in their native habitat have lower productivity compared to other animals (Topps, 1975; Mousa *et al*, 1983). Fibrous crop residues and agro-industrial by-products play an important role as a source of feed for ruminants, but the utilisation of these feeds is limited because of poor nutrient content and digestibility. Furthermore, the availability of straws is widespread and they play a strategic role particularly in times of scarcity. Crop residues is a by-product from the main agriculture produce and feeding value of these crop residues can be increased by incorporating them into total mixed rations (TMR) by fortifying them with required nutrients (Sharma *et al*, 2010). Present study was therefore planned to observe the effect of total mix ration with different proportions of soybean straw and gram straw on digestibility, nutrient intake, body weight changes, draught performance and physiological responses in draught camels.

Materials and Methods

This study was conducted in 3x3 latin square design using 3 adult (~8-10 years) dromedary Mewari draught camels weighing 498-531 kg to investigate the effect of total mix ration with different proportions of soybean straw and gram straw on nutrient utilisation, draught performance and physiological responses. The animals were fed on total mix ration (TMR) containing roughages and concentrate mixture in the ratio of 70:30. In roughages, soybean straw (*Glycine max* L.) and gram straw (*Cicer arietinum* L.) were mixed in one of the three ratios 75:25, 50:50 and 25:75 which were designated as T_1, T_2 and T_3, respectively. The concentrate mixture was fed as per requirement of draught camels (ICAR, 1985). Concentrate mixture prepared by grinding of feeds ingredients in hammer mill and feed mixer was used for evenly mixing of all the ingredients. Concentrate was prepared at monthly intervals in which ingredients purchased at the start of experiment were used.

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The experiment was conducted for a period of nine weeks having three different phases of three weeks each. Each animal was offered one of the treatments at a time for a period of 3 weeks. The 3rd week of each phase under each treatment was considered as experimental period for collection of data. During the collection period, the daily feed consumption, leftover as well as faeces voided during preceding 24h were recorded at 9.00 hrs. The total dung voided by each camel during digestibility period (3^{rd} week of each phase) for 24 hours was collected with the help of specially designed faecal collection bags. The representative samples of feeds and faeces were pooled and analysed for proximate principals (AOAC, 2005).

The camels were operated at a draught level of 14 per cent of their BW with a work (W)-rest (R)-cycle of 2h W-1h R-2h W-4h R-2h W-1h R-2h W during the experimental period (Gupta *et al,* 2014). The animals were trained for carting and were developed endurance of working for 4-6 hr daily. For applying the load cells (Dynometer of 500 kg Ecl, UK) between the body of the cart and the beam for measuring the draught, four wheeled camel cart was used as a loading device. The cart was pulled on track to cover an approximate distance of 25.5 km daily in 4 to 5 hrs. The camels were allowed to pull payload in such way that the experimental camels could exert an average draught of 18 per cent of their body weight. The speed (km/h) and draught (kgf) were recorded for 5.1 km span and cumulative 25.5 km distance during the experiment and power was calculated using the standard formula:

$$P=\frac{dxs}{270}$$

Where, P= Power developed (hp), d= Draught (kgf), s=Average speed (kmh^{-1})

Physiological responses such as respiration rate, pulse rate and rectal temperature of the camels were recorded before and after carting. The data obtained were analysed by using one-way ANOVA as per the procedure prescribed by Snedecor and Cochran (1994).

Results and Discussion

Chemical composition: The chemical composition of feeds and fodders fed to the draught camels during the investigation is represented in Table 1. Crude protein (CP) content was 6.97 per cent in gram straw while, it was 6.61 per cent in soybean straw. Gram straw and soybean straw had ether extract (EE) of 1.21 and 1.52 per cent, respectively. Crude fibre (CF), nitrogen free extract (NFE) and organic matter (OM) were higher in soybean straw as compared to gram straw while, total ash (TA) was more in gram straw as compared to soybean straw. These results are in confirmation with Gupta *et al* (2014) who reported similar composition of gram straw. However, Gupta and Murdia (2002) and Gupta *et al* (2011) reported lower values of CP, EE, NFE and TA in gram straw as compared to present study. Significantly higher CP contents were reported by Nagpal *et al* (2010) in guar phalgati (7.42%) and groundnut haulms (8.25%) as compared to gram and soybean straw. Similarly, higher crude protein content (14.2%) was reported by Bui (1998) in peanut haulms.

Nutrient Utilisation: The digestible crude protein (DCP) and total digestible nutrient (TDN) contents were 5.77 and 54.10; 6.12 and 55.11 and 6.09 and 53.49, respectively in T_1, T_2 and T_3 (Table 2). The DCP and TDN contents were higher in camels fed on total mix ration containing 50:50 proportion of soybean straw (*Glycine max* L.) and gram straw (*Cicer arietinum* L.) as compared to other treatment groups. The DCP and TDN contents were higher that that reported by Gupta *et al* (2011) on feeding different levels of energy diets along with gram straw based ration. Gupta *et al* (2012b) also reported lower values of DCP and TDN as compared to present study in dromedary camels fed on different proportions of groundnut haulms and cluster bean straw. However, Choudhary *et al* (2008) reported significantly higher

Table 1. Proximate chemical composition (% DM) of feed and fodder offered to draught camels.

Feed	DM	CP	EE	CF	TA	NFE	OM
Cotton seed cake (*Gossypium hirsutum*)	89.91	21.54	7.56	25.91	6.23	38.76	93.77
Barley (*Hordeum vulgare*)	90.22	10.21	2.71	6.91	2.89	77.28	97.11
Wheat Bran (*Triticum aestivum*)	90.31	10.93	3.05	9.56	5.24	71.22	94.76
Green gram churi (*Vigna radiata*)	89.82	18.21	3.08	15.42	8.2	55.09	91.80
Gram straw (*Cicer arietinum*)	90.01	6.97	1.21	37.28	12.86	40.12	89.29
Soybean straw (*Glycine max*)	90.74	6.61	1.52	42.06	11.29	40.35	91.27

values of DCP and TDN contents as compared to present investigation.

Table 2. Nutrient utilisation in draught camels.

Attribute	Treatments			SEM
	T_1	T_2	T_3	
Nutrient Intake:				
DDMI (kg/d)	6.37	7.35	6.36	0.347
DOMI (kg/d)	6.42	7.04	6.53	0.617
DMI (kg/$w^{0.75}$)	95.42^b	102.07^a	95.80^b	1.012
CPI (g/kg$W^{0.75}$)	8.79^b	9.49^a	8.94^b	0.109
DCPI (g/kg$W^{0.75}$)	5.51^b	6.27^a	5.83^b	0.197
TDNI (g/kg$W^{0.75}$)	51.66^b	56.41^a	51.25^b	1.574
Nutritive Value:				
DCP (%)	5.77^b	6.12^a	6.09^b	1.155
TDN (%)	54.10^b	55.11^a	53.49^b	1.052

^{a,b,c} Values with different superscripts differ significantly from each other.

The nutrient intake analysis indicated that dry matter intake (kg/w0.75) was higher in T_2 (102.07) while, T_1 (95.42) and T_3 (95.80) were on par with each other. These results were in agreement with the findings of Rai *et al* (1994). However, Gupta *et al* (2012b) reported non-significant difference between the treatments for dry matter intake on metabolic size basis on feeding different levels of groundnut haulms and cluster bean straw in draught camels. There was significant difference between the treatments for digestible dry matter intake (DDMI) and digestible organic matter intake (DOMI). Shalash (1984) reported that DDMI and DOMI were significantly high in camels fed on 75% groundnut haulms and 25% cluster bean straw which may be due to high palatability of groundnut haulms as it has more proportion of leaves as compared to cluster bean straw which confirms the present results. The crude protein intake (g/kgW0.75) was significantly higher in T_2 whereas, T_1 and T_3 exhibited non-significance difference. The digestible crude protein intake and total nutrient intake (g/kgW0.75) were highest in 50:50 proportion of soybean and gram straw followed by 75:25 and 25:75 proportions but didn't differ significantly. The TDN intake also follows the same trend i.e., the values were significantly higher in T_2 but T_1 and T_3 were at par with each other. Nagpal *et al* (2005) reported significantly higher DCP and TDN intakes in camel calves fed on complete ration containing gram straw, groundnut forage and concentrate in the ratio of 60.3:25.0:14.7 in feed blocks which confirm the results of present investigation. Likewise, Nagpal *et al* (1996), Nagalaksh and Reddy (2001) and Nagpal and Arora (2002) reported higher nutrient intake in complete ration fed animals than those fed on conventional diets.

Body Weight and Draught Performance: The results indicated non-significant difference for the initial body weight, final body weight, body weight gain and average daily gain (g/day) among the three treatments but the values were comparatively higher in 50:50 proportions of soybean and gram straw. The draught performance of camels is depicted in Table 3. The feeding of different proportions of gram and soybean straw didn't affect the draught (kgf) exerted by the camels. The speed of travel was significantly higher in T_2 (3.25 km/h) while, T_1 (3.00 km/h) and T_3 (3.04 km/h) were at par with other. Similarly, significant high power was developed in 50:50 proportions of soybean and gram straw but 75:25 and 25:75 proportions had non-significant difference. Similar results for draught and power output were confirmed by Gupta *et al* (2011). The speed varied in range from 2.97 to 2.75, 2.5 to 2.43 and 2.26 to 2.2 km/h in I, II, III and IV session and the rate of decrease was 7.4, 3.0, 7.6 and 2.6%, respectively on feeding gram straw along with supplementation (Gupta *et al*, 2014).

Table 3. Body weight and draught performance in camels.

Attributes	Treatments			SEM
	T_1	T_2	T_3	
Body weight (BW)				
Initial body weight (kg)	515.33	516.67	518.33	10.645
Final body weight (kg)	560.00	563.67	562.33	7.034
Body weight gain (kg)	44.66	47.00	44.00	9.767
Average daily gain (g/day)	709.00	746.03	698.41	15.011
Draught Performance				
Draught (kgf)	100.80	101.46	101.22	1.266
Speed (km/h)	3.00^b	3.25^a	3.04^b	0.142
Power (hp)	1.12^b	1.25^a	1.14^b	0.049

^{a,b,c}Values with different superscripts differ significantly from each other.

Physiological Responses: The values of rectal temperature, pulse rate, respiration rate, breaths /minute are depicted in Table 4. There was no significant effect on the rectal temperature among the three proportions of soybean and gram straw before work while, T_1 (39°C) had significantly higher

effect on rectal temperature followed by T_2 (38.4°C) and T_3 (38.8°C) after carting. There was no significant effect on the pulse rate and respiration rate before and after work. However, the per cent increase was higher in 25:75 proportions of soybean and gram straw, followed by 75:25 and 50:50 proportions. In contrast, non-significant difference for rectal temperature and significant difference for pulse rate and respiration rate was reported by Gupta *et al* (2012a). The physiological responses of the camels viz, pulse rate, respiration rate and rectal temperature increased with duration of work where as speed of operation decreased with duration of work (Gupta *et al*, 2014).

Table 4. Physiological responses in camels and respiration rate, breaths/minute.

Attributes	Treatments			SEM
	T_1	T_2	T_3	
Rectal Temperature, °C				
Before work	36.50	36.46	36.73	0.364
After work	39.00[a]	38.40[b]	38.80[b]	0.226
% Increase	6.85	5.30	5.63	–
Pulse Rate, beats/minute				
Before work	45.33	46.00	45.66	1.122
After work	52.00	52.33	52.66	1.123
% Increase	14.71	13.77	15.33	–
Respiration rate, breaths/minute				
Before work	8.33	8.66	8.67	0.471
After work	16.66	16.00	16.67	0.902
% Increase	100.00	92.00	100.00	–

[a,b,c]Values with different superscripts differ significantly from each other.

Conclusion

It can be concluded that feeding of soybean straw and gram straw in ratio of 50:50 had positive effect on nutrient utilisation and power out as compared to 75:25 and 25:75 proportions. Thus, feeding of total mix ration with equal proportions of soybean straw and gram straw may be recommended for improved nutrient utilisation in dromedary camels undergoing sustained working.

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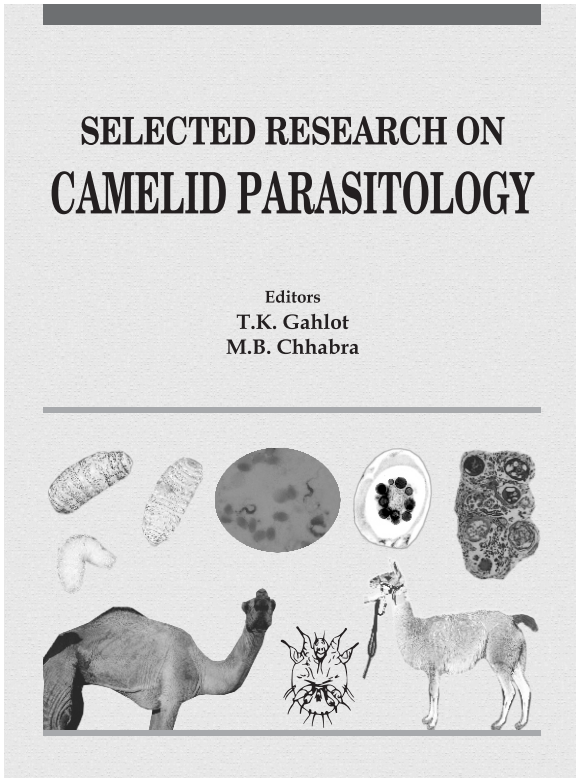

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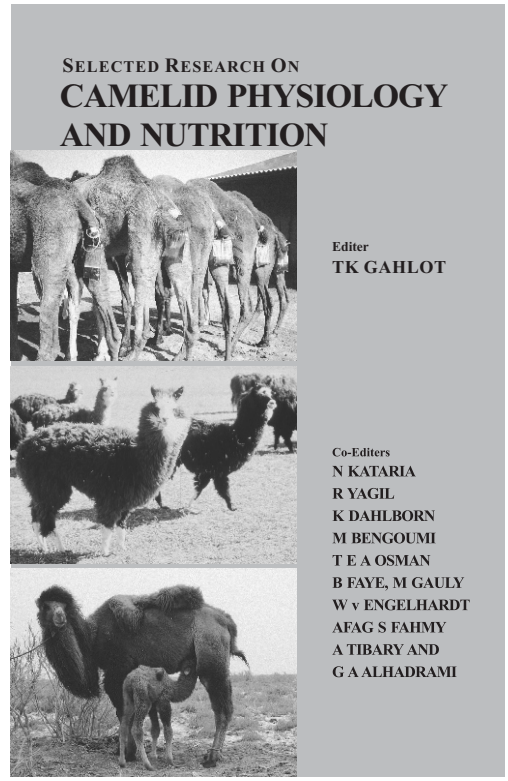

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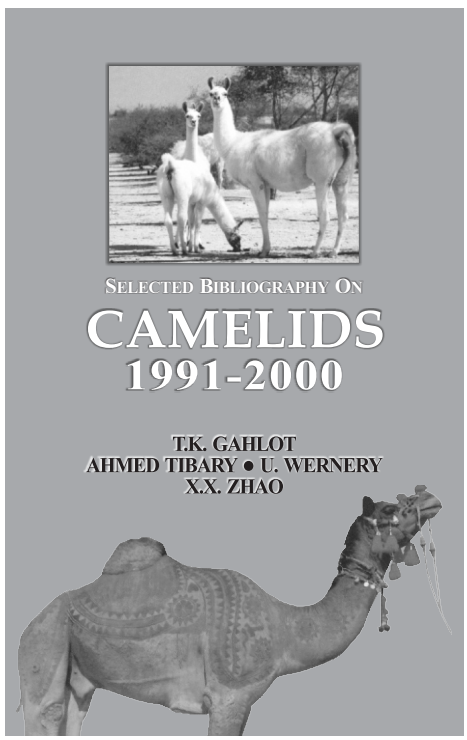

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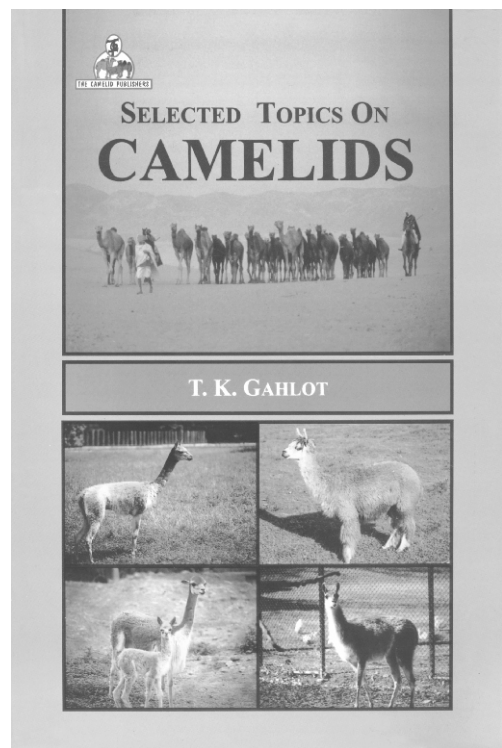

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GROSS ANATOMICAL PECULIARITIES OF TONGUE OF INDIAN DROMEDARY CAMEL

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ABSTRACT

The present study was conducted on tongues obtained from 10 cadavers of adult camels (*Camelus dromedarius*). Gross anatomical features were studied. The tongue of camel was muscular and spatula shaped, comprised of 3 parts - the apex, body and the root. The mean length of tongue was 41.21 ± 0.527 cm. A median groove was present on the dorsal surface of tongue. The caudo-dorsal part had convexity and formed an elliptical dorsal prominence (*torus linguae*), and bounded rostrally by the *fossa linguae*. A median fibrous ridge like structure, "lyssa" was present on the rostral third of its ventral surface. Five types of papillae were identified on the tongue of camel.

Key words: Dromedary camel, tongue

The Indian camel breeds have variable behavioural preferences for diet, feed resources because the anatomy and physiological function of the tongue is well adapted to wide range of feed resources (Serkan *et al,* 2016) in camels living in diverse and extreme climate conditions. Although, previous research studies have been performed on the tongue has been reported *viz.* Indian buffalo (Dhingra and Baranwal, 1979), Bactrian camel (Ye *et al,* 2008). The ultrastructural studies using scanning electron microscopy studies on the papillary body of dromedary tongue have been conducted by Saber *et al* (2011). Gross morphology and morphometry of tongue of antenatal and adult dromedaries (Saidu *et al,* 2015), histology of dromedary tongue (Qayyum *et al,* 1991) and morphological study of muscle of camel tongue (Allouch, 2014) have been studied previously. The paucity in the literature on the gross structure of tongue of the Indian dromedary camel evoked interest to carry out the present study.

Materials and Methods

The tongues were dissected out from 10 cadavers of recently dead adult camels (*Camelus dromedarius*) irrespective of age and sex from clinics of Veterinary Clinical Complex, RAJUVAS, Bikaner. These were free from any pathological condition of tongue and mouth.

Each tongue was then used to study the gross and biometric parameters. The tongue was weighed on a weighing scale. The width and thickness of each tongue was measured by Vernier caliper. The maximum length measured from the tip of the tongue to the median glosso-epiglottic fold was recorded by a measuring scale. Volume of each tongue was recorded by water displacement method. The number of papillae present on the tongue was counted grossly. The data was analysed using standard statistical methods as described by Kaps and Lamberson (2004).

Results and Discussion

Shape and Colour

The tongue of camel was muscular and spatula shaped and comprised of 3 parts - the apex, body and the root (Fig 1). Similar findings were reported by Smuts and Bezuidenhout (1987), Kumar *et al* (1995) and Ramayya *et al* (2012) in dromedary camel, Ye *et al* (2008) in Bactrian camel, Raghavan (1964) in Ox, Parvez and Rahaman (2005) in cow and Mahabady *et al* (2010) in Iranian buffaloes. The apex was free, flattened, wide and rounded as described previously (El Sharaby *et al,* 2012) in camels. It presented dorsal and ventral surfaces and a median groove was present on the dorsal surface (Fig 1). Similar findings were obeserved by Ye *et al* (2008) in Bactrian camel. Although, Ye *et al* (2008) reported crinkled appearance of the apex of tongue on dorsal surface, but no such observations have been recorded in the present study. The presence of papillated structures on the ventral margin of the apex of tongue (Fig 2) was

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in congruence with the observations of Harłajczuk *et al* (2015) in Alpacas. A median fibrous ridge like structure "*lyssa*" was observed on the rostral third of the tongue at ventral surface, situated 2 - 3 cm away from the tip and extended caudally to the level of *frenum linguae* (Figs 1 and 2) was in conformity with the findings in camels (Shoeib *et al,* 2014) and in dogs (Eubanks, 2007). However, Besoluk *et al* (2006) reported the helical shaped *lyssa* in cats and more or less J-shaped in the dogs. Shoeib *et al* (2014) reported the *lyssa* as rod-shaped in dog and strip-like structure in cat.

The body of the tongue was wider and thicker caudally than the apex and narrower rostrally (Fig 1). These findings were not in conformity with the Ye *et al* (2008) who reported long and slender body of the tongue in camels. It had 4 surfaces *viz.* dorsal, 2 lateral and ventral surfaces. The rostral part of the dorsal surface of the tongue was flat and the caudal part had convexity and formed an elliptical dorsal prominence - *torus linguae*, which appeared to be more, pronounced in the centre than on its lateral margins (Fig 1). These findings were in close agreement with the reports in Indian buffalo (Dhingra and Barnwal, 1979), cow (Parvez and Rahaman, 2005), Bactrian camel (Ye *et al,* 2008) and Egyptian water buffalo (El-Bakary and Abumandour, 2017). However, Bradley (1971), in ruminants named this elliptical dorsal prominence as intermolar eminence. The *torus linguae* has been found bounded by the *fossa linguae*, rostrally which restricted the torus between 2nd and 3rd cheek teeth in the middle of the tongue (Fig 1). Similar observations were reported by Gupta *et al* (1989) in tongue of buffalo. Ramayya *et al* (2012) reported the absence of *fossa linguae* in the tongue of camel. The *fossa linguae* was crescent shape (Fig 1) as reported by Dhingra and Barnwal (1979) in Indian buffalo. The lateral surfaces were nearly flat for the most part, but rostrally became rounded and narrower. The rostral part of the ventral surface of the tongue was free and the caudal part was related to the mylohyoid muscle, which was similar to the findings reported in ox (Raghavan, 1964). The root of the tongue was slightly narrower than body and sloped caudo-ventrally. The tongue of the camel had light pink appearance of dorsal surface and grayish pink ventral surface (Figs 1 and 2). Black pigmented patches or spots were occasionally found on the ventral surface of the tip and body (Fig 3). Whereas pigmented spots on the dorsum of mucous membrane of tongue of ox were reported by Sisson and Grossman (1958).

Size and Weight

The morphometric studies demonstrated the mean length of the tongue from the tip to the root 41.21 ±0.527 cm. Similar findings were reported by Kumar *et al* (1995) and Ramayya *et al* (2012) in camels. However, Ye *et al* (2008) reported the length of tongue in a range 25-32 cm in Bactrian camels and El-Bakary and Abumandour (2017) reported 47 ± 1.2 cm in Egyptian water buffalo. The mean length from root to lingual fossa and lingual fossa to tip of tongue was 16.83 ±0.202 cm and 24.38 ± 0.462 cm, respectively.

The mean thickness of tongue at *torus linguae* was 6.825 ±0.409 cm, at lingual fossa 4.362 ± 0.284 cm and at tip 1.235 ± 0.108 cm. The mean width of tongue at root was 8.117 ±0.209 cm, at lingual fossa 3.916 ± 0.191 cm and 5.479 ± 0.117 cm at the tip. Kumar *et al* (1995) reported maximum width at root of tongue 7.12 ± 0.94 cm in camel, however, Ramayya *et al* (2012) noticed maximum width at the level of *torus linguae* in same species. According to El-Bakary and Abumandour (2017), the tongue of Egyptian water buffalo was 7 ± 0.51 cm wide at its middle part. The mean weight of tongue recorded 0.570 ± 0.021 Kg and mean volume 0.572 ± 0.021 litres.

Topography

The tongue rests on the floor of mouth cavity, between the two horizontal rami of mandible (Figs. 1 and 3). It was in agreement with Raghavan (1964) in ox, Gupta *et al* (1989) in buffalo and Ye *et al* (2008) in Bactrian camel. It extended from glosso-epiglotic fold to the lingual surface of central incisors (Fig 1), whereas, Gupta *et al* (1989) found the tongue extended from the glosso-epiglottic fold to about 1 cm rostral to the level of sublingual caruncle in buffalo. Kumar *et al* (1995) found camel tongue extended from the rostral part of the floor of mouth cavity to the level of the oropharynx.

The caudal 3/4th portion of the tongue was fixed and rostral 1/4th was free which was also reported by Dhingra and Barnwal (1979) in Indian buffalo. On the Contrary, Ye *et al* (2008) reported the caudal 4/5th of tongue was fixed while the rostral 1/5th was free in Bactrian camel. The ventral surface of fixed part was placed on the mylohyoid muscle and attached to the dorsal surface of basal part of hyoid bone was in partial accordance with the findings of Ye *et al* (2008) in Bactrian camel. The dorsal surface of fixed part was attached with anterior pillars of the soft palate, and the glosso-epiglottic fold (Fig 1). Similar findings were also reported by Raghavan (1964) in ox. The ventral surface of the free part of the tongue was attached to the floor of the oral cavity by a median

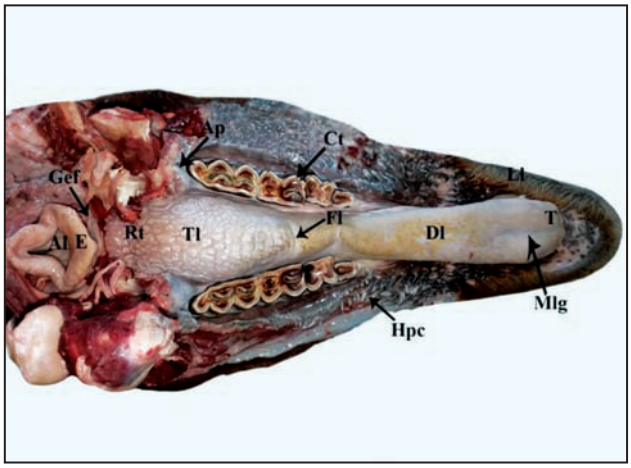

Fig 1. Photograph showing mouth cavity and pharynx of camel. Al - Aditus laryngis, E - Epiglottis, Gef - Glosso - epiglottic fold, Tl - *Torus linguae*, Ct - Cheek tooth, Fl - *Fossa linguae*, Dl - Dorsum linguae, Hpc - Horny papillae of cheek, T - Tip of tongue, Ll - lower lip, Ap - Anterior pillar, Mlg - Median longitudinal groove, Rt - Root of tongue.

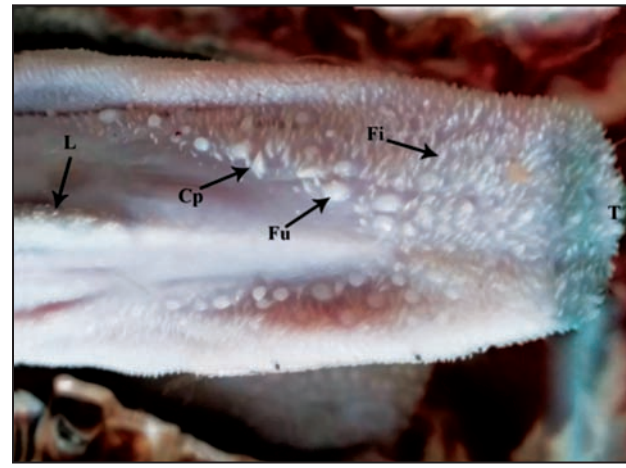

Fig 2. Photograph of ventral surface of tongue of camel showing papillae. Fi - filiform papillae, Fu - Fungiform papilla, Cp - Conical papilla, L - Lyssa, T - Tip of tongue.

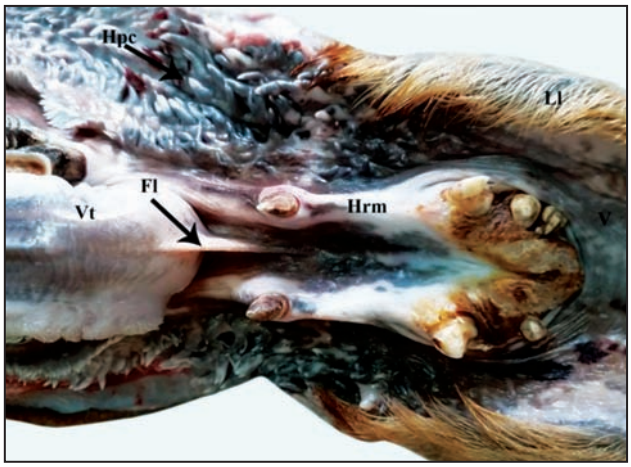

Fig 3. Photograph showing floor of mouth cavity of camel. Vt - Ventral surface of tongue, Fl - Frenulum linguae, Hpc - Horny papillae of cheek, Hrm - Horizontal ramus of mandible, V - Vestibule, Ll - Lower lip.

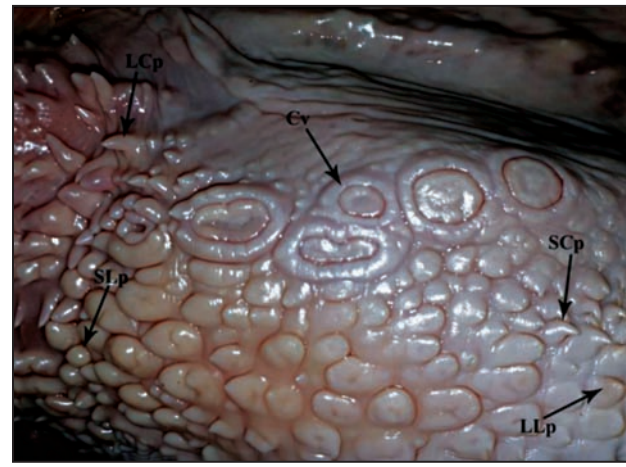

Fig 4. Photograph of dorsal surface of tongue of camel showing circumvallate papilla. Cv - Circumvallate papilla, LCp - Large conical papilla, SCp - Small conical, LLp - Large lenticular papilla, SLp - Small lenticular papilla.

fold of mucosa, the *frenulum linguae* which was extended from caudal end of *lyssa* to 1 - 2 cm rostrally to the 1st premolar (Fig 3). It was in congruence with the observations of Gupta *et al* (1989) in buffalo, Ye *et al* (2008) in Bactrian camel and El-Bakary and Abumandour (2017) in Egyptian water buffalo.

Papillae

Five types of papillae identified on the tongue of camel were categorised as mechanical and gustatory papillae. The filiform, conical and lenticular papillae were categorised as mechanical papillae whereas, fungiform and circumvallate were categorised as gustatory papillae. These were in conformity with the findings of Smuts and Bezuidenhout (1987) in dromedary camel and Peng *et al* (2008) in Bactrian camel. However, it partially contradicted the statement of Mahabady *et al* (2010), who found filiform, conical, lenticular and fungiform papillae as mechanical papillae and circumvallate papillae as gustatory papillae in Iranian buffaloes.

Mechanical papillae

Filiform Papillae

Filiform papillae were the most numerous papillae randomly distributed approximately on the anterior half of the surface of the tongue (Fig 5). It was

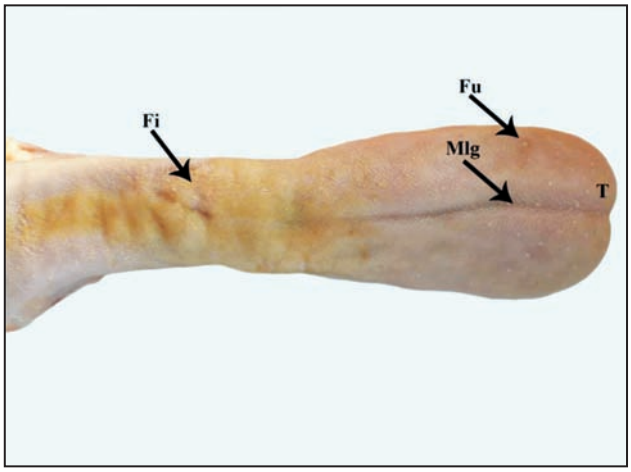

Fig 5. Photograph of dorsal surface of tongue of camel showing filiform and fungiform papillae. Fi - Filiform papillae, Fu - Fungiform Papillae, T - Tip, Mlg - Median longitudinal groove.

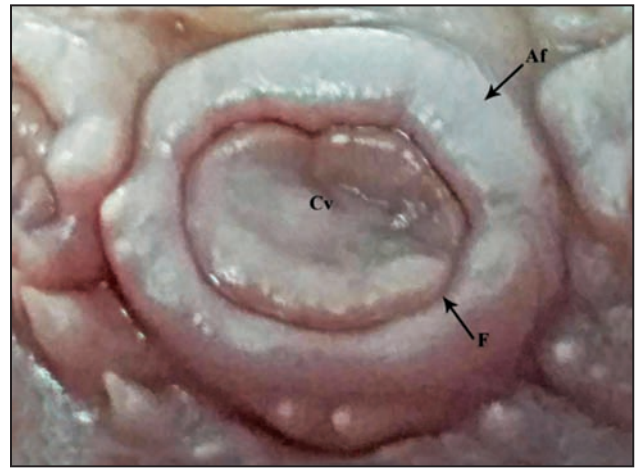

Fig 6. Photograph showing single circumvallate papilla on the tongue of camel. Cv - Circumvallate papillae, F - Furrow, Af - Annular fold.

in agreement with Smuts and Bezuidenhout (1987) in dromedary camel and Ye *et al* (2008) in Bactrian camel. These were thickly populated on the dorsal surface, in front of the *fossa linguae* up to the tip and moderately populated on the ventral surface of the tip (Figs 2 and 5), Similar finding were reported (Parvez and Rahaman, 2005) in cow. No filiform papillae were found on *torus linguae* and root (Fig 4). It was also reported by Ye *et al* (2008) in Bactrian camel however, contradicted with the findings of Mahabady *et al* (2010), according to which filiform papillae were distributed over the entire dorsal surface of the tongue, with the filiform papillae on *torus linguae* shaped as caudally directed pointed spines or conical shape in Iranian buffaloes. The height of the papillae located on the tip of the tongue was low, but increased towards the body of the tongue, which was in conformity with the findings of Ye *et al* (2008) in Bactrian camel. The papillae were triangular or leaf-like shape with a sharp tip pointed backward. It was also reported by Mahabady *et al* (2010) in Iranian buffaloes. The degree of inclination of the filiform papillae increased towards *fossa linguae*.

Conical Papillae

Two types of conical papillae, i.e. large and small were located only on the *torus linguae*. The larger papillae were located mainly on caudal margin of *torus linguae* and directed caudally, with flat and blunt free end. The small conical papillae were located on lateral margins of torus rostrally to the circumvallate papillae and directed cranially. The free ends of conical papillae were pointed (Fig 4). Few horny conical papillae were also found on the ventral surface of the tip (Fig 2). Parvez and Rahaman (2005) reported that the large conical papillae were located in the rostral two-thirds of the dorsum of the tongue with a higher concentration in the middle portion of the tongue in cow (*Bos indicus*).

Lenticular Papillae

Lenticular papillae were largest among the mechanical papillae, limited on the *torus linguae* (Fig 4) as described by Smuts and Bezuidenhout (1987) in camel and Mahabady *et al* (2010) in Iranian buffaloes. These were of two types, i.e. larger and smaller. The larger papillae were arranged in two parallel rows and located in the middle of the anterior two-third of the torus while the smaller papillae were distributed laterally in the posterior third of the torus. Their free surfaces were round or flattened (Fig 4).

Gustatory papillae

Fungiform Papillae

The fungiform papillae were small, round and club shaped (Fig 5) as also reported by Raghavan (1964) in ox. These were irregularly distributed among the filiform papillae in the anterior two-thirds of the dorsal surface and ventro-lateral surface of the tongue (Fig 5), also reported previously in camels (Smuts and Bezuidenhout, 1987). These papillae were maximally distributed on the dorsal and lateral aspects of the tip of the tongue, with few on the ventral surface of the tip (Figs 2 and 5) which were in agreement with Raghavan (1964) in ox and El-Bakary and Abumandour (2017) in Egyptian water buffalo. No fungiform papillae were found on *torus linguae* and root (Fig 4) whereas, it was contrary to

the findings of Mahmoud *et al* (2002) in donkeys. These were slightly elevated from the lingual surface as described by Mahabady *et al* (2010) in Iranian buffaloes. Fungiform papillae decreased in number but increased in size from the tip to lingual fossa which, were in the accordance with the findings of Gupta *et al* (1989) in buffalo. The ventral surface was also papillated. The filiform and fungiform papillae extended for about 5 to 6 mm beyond the tip (Fig 2).

Circumvallate Papillae

Circumvallate papillae were found arranged in a single row on both rims of the *torus linguae*. These were 12-16 in number, with 6-8 distributed on either side (Figs 1 and 4). These findings were in partial disagreement with Smuts and Bezuidenhout (1987), as the vallate papillae were located on the torus, along its lateral borders, and consisted of a single row of 3 to 6 prominent papillae in same species. Kobayashi *et al* (2005) reported 15 or more vallate papillae at the posterior area of the lingual prominence in cattle although, Parvez and Rahaman (2005) reported 12-20 papillae in number on either side in cattle. Further, El Sharaby *et al* (2012) also reported 4-6 large vallate papillae arranged on each side closer to one another forming two lines almost parallel to the rim of lingual torus in one-humped camel whereas, Ramayya *et al* (2012) reported that circumvallate papillae were 4 on right side and 5 on left side on caudo-lateral aspect of the tongue in camel. The papillae were round in shape with minute elevation from the tongue surface. Each papilla was separated from the surrounding thick annular fold by a prominent furrow (Fig 6). The shape and size of these papillae varied greatly and these were not identical or symmetrical in the lines of either side even in the same specimen (Fig 1 and 4) also reported in camels previously (El Sharaby *et al* 2012). In some specimens, two papillae were found surrounded by a common annular pad and primary grooves (Fig 4).

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Hyalomma dromedarii TICKS INDUCE A DISTINCT ACUTE PHASE REACTION IN DROMEDARY CAMELS

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ABSTRACT

The effect of tick infestation in camels on the serum concentrations of the acute phase proteins (APPs) haptoglobin (Hp) and serum amyloid A (SAA) has been described. Twenty-three dromedary camels, naturally infested with *Hyalomma dromedarii* ticks were used. Twelve clinically healthy adult camels were used as controls. From both groups, sera were harvested and analysed for Hp and SAA concentrations. There were statistically significant elevations of Hp and SAA in tick-infested camels compared to uninfested healthy controls. Increases in Hp concentrations in the disease group were estimated to be 4.6 fold (0.52 ± 0.27 mg/L in controls *vs* 2.42 ± 0.51 mg/L in tick infested camels). However, increase in serum SAA were 10.9 folds (0.61 ± 0.25 ng/mL in controls vs 6.67 ± 1.97 ng/mL in tick infested camels). In conclusion, a remarkable acute phase reaction occurs in camels naturally infested with *Hyalomma dromedarii* ticks, and SAA is a more sensitive biomarker for tick infestation in camels than Hp.

Key words: Acute phase reaction, biomarkers, dromedary, *Hyalomma dromedarii*, ticks

Ticks are obligatory haematophagous arthropodes and transmit many tick borne diseases. It may cause severe toxic conditions such as paralysis and toxicosis, irritation and allergy, local skin necrosis resulting in low-quality hides and lowered productivity in terms of weight gain and milk yield (Mehlhorn, 2008).

The acute phase proteins (APPs) are a group of serum proteins that change their concentration in animals following external or internal challenges, and likely play a role in host immunological defense. This response is called the acute-phase response or acute phase reaction (APR), occurring secondary to many types of tissue injury and might be a physiological protective mechanism during inflammatory events (Petersen *et al,* 2004). APPs are synthesised in the liver in response to the systemic presence of high levels of pro-inflammatory cytokines. The principal pathway leading to production of APPs involves initial release of pro-inflammatory cytokines by macrophages at the site of infection or inflammation. The most important inducers of APP are cytokines of the interleukin-1 (IL-1), tumor necrosis factor (TNF) and interleukin-6 (IL-6) families (Eckersall, 2000).

In camel medicine, haptoglobin (Hp) and serum amyloid A (SAA) are the commonly used APPs. The APR can be used in camels for general health assessment. The serum concentrations of APPs has been studied in camels in the non-diseased state during the periparturient period (Tharwat and Al-Sobayil, 2015), following stimulation by electroejaculation (Tharwat and Al-Sobayil, 2018a), and following race (Tharwat and Al-Sobayil, 2018b). These studies suggest that Hp and SAA are sensitive biomarkers and can be used in camels as biomarkers of infection and inflammation (Tharwat, 2020). The aim of the present study was to evaluate the effect of tick infestation in dromedary camels naturally infested with ticks on the serum concentrations of the two major APPs, namely Hp and SAA as markers of APR.

Materials and Methods

Camels, experimental design and blood sampling

The experimental protocol was approved by the Animal Ethical Committee, Deanship for Scientific Research, Qassim University, Saudi Arabia. The experimental design had been reported recently (Tharwat and Al-Sobayil, 2014). Briefly, 23

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Arabian camels (*Camelus dromedarius*), 14 females and 9 males (age: 6.8±2.4 y; weight: 460±115 kg) were admitted to the Veterinary Teaching Hospital, Qassim University for treatment of tick infestation. During the course of the disease, camels showed symptoms of anorexia, incoordination of movement, unsteady gait, recumbency, opisthotonus, anaemia and reduced production. *Hyalomma dromedarii* was identified as the predominant tick species. Twelve clinically healthy adult camels were used as controls. From both groups, blood samples were collected in plain tubes and sera were harvested and stored pending Hp and SAA analysis.

Acute phase protein assays

The serum concentration of Hp was determined in the sera using a colorimetric assay (Haptoglobin kit, second generation, Tridelta Ltd., Ireland) as reported recently (Tharwat and Al-Sobayil, 2015; Tharwat and Al-Sobayil, 2018a; Tharwat and Al-Sobayil, 2018b). The analytical sensitivity of the assay was 0.0005 mg/mL, and intra- and inter-assay CVs were 5-6% and 4-6%, respectively. The SAA was measured in the sera using a commercially available ELISA kit (Multispecies SAA ELISA kit, Tridelta Ltd., Ireland). A monoclonal antibody specific for SAA has been coated onto the wells of the microtitre strips provided. The analytical sensitivity of the assay was 0.15 µg/mL and intra- and inter-assay CVs were 4.5% and 6%, respectively.

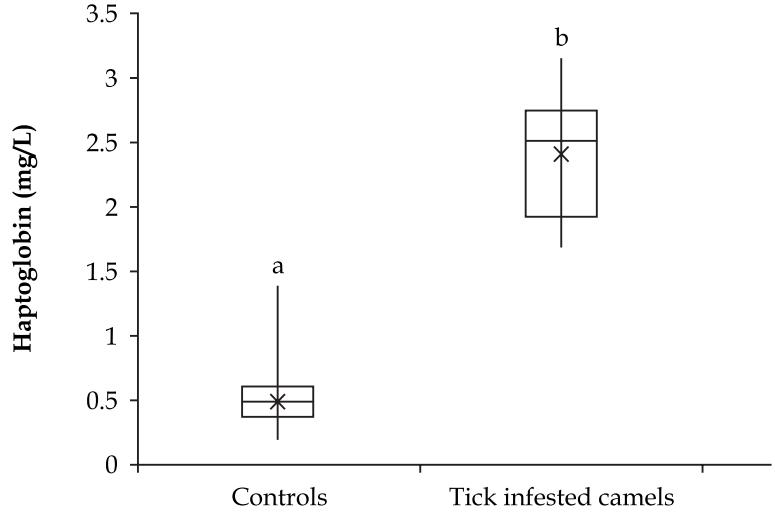

Fig 1. Box and whiskers plots of serum haptoglobin in diseased camels with tick infestation compared to healthy controls. [a,b]Values with different letters differ significantly ($P<0.0001$).

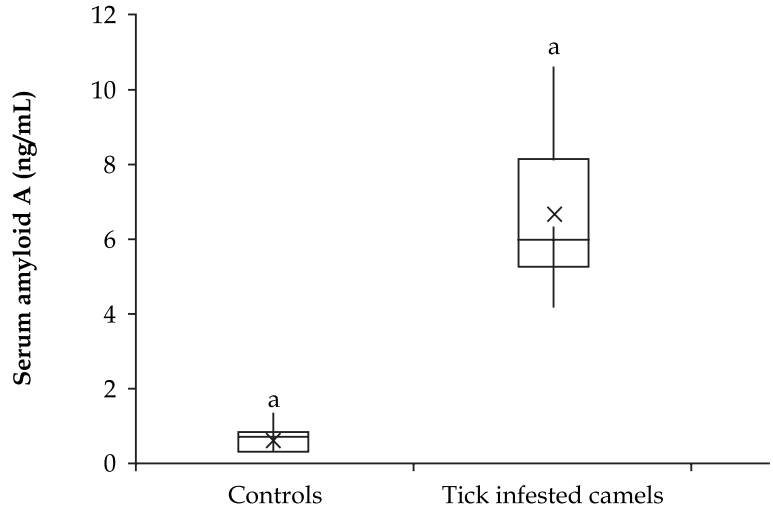

Fig 2. Box and whiskers plots of serum amyloid A in diseased camels with tick infestation compared to healthy controls. [a,b]Values with different letters differ significantly ($P<0.0001$).

Statistical analysis

Data are presented as means ± standard deviation. A statistical program was used to perform the statistical analyses (SPSS, 2009). The graphical representation of the results was performed using MedCalc Software (Mariakereke). A paired *t-test* for repeated samples was used for comparisons between Hp and SAA values in camels infested with ticks and control values. Significance was set at P<0.05.

Results and Discussion

To the author's knowledge, this is the first report to evaluate the effect on *Hyalomma dromedarii* tick infestation in dromedary camels on the serum concentration of APPs as a marker of APR. The minimum, 25th, 50th and 75th, maximum and mean of serum Hp in camels with tick infestation were 1.7, 1.9, 2.5, 2.7, 3.2 and 2.4 mg/L, respectively compared to 0.2, 0.4, 0.5, 0.6, 1.2 and 0.5 mg/L in controls (Fig 1). Increases in Hp concentrations in the camel with naturally-acquired tick infestation compared to healthy uninfested controls were estimated to be 4.6 fold (0.52 ± 0.27 mg/L in controls *vs* 2.42 ± 0.51 mg/L in tick infested camels) (P<0.0001).

The minimum, 25th, 50th, 75th, maximum and mean of SAA in camels with tick infestation were 4.2, 5.3, 6.0, 8.1, 10.6 and 6.7 ng/mL, respectively compared to 0.3, 0.3, 0.7, 0.8, 1.1 and 0.6 ng/mL, respectively in the controls (Fig 2). Increase in serum

SAA were 10.9 folds (0.61 ± 0.25 ng/mL in controls *vs* 6.67 ± 1.97 ng/mL in tick infested camels) (P<0.0001).

The effect of scab, blood parasites, gastrointestinal and pulmonary nematodes and intestinal protozoa on the APR has been reported in animals. The levels of Hp and SAA has increased in serum following experimental infestation of sheep with *Psoroptes ovis,* becoming statistically significantly elevated from pre-infestation levels at 4 weeks post-infestation. Following successful treatment of infested sheep in the previous study with an endectocide, Hp and SAA serum levels declined rapidly (Wells *et al,* 2013). The serum concentrations of the APPs SAA, Hp, α1-acid glycoprotein (AGP) and ceruloplasmin (Cp) were also higher in Alpine Ibex with clinical signs of *Sarcoptes scabiei* mange when compared to healthy animals (Rahman *et al,* 2010). From the previous study, it was found that SAA and AGP is a major APPs as they increased more than 10 folds, while the increases of Hp and Cp were 2-5 folds so they were classified as minor APPs, a similar finding to our results.

In cattle infected with hydatid cysts, SAA was found to be the major marker in the detection of infection, however Hp was not sensitive marker as it was higher in the control group than diseased one (Sevimli *et al,* 2015). Significant increase were also reported in calves with *Dictyocaulus viviparus* infection (Ganheim *et al,* 2004). Hp concentration was also increased in calves with gastrointestinal and pulmonary nematodes (de Cezaro *et al,* 2016). In calves infected with *Eimeria zuernii* oocysts, serum Hp and SAA levels has increased during the monitoring period of 28 days post-infection (Lassen *et al,* 2015). However, it was reported in the later study that Hp is a more sensitive marker than SAA. In cattle infected with *Theileria annulata*, similar findings were reported (Glass *et al,* 2003). The behaviour of APPs in dairy cattle herd naturally infected with *Trypanosoma vivax* has been investigated (Machado *et al,* 2015). In camels, the serum concentration of the APPs Hp, SAA, Cp and fibrinogen has also been studied in dromedary camels naturally infected with *Trypanosoma evansi* (El-Bahr and El-Deep, 2016).

In the present study, the 4.6-fold and 10.9-fold significant elevations of Hp and SAA, respectively in tick-infested camels reflect the occurrence of severe systemic reaction, probably because the skin inflammation was sufficiently intense to induce a remarkable APR. The local skin necrosis as well as the tick salivary secretions may be the cause of APR induction in this study. The significant elevations of Hp and SAA in tick-infested camels may be attributed to the initial secretion of pro-inflammatory cytokines by macrophages at skin. To secure uninterrupted blood uptake, ticks suppress and evade the complex physiological host immune and homeostatic responses that are raised against them. Haemostasis, which includes coagulation, vasoconstriction, and platelet aggregation, is the first innate host defense mechanism against the mechanical injury caused by intrusion of tick mouthparts into the host skin. This early response further includes complement activation and inflammation, with the host inflammatory response including, among other factors, rapid leukocyte infiltration after skin injury. Pro-inflammatory chemokines and cytokines including IL-8, TNF, and IL 1- β (IL-1β) are released to recruit neutrophils and other inflammatory cells to the area of tick infestation (Kotal *et al,* 2015). Unfortanately, pro-inflammatory cytokines were not determined in this study. In camels, naturally infected with *T. evansi*, there were significant increases in the pool of pro-inflammatory cytokines IL 1- α, IL 1- β, IL-10, IL-6, TNF-α and interferon-γ (El-Bahr *et al,* 2016). It is concluded from this study that a remarkable APR occurs in camels naturally infested with *Hyalomma dromedarii* ticks and SAA appears to be a more sensitive biomarker for tick infestation in camels than Hp.

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LUNG LESIONS ON CAMELS (*Camelus dromedarius*) SLAUGHTERED IN TAMBOUL ABATTOIR, SUDAN: PATHOLOGICAL AND HAEMATOLOGICAL STUDY

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ABSTRACT

This investigation was carried out in Tamboul Abattoir, Central Sudan, to find out and describe the macroscopic and microscopic lesions in camels slaughtered during a period of three months. All examined camels were subjected to haematological analysis before slaughter. Out of 100 condemned lungs 41% had pneumonia, 19% hydatid cysts, 11% emphysema, 9% congestion, 6% fibrosis, 3% aspirated blood, 3% oedema, 3% adhesions, 3% abscesses, 2% calcification and 1% had necrotic foci. The results of the haematological analysis indicated insignificant changes in the erythrocytic indices of camels with pulmonary lesions compared with those of camels with normal lungs. However, the total leucocytes counts were significantly higher in the camels that had lung lesions compared with those without lung lesions. Neutrophils and lymphocytes counts were relatively higher in the camels with lung lesions. Pneumonia and hydatidosis were the main causes of condemnation of lungs of camels slaughtered at Tamboul Abattoir.

Key words: Camels, haematology, lungs, pathology

The non-sedentary nature of camel herds which are constantly moving in search of grazing and water is one of the major constraints that face research in camel diseases including respiratory diseases. Pulmonary lesions as evidence of respiratory diseases of camels have been investigated. Bani Ismail (2017) diagnosed bacterial and viral agents causing pnuemonia in camels. Affected camels had elevated leukocytes, creatinine, urea and protein. Jenberie *et al* (2012) diagnosed pulmonary lesions of camels. Pulmonary lesions in camels were also reported by Abdelrahim *et al* (1990) in Libya, Zubair *et al* (2004) in Pakistan, Nourani and Rohani (2009) in Iran, Abubakar *et al* (2011) in Nigeria and Muskin and Moti (2011) in Ethiopia. Similarly, few studies have been published in this field in the Sudan (Tigani *et al,* 2007 and Nasar Eldien, 2010).

The haematological effects of pulmonary lesions on camels also seem to be investigated by few workers including Abubakar *et al* (2011) in Nigeria. The present work was, therefore, carried out to investigate the pathological and haematological effects of lung lesions on camels in Central Sudan.

Materials and Methods

The study area

The study was conducted at Tamboul Abattoir, 150 km south of Khartoum North, Central Sudan. Tamboul is the largest camel market in the Sudan with more than 90 camels slaughtered every week.

Collection of condemned lungs, histopathological and blood samples

One hundred lungs condemned due to pulmonary lesions were randomly collected from slaughtered camels of both sexes and different ages. The collected lungs were grossly examined and the observed pathological changes were described. Samples were taken from prominent lesions and fixed in 10% neutral buffered formalin. These were processed, embedded in paraffin wax and sectioned at 5µm. Sections were dewaxed and stained with haematoxylin and eosin (H & E) for histopathological examination.

Blood samples for the determination of blood indices were collected before slaughter of all camels in clean vials containing ethylene diamine tetracetic acid (EDTA). Samples from camels identified later as having

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lung lesions were kept while others were discarded except the samples required as normal controls. Twenty three blood samples were randomly chosen from the samples from camels with lung lesions and 14 samples were chosen as controls from normal camels without lung lesions. Haemoglobin (Hb) values were determined by the cyanomethaemoglobin method using a haemoglobinometer (Evans Electroselenium, UK). Packed cell volume (PCV) was measured by a haematocrit centrifuge (Hawksley and Sons Ltd, UK). Total red (RBC) and white (WBC) blood cell counts were made in an improved Neubauer haemocytometer (Hawksley and Sons, UK). The mean corpuscular volume (MCV) and mean corpuscular haemoglobin concentration (MCHC) were calculated according to Kerr (1991). Differential leucocyte counts were made on thin blood films stained with Wright-Leishman stain and at least 200 cells were counted on each film using the battlement technique (Kerr, 1991).

Statistical analysis

Haematological values were analysed using the statistical package of social science (SPSS). Analysis of variance was used to assess the significance of difference. P values higher than 0.05 were considered insignificant.

Results

Causes of lung condemnation:

The different causes of lungs condemnation of slaughtered camels in Tamboul Abattoir are listed in Table 1.

Table 1. Causes of condemnation of lungs from camels slaughtered at Tamboul Abattoir.

Cause of condemnation	Condemned lungs	
	No. of lungs	Rate of condemnation (%)
Pneumonia	41	41
Hydatid cysts	19	19
Emphysema	11	11
Congestion	9	9
Fibrosis	6	6
Aspiration of blood	3	3
Oedema	3	3
Adhesions	3	3
Abscesses	2	2
Calcification	2	2
Necrotic foci	1	1
	100	100

Pneumonia: Pneumonia was found to be the cause of the highest percentage of lung condemnations (41%) as compared with other causes. All stages of pneumonia were seen including congestion, red hepatisation, grey hepatisation (Fig 1) and consolidation. Many lungs which showed grey hepatisation had multiple foci of consolidation with dark greyish colour and lobular appearance. The lesions were more pronounced in the apical lung lobes. Histopathological examination indicated evidence of bronchopneumonia (Fig 2), interstitial pneumonia (Fig 3) and necrotic pneumonia.

Hydatid cysts

Hydatidosis was found to be the second cause of lungs condemnation (Fig 4). The number of hydatid cysts varied from 1 to 10 and their sizes ranged from 2 to 20 centimetres, in a single lung. Some cysts were caseated.

Emphysema

Emphysematous camel lungs were enlarged in size, flabby, pale in colour and easily compressed by finger.

Fibrosis

Fibrosis was seen as excessive fibrous connective tissue in the lung.

Aspiration of blood

Raised plaques were found scattered all over the lung due to aspirated blood.

Oedema

Oedematous lungs were enlarged, increased in weight and fluid was released when the surface of the lung was cut.

Abscesses

Abscesses were found in the form of nodules with various sizes. They had raised surfaces and were greyish white in colour. Upon cutting, they yielded inspissated pus which was sometimes gritty in nature.

Calcification

White to grey calcium deposits were found in some lungs. The deposits were irregularly round with a gritty feeling when sliced with a knife.

Other lesions

These included congestion of some lungs which looked bright red in colour with areas of atelectasis. These areas appeared dull red, hard and depressed at the lung surface. Other lungs showed areas of adhesions on their surface.

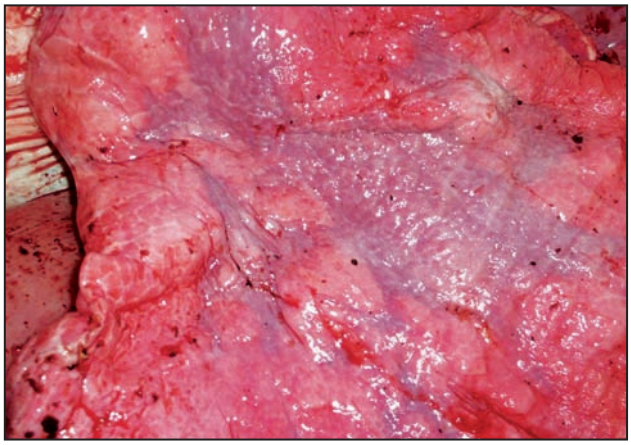

Fig 1. Grey hepatisation indicating pneumonia.

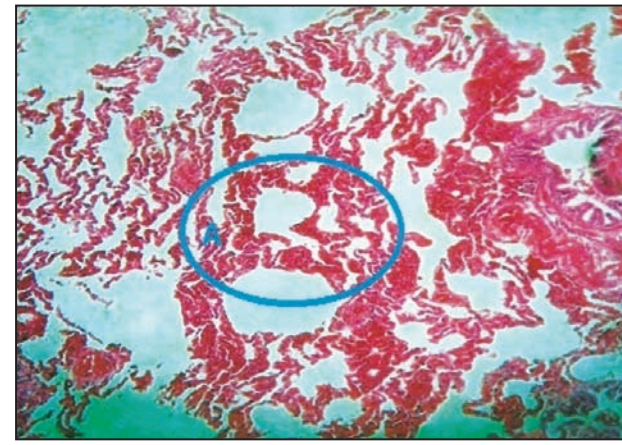

Fig 2. Bronchopneumonia (mononuclear cells and neutrophils infiltration inside the bronchi and hyperplasia of the bronchiolar epithelium, H&E X100).

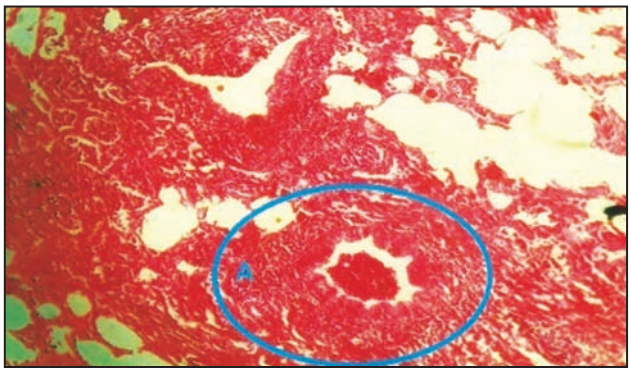

Fig 3. Interstitial pneumonia (thickening of the interstitial tissue and infiltration of inflammatory cells between the alveoli, H&E X100).

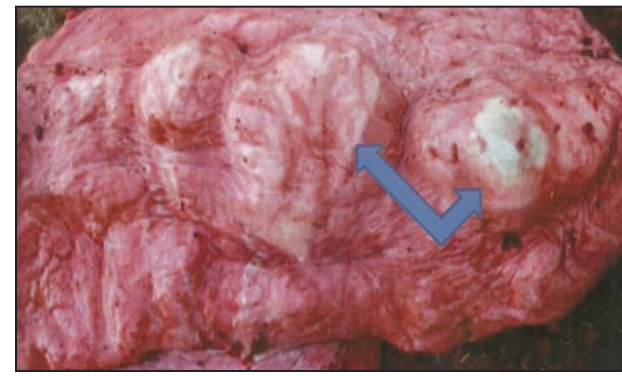

Fig 4. Hydatid cyst.

Haematological findings

Erythrocytic indices

Table 2 shows the erythrocytic indices of camels without pulmonary lesions compared with those with pulmonary lesions. The differences between the values in the two groups were statistically insignificant.

Table 2. Erythrocytic indices of camels without and with lung lesions.

Parameter	Camels without lung lesions (n = 14)	Camels with lung lesions (n = 23)
RBC ($X10^6$/µ)	3.8±09	4.6±0.5
Haemolglobin (g/dL)	9.8±0.5	10.5±0.5
PCV %	31.4±2.3	31.5±1.2
MCV (fl)	55.5±4.9	55.6±11.0
MCH (pg)	32.2±10.0	28.4±3.8
MCHC/(g/dl)	33.2±0.1	33.3±0.08

Leucocytic indices:

The leucocytic indices of camels without and with pulmonary lesions are shown in Table 3. The total leucocytes counts were significantly increased ($P< 0.05$) in camels with lung lesions. The counts of neutrophils, eosinophils, basophils, lymphocytes and monocytes were, however, not statistically different in the two groups, but relative increases in the neutrophils and lymphocytes in camels with lung lesions were found.

Table 3. Leucocytic indices of camels without and with lung lesions.

Parameter	Camels without lung lesions (n = 14)	Camels with lung lesions (n = 23)
WBC ($X10^3$/µl)	7.3±0.5	9.3*±0.6
Neutrophils ($X10^3$)	3.39±0.5	4.48±1.2
Eosinophils ($X10^3$)	0.33±1.3	0.31±0.4
Basophils ($X10^3$)	0.03±0.04	0.006±0.05
Lymphocytes ($X10^3$)	3.32±0.8	3.93±1.3
Monocytes ($X10^3$)	0.42±1.2	0.52±0.5

*P<0.05 was considered statistically significant

Discussion

The results showed that pneumonia was the most important cause of condemnation of slaughtered

camels lungs in Tamboul Abattoir at Central Sudan (41%). Comparable findings indicating pneumonia as an important cause of condemnation of slaughtered camel lungs were previously reported by Tigani *et al* (2007) and Nasar Eldien (2010). The former and latter workers reported condemnation rates of lungs of camels slaughtered in Nyala (Western Sudan) and Tamboul (Central Sudan) as 32% and 57%. Likewise. Zubair *et al* (2004) found a condemnation rate of 45% of slaughtered camel lungs due to pneumonia in Pakistan and Abubakar *et al* (2011) reported a condemnation rate of 46.4% in Nigeria.

The second important cause of lung condemnation in the present investigation was hydatidosis (19%). In a similar survey in Ethiopia, Muskin and Moti (2011) reported a condemnation rate of 22.6% of slaughtered camel lungs. Tigani *et al* (2007) and Nasar Eldien (2010) reported higher condemnation rates of camel lungs infected with hydatidosis in Western and Central Sudan. These were 40.4% and 31.2% consecutively. Likewise, Nourani and Rohani (2009) found that hydatidosis was the most prevalent pulmonary infection in Iran (51%). Etana Debela *et al* (2015) reported that the highest proportion of hydatid cysts were recorded in the lungs (56%) followed by the liver (33.9%), the spleen (7.3%) and the kidneys (2.8%).

Mixed infections of hydatidosis and pneumonia were found in some condemned lungs in the present investigation indicating the possible role of hydatidosis as a predisposing factor for secondary infections which lead to pneumonia.

Congestion and fibrosis were the causes of 9 and 6% of condemned camel lungs in Tamboul Abattoir as shown by the results of the present investigation. Similar rates (9.2 and 6.0%) were previously reported by Nasar Eldien (2010) in an investigation carried out in the same abattoir.

The condemnation rate due to fibrosis was found to be slightly lower (4.4%) in Western Sudan by Tigani *et al* (2007).

Aspiration of blood as a cause of camel lungs condemnation was found to be 3% in the present investigation. A similar rate was reported by Tigani *et al* (2007) in Western Sudan. The rate reported by Nasar Eldien (2010) in Central Sudan was about 5 times higher (14.4%). This may be due to differences in slaughtering methods.

The percentages of adhesions and abscesses as causes of condemnation of slaughtered camel lungs in the present investigation were similar to those reported by Tigani *et al* (2007) in Western Sudan. They were, however, higher than the percentages previously reported by Nasar Eldien (2010) in Tamboul Abattoir (0.8%).

Pulmonary lesions had little effect on the erythrocytic indices of affected camels as the differences between these indices in the affected and normal camels were statistically insignificant. These findings were comparable to those reported by Abubakar *et al* (2011) in a similar investigation in Nigeria.

The values of these erythrocytic indices were found comparable to those reported by Abdelgadir *et al* (1979) and Barakat *et al* (2007) in the Sudan. They were also comparable with those reported by Abubakar *et al* (2011) in Nigeria and Farooq *et al* (2011) in Pakistan.

The results, however, showed a statistically significant increase in the total leucocytes count of camels with pulmonary lesions when compared with those which had no pulmonary lesions. It is likely that this increase is associated with the inflammatory process initiated by the various etiological factors which lead to the pulmonary lesions in the affected camels. Nigerian camels with pulmonary lesions also showed an increase in the total leucocytes counts when compared with the counts in normal camels (Abubakar *et al* 2011).

The relative increase in neutrophil counts in the camels with pulmonary lesions in the present investigation may be attributed to purulent inflammations in the camels which had abscesses. The slight increase in the lymphocyte counts, on the other hand, may be due to chronic inflammatory processes in some of the camels with pulmonary lesions. Lymphocytes, together with macrophages, are known to comprise the main components of the inflammatory cells which prevail during chronic inflammations, particularly those initiated by bacterial infections (Johnes *et al*, 1997; Muna *et al*, 2017).

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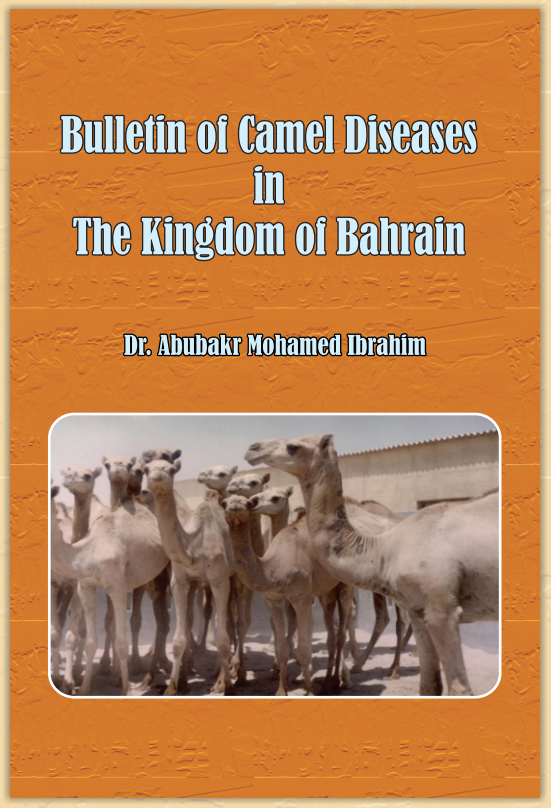
Bulletin of Camel Diseases in The Kingdom of Bahrain

This is a unique book which contains chapters on infectious and non-infectious diseases. The chapter on infectious diseases contains six sections. The section of bacterial diseases is subclassified as corynebacterium abscesses, paratuberculosis, hepatic necrobacillosis, mastitis, *Streptococcus zooepidemicus,* bacterial Infection in young camels, uterine Infection, infection of the vagina and vulva and other disorders. The section of protozoal diseases has narrations on trypanosomiasis, anaplasmosis and babesiosis. The section on parasitic infections is composed of gastrointestinal parasites in young camels, echinococcosis and mange. The section of mycotic diseases contains phycomycosis and ringworm. The section of viral diseases contains subsections on camel pox and contagious ecthyma. Edema Disease is described in miscellaneous section. The chapter on noninfectious diseases has three sections. Other section on poisoning describes pyrethroid, nitrate and toxic jaundice. The section describes zinc deficiency. The miscellaneous section describes foreign bodies, sand colic, bloat, caecal impaction, hydrocephalus, corneal opacity and osteochondroma.

About the Author

Dr. Abubakr Mohamed Ibrahim is a Veterinary Pathologist and worked for a long period as head of Royal Court Veterinary Laboratory. Kingdom of Bahrain which led to genesis of this publication out of his rich experience in diagnosing camel diseases in the Kingdom of Bahrain. This would be counted as his significant contribution and future researchers will find it easy to understand the pattern of camel diseases in this part of the world. Dr. Abubakr had majority of his publications based on camel diseases of Bahrain. Thus publication of this book would prove an important reference book for the camel practitioners and researchers.



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LUMPY JAW AND WOODEN TONGUE IN AN ADULT DROMEDARY CAMEL: CASE REPORT

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Lumpy jaw is caused by different bacterial species and therefore it is important to identify the pathogen. Actinomycosis caused by *Actinomyces* (A.) *bovis,* a Gram-positive, anaerobic, organism causes the classical lumpy jaw in cattle, a suppurative alveolar periostitis often involving the third and fourth molars. In chronic cases the osteomyelitis is surrounded by periosteal new bone and fibrous tissues producing cavities in the bones; the bone is "eaten away". The pathogen penetrates the wound of the oral mucosa caused for example by a wire, coarse hay stems or sticks. *A. bovis* is part of the normal oral flora of ruminants. The lesions most frequently involve the mandible and the molars, the maxillae and other bones of the head.

Actinobacillosis with 22 different species of which few are important pathogens like *Actinobacillus (A.) pleuropneumoniae, A. suis, A. equuli* and *A. lignieresii* are responsible for different diseases. *A. lignieresii* occurs in sheep, horses, dogs and ruminants. It is a Gram-negative anaerobic coccobacillus, which causes tumorous abscesses mainly of the tongue in bovines. A hard granulomatous mass develops in the tongue causing anorexia and excess salivation. The tongue is hard and wooden and it is therefore named "wooden or timber" tongue (Markey *et al,* 2013). The disease resembles lumpy jaw disease as often the jaws are also involved. Wooden tongue disease is caused by wounds in the mouth mucosa by stemmy feed or pastures covered with penetrating plant awns like thistles.

Fig 2. Lumpy jaw cut open displaying white necrotic material

Fig 1. Lumpy jaw presented as an orange-size granuloma of the right mandible

Fig 3. "Wooden tongue"

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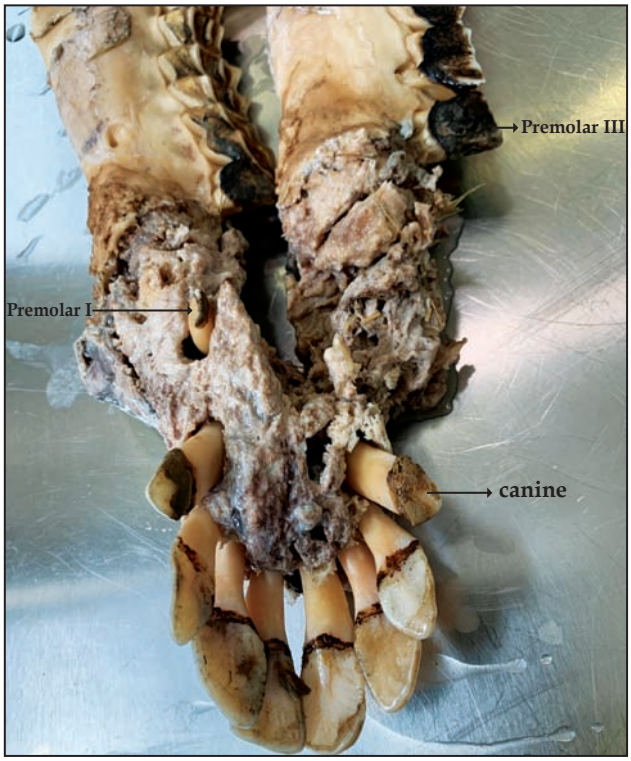

Fig 4. Large parts of the Pars incisiva of Corpus mandibulae were eaten away between canine and premolar III of both mandibles

A pregnant dromedary camel in poor condition was sent to CVRL where a necropsy was performed. An orange-size hard granuloma was observed on the right lower jaw. The skin was intact (Fig 1). When opened dry necrotic mass protruded (Fig 2). The tongue lost its lining displaying necrotic material inside the tongue. When touched, it was hard like wood, therefore named wooden tongue (Fig 3). Both mandibles were macerated and it was observed that both lower jaws were affected. Large parts of the *Pars incisiva* of *Corpus mandibulae* were eaten away between canine and premolar III (Fig 4). Histology of the jaw lesions displayed a granuloma containing fungal elements surrounded by pyogranulative inflammation (Fig 5).

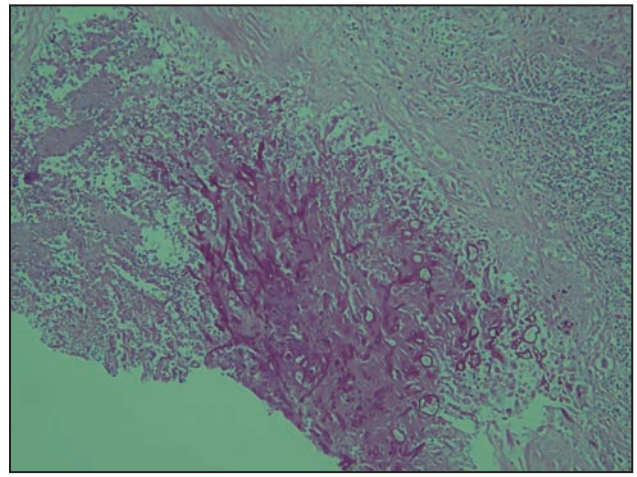

Fig 5. Histology of lumpy jaw in PAS stain showing fungal filaments and abscess formation

From the jaw's abscess, *Fusobacterium mortiferum, Streptococcus sanguinis, Pediococcus acidilactici, Aspergillus ochraceus* and *Trichosporon mucoides* were isolated. It is not clear which bacterial or fungal species had caused the lumpy jaw, but most probably all together. Actinomyces or Actinobacillus bacteria were not isolated. Lumpy jaw in camels is rare and has very seldom been reported. Fowler (2010) reported Actinomycosis in a llama and Wernery *et al* (2014) in dromedaries. However, according to Manefield and Tinson (1996) in contrast to other species Actinomycosis is almost never seen in dromedaries.

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USE OF SALICYLIC ACID, ZINC SULPHATE, FENVALERATE AND SULPHUR WITH PETROLEUM JELLY AS BASE FOR TREATMENT OF SARCOPTIC MANGE IN A CAMEL

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ABSTRACT

The present study describes use of ointment containing salicylic acid, zinc sulphate, fenvalerate and sulphur with petroleum jelly as base for successful and effective treatment of severe mange in an adult male dromedary camel. The affected camel had severe lesions on neck, axilla, inner surface of thigh, inguinal region, perineal region, root of tail, flank region, head, lip necckand brisket region. The case did not respond to Ivermectin treatment. Itching disappeared 7 days after application of acaricidal ointment and fresh shiny skin appeared at 7 day after second application.

Key words: Camels, petroleum jelly, salicylic acid, sarcoptic mange

Sarcoptic mange in camels caused by *Sarcoptes scabiei var cameli* is considered to be one of the most serious, contagious, zoonotic (Basu *et al,* 1996) and debilitating disease (Singh *et al,* 1996).

Synthetic pyrethroid like fenvelarate used topically was reported to be successful in treating the sarcoptic mange (Bowman, 1999). The systemic use of ivermectin, though costly but having distinct advantage over topical therapy with acaricides gained preference as treatment of sarcoptic mange (Abu-Samra, 1999) but its efficacy has been in question after documentation of *in vivo* and *in vitro* ivermectin resistance in *Sarcoptes scabei* (Currie *et al,* 2004). The importance of concomitant keratolytic therapy and topical scabicides to kill mites in the thick crusts has been emphasised (Chosidow, 2000). In order to utilise antiinflammatory properties of salicylic acid (Weirich *et al,* 1976) and zinc sulphate (Gupta *et al,* 2014) together with acaricidal effect of fenvalerate (Bowman, 1999) and sulphur (Guichou *et al,* 2002), a new formulation was tried in petroleum jelly to treat the the mange in a male camel and is reported here.

Materials and Methods

A male camel (breeding stud) aged 10 yrs belonging to the herd of ICAR- National Research Centre on Camel, Bikaner was presented with mange. The camel was treated with ivermectin previously alongwith other camels of the herd. But it did not respond to the treatment. The severe lesions were found on neck, in the axillae, inner surface of thigh, inguinal region, preputial sheath, perineal region, root of the tail, entire flank region, head lips neck and brisket region. Keratinisation, thickening, corrugation and wrinkling of the skin, exudation, fissured skin and scab formation were also noticed. Alopecia was severe on legs, head, neck, trunk, abdomen, flank, perineum (Fig 1). Histopathological study of skin of mange affcated camels is well studied (Mathur *et al,* 2005).

The affected camel lacked proper rest because of intense pruritis and spent much time in biting, scratching and rubbing against objects and was totally restless. As a result substantial loss of libido was observed during the breeding season.

An ointment with following formulation was prepared,

1. Salicylic acid– 3%
2. Zinc sulphate– 3%
3. Fenvalerate dust (0.04%)– 6%
4. Sulphur– 6%
5. Petroleum jelly (I.P.)– Base

The ingredients were homogenously mixed in a bucket. The camel was restrained in sitting position and ointment was applied and spread on the affected skin area with the help of muslin cloth. In

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the present case of severe mange where whole body was involved about 2 kg ointment was sufficient to cover the affected portion in an adult camel. A second application of ointment was made after 15 days.

Results and Discussion

The ointment was found to be effective in treating the mange in dromedary camel. The recovery of skin and return to normalcy of "skin coat" was rapid and is characteristically shown in fig 2. There was marked clinical improvement in appearance with reference to healing of skin lesions and texture. The crusts and wrinkling disappeared and there was substantial reduction in skin folds. The disappearance of clinical signs of itching was noticed by 6-7 days. Appearance of fresh shiny skin was observed at 7 days after second application.

Salicylic acid is a lipid soluble, miscible with epidermal lipids and considered as a keratolytic, desmolytic and peeling agent because of its ability to disrupt cellular junctions and breaking or lysing intercellular keratin filaments (Arif, 2015) therefore it helps in penetration of drugs deep into the skin. Salicylic acid possess anti-inflammatory properties (Weirich *et al,* 1976). Zinc sulphate has been found useful in several dermatological ailments owing to its anti-inflammatory properties (Gupta *et al,* 2014). Fenvelarate is the 3rd generation synthetic pyrethroids having acaricidal effect (Bowman, 1999). For more than one hundred years Sulphur is being used as fungicide and acaricide (Guichou *et al,* 2002).

The male mite does not burrow into the skin and remains on the surface but the female mite cuts skin rapidly with its mouth parts and claws becoming completely embedded in two and a half minutes. She remains within the horny layer of the skin forming tortuous tunnels for about two months laying eggs. The tunneling and the secretory and excretory products produced by the mites produce an itching sensation in the infested camel (Roberts and Janovy, 2000). It was suggested that ivermectin is not ovicidal because of inadequate penetration of the thick eggshell (Currie *et al,* 2004) and this may be the reason for its ineffectiveness in treating the mange completely in some animals. Petroleum Jelly is a purified mixture of semi-solid, saturated hydrocarbons, mainly of paraffinic nature, obtained from petroleum. The application of acaricides in a petroleum jelly base has benefit of spot treatment of acaricides to control ticks at specific body sites such as the perineum region. The acaricides in this spot on treatment can act for longer time. It has superiority over vegetable oils because it helps in healing of skin crevices. Secondly, it is retained longer than vegetable oil on the affected part and hence is more effective vehicle for the drugs. The application of acaricide by spraying tends to be uneconomical as excess acaricide solution, which drips off animals, is not recovered. This loss of medicament when applied through sprays is reduced with petroleum jelly as base. However, a herbal formulation has also been used successfully for treating mange in camels (Pathak *et al,* 1996).

Salicylic acid together with zice sulphate, sulphur, fenvalerate and petroleum jelly commonly used in human beings have not been reported for the treatment of skin affection in camel. Therefore, in the present study these were used as an ointment which was found to be effective in treating the skin affection in severe case of mange in an adult dromedary male camel.

Fig 1. Extensive mange infestation in a male camel.

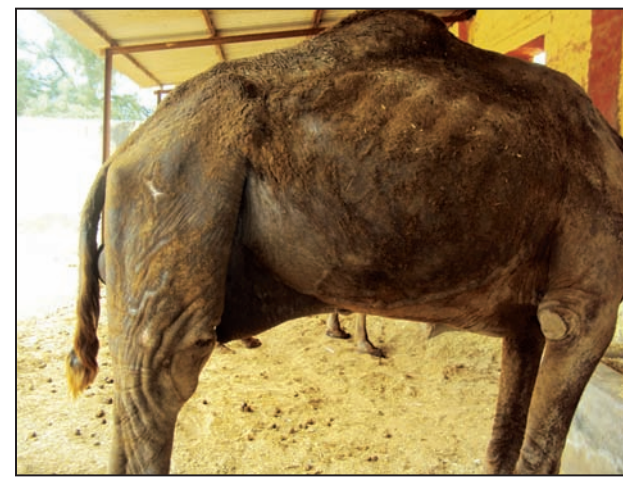

Fig 2. The effect of ointment with petroleum jelly as base in mange infested camel.

Acknowledgement

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SOME PRODUCTIVE CHARACTERISTICS OF SHAMI CAMELS (*Camelus dromedarius*)

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ABSTRACT

This study was carried out on Shami camels at Dier-Alhajar station in Syria to evaluate the effect of some environmental factors on milk parameters daily milk yield (kg), milk fat%, milk protein%, lactose%, non-fat solids%, and total solids and body weight at birth and at 6 months intervals until 4 year-old (6, 12, 18, 24, 30, 36, 42 and at 48 months of age). A total of 1968 records of milk and 2018 records of body weight were used. Data were analysed using 2 fixed linear models by SAS (2012). The studied milk and bodyweight traits were estimated. All studied milk traits were affected by year of production and parity except lactose% was not affected by year of production. Daily milk yield was affected by the interaction between the year of production and the time of milking. All weight traits were not affected by calf sex, birth year, or their interactions except body weight at birth and weight at 18 months of age which were affected by year of production and calf sex, respectively. This study concluded that year of production and parity might influence some milk traits and also birth year might influence body weight at birth of Shami camel.

Key words: Body weights, camels milk traits, shami camel

Camels have unique characteristics that make them more suitable to raise in arid land than other animals. They are also less prone to environmental damage compared to other animals under the pastoral system (Raziq *et al,* 2010). The camel population in Syria is estimated at 46140 animals (FAO, 2018). The greater part of camels is raised under the pastoral system. There are 2 stations for rearing camels in the countryside Damascus and Hama cities. Camel milk yield varies with management and genetic factors (Kariuji, 1997). Konuspayeva *et al* (2009) reviewed that camel milk components differed depending on geographical region, breed, nutritional condition, seasonal and also physiological condition. Milk production in camel is low in amount but with a large variation, however, improvement is possible by management and selection (Hermas, 2002). Four factors should be considered for selecting dairy camel *viz* general appearance, milk production, body capacity and mammary system (Shareha, 2004). In addition, camels that are producing great quantities of milk and meat are due to good management and selection across generations by breeders. Identifying environmental factors that affect milk yield, milk composition and calves' weight is important for the best management of camel herds. The objective of this study was to evaluate the effect of some environmental factors on daily milk yield, milk composition and calves weight from birth to 48 months of age under Syrian conditions.

Materials and Methods

Camels were kept, under open shades and housed in cement barns during the night and bad weather to protect them from rain time in winter and spring season. Camels were fed on Atriplex Salty or *Solsola* rigida plant growth for 8 hours a day. Barley was given as additional ration by 1.5-2.5 kg for females according to production and 1.5 kg for males (kg/head/day). Also, camels were given a bran and cotton meal when available. Water was provided *ad-lib*. Natural mating was allowed with males from the same station. Males were assigned to female according to reproductive efficiency. Females were mated from November to March so calving expected to take place during the months of February through May. The young camels were weaned at 13-15 months, depends on their weight. Both females and males were allowed to mate when their ages reach between 4-5 years according to their weights.

The data were collected from Dier-Alhajar station for Shami camels in the countryside Damascus. Syria. Milk samples were taken 3 times a day during the lactation period. Immediately after

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milking they were weighed using scale and were analysed using the milk scan apparatus. Daily milk yield (kg) and milk fat, milk protein, lactose, non-fat solids and total solids (%) were recorded. Camels calves were weighed (kg) immediately after birth and at 6 months intervals until 4 years old using a fixed scale in the station. Data covering six and three years from 2002 to 2007 and from 2004 to 2006 included 1968 and 218 records for milk and weight traits, respectively of Shami camels.

Data were statistically analysed using SAS (2012) program according to General Linear Models, GLM that were fitted into two models. Duncan multiple range test was used to detect the differences among means of effects (Duncan, 1955).

Results and Discussion

The least-squares means for daily milk yield, kg, milk fat%, milk protein%, lactose%, non-fat solids% and total solids%, are included in table 1. The daily milk yield (DMY) was close to the values of 3.2, 3.8 and 3.96 kg which were determined by Hermas (2002), Zeleke (2008) and Hadef *et al* (2018), respectively. While DMY was higher than 2.4, 2.0 and 1.02-2.0 which were determined by Bakheit *et al* (2004), Moslah *et al* (2005) and Chimsa *et al* (2014), respectively. But DMY was below the values of 16.6, 13.0 and 9.62 kg determined by Ismail and Al-Mutairi (1990), Hanif Khan (1996) and Musa *et al* (2006). The milk fat% (MF%) was close to Amer *et al* (2005), El-tahir *et al* (2014) and Hadef *et al* (2018), which were 3.38%, 3.63% and 3.72%, respectively. The current MF% was less than those estimated by Riek and Gerken (2006), Park and Haenlein (2006) and Gorakh and Pathak (2010), which were 4.70%, 4.9% and 5.5%, respectively. While MF% was higher than 2.47%, 2.72%, 2.92% which were reported by Zeleke (2008), Ismaili *et al* (2019) and Kaskous (2019). The milk protein% (MP%) was similar to (3.87)%, (3.60)% and (3.37)%, which were estimated by Gorakh and

Table 1. Least square means±standard errors (LSM±SE) and variance analysis of milk traits of Shami camel.

Factors	Least Squares Means±Standard Errors					
	Daily milk yield, kg	**Fat%**	**Protein%**	**Lactose%**	**Non-fat solids%**	**Total solids%**
μ	3.52±0.15	3.75±0.11	3.14±0.08	4.74±0.07	8.90±0.10	12.64±0.17
Year of Production (YP)	**	**	**	**Ns**	**	**
2004	4.41±0.17^{a}	4.68±0.12^{a}	4.28±0.09^{a}	4.88±0.07	10.20±0.12^{a}	14.87±0.19^{a}
2005	3.59±0.07^{a}	3.78±0.05^{b}	3.01±0.03^{b}	4.75±0.03	8.77±0.04^{b}	12.54±0.07^{b}
2006	2.57±0.40^{b}	2.78±0.28^{c}	2.15±0.21^{c}	4.58±0.17	7.72±0.27^{c}	10.50±0.44^{c}
Time of Milking (TM)	**Ns**	**Ns**	**Ns**	**Ns**	**Ns**	**Ns**
Morning	3.52±0.25	3.72±0.18	3.15±0.14	4.76±0.12	8.92±0.18	12.64±0.27
Afternoon	3.35±0.24	3.77±0.17	3.16±0.13	4.69±0.10	8.88±0.17	12.62±0.29
Evening	3.70±0.26	3.76±0.16	3.12±0.15	4.77±0.11	8.89±0.19	12.63±0.28
Parity (PR)	**	**	**	**	**	**
1st	1.53±0.17^{e}	3.89±0.12^{ab}	2.76±0.09^{de}	4.86±0.08^{b}	8.59±0.12^{c}	12.48±0.19b^{c}
2nd	1.32±0.31^{e}	3.46±0.22^{c}	4.05±0.16^{a}	5.33±0.13^{a}	10.40±0.21^{a}	13.86±0.34^{a}
3rd	5.21±0.15^{ab}	3.56±0.10^{bc}	2.81±0.08^{de}	4.85±0.06^{b}	8.68±0.10^{c}	12.22±0.16^{bcd}
4th	3.56±0.17^{c}	3.41±0.12^{c}	2.60±0.09^{e}	4.81±0.07^{b}	8.39±0.12^{c}	11.80±0.19^{d}
5th	5.66±0.18^{a}	3.69±0.13^{bc}	3.03±0.10^{cd}	5.20±0.08^{a}	9.26±0.12^{b}	12.94±0.20^{b}
6th	2.57±0.27^{d}	4.10±0.19^{a}	3.59±0.14^{b}	3.52±0.12^{d}	8.15±0.18^{d}	12.23±0.29^{cd}
7th	4.80±0.22^{b}	4.13±0.16^{a}	3.18±0.12^{c}	4.61±0.10^{c}	8.81±0.15^{c}	12.92±0.24^{b}
YP × TM	**	**Ns**	**Ns**	**Ns**	**Ns**	**Ns**
PR × TM	**Ns**	**Ns**	**Ns**	**Ns**	**Ns**	**Ns**
MSE	3.247	1.2655	0.879	0.605	1.534	3.952
CV%	40.74	33.89	33.39	15.99	14.27	16.03

* : p<0.05. ** : p<0.01. Ns : insignificant effect. μ : Overall mean. MSE: Mean Square Error.

$^{abc...}$Means in the same column without common letter are different at p<0.05. CV% : Coefficient of Variation.

Pathak (2010), El-tahir *et al* (2014) and Hadef *et al* (2018), respectively. While MP% was lower than the estimates of Urazakov and Bainazarov (1974) and Riek and Gerken (2006) which were 4.93% and 4.23%, respectively. The MP% was higher than 2.85%, 2.55% and 2.28% which was determined by Zeleke (2008), Ismaili *et al* (2019) and Kaskous (2019), respectively. The lactose% (Lac%) was similar to estimates of 4.74%, 4.13%, 4.37%, that were determined by El-Tahir *et al* (2014), Hadef *et al* (2018) and Ismaili *et al* (2019), respectively. While Lac% was lower than 5.93% and 5.10% which were determined by Riek and Gerken (2006) and Park and Haenlein (2006), respectively. But Lac% was higher than 2.90% and 3.91% that were determined by Brezovecki *et al* (2015) and Kaskus (2019), respectively. Estimation of non-fat solids% (NFS%) was around 9.09% and 8.84%, that was also found by Amer *et al* (2005) and El-Tahir *et al* (2014), respectively. While NFS% was lower than that 10.95%, 14.31%, 10.44% which were reported by Elamin and Wilcox (1992), Zhang *et al* (2005) and Zeleke (2008), respectively. A total solids% (TS%) estimate was close to 12.48%, which was reported by Amer *et al* (2005). While TS% was higher than 9.99%, which was stated by Hadef *et al* (2018). But TS% was lower than 14.40% and 14.68%, which were reported by Park and Haenlein (2006) and Gorakh and Pathak (2010), respectively. These differences in estimates may be due to dissimilar herd management, rearing systems and genotype.

Estimates of DMY, MF%, MP%, NFS% and TS% were lowered significantly (P<0.01) from 2004 to 2006 while the reduction in Lac% was not significant (P>0.05) as shown in table 1. The effect of year of production (YP) on DMY agrees with Aslam *et al* (2002). The effect of YP on MF% and MP% was compatible and was not for Lac% with Ismaili *et al* (2019). The difference between productive years may be due to differences in management, nutritional and climatic conditions. The parity (PR) effect was significant (P<0.01) on all studied milk traits. Estimating the PR showed an unclear trend in all milk traits (Table 1) which may be due to weight and age differences among females. The effect of PR for DMY was consistent with Zeleke (2008), Sallal *et al* (2010) and Chimsa *et al* (2014) but disagreed with Aslam *et al* (2002). The effect of time of milking (TM) was non-significant (P>0.05) on the studied milk traits, where there are no similar research reviews of camels. The interactions of (YP by TM) and (PR by TM) were non-significant on studied milk traits except for DMY, which was significant (P<0.01). These show that the differences in TM within YP and also TM within PR were homogeneous for most studied milk traits except DMY which was heterogeneous.

The least-square mean weights for birth weight (WtB), weight at 6 months age (Wt6), weight at 1st a year age (Wt12), weight at 1.5th years age (Wt18), weight at 2nd years age (Wt24), weight at 2.5th years age (Wt30), weight at 3rd years age (Wt36), weight at 3.5th years age (Wt42) and weight at 4th years age (Wt48) were estimated in table 2. Estimated WtB was lower than 40.6, 37.3 and 37.45-37.60 kg, which was reported by Bissa *et al* (1998), Sallal *et al* (2010) and Bakheit *et al* (2017), respectively. The estimated WtB for males and females were also lower than those determined by Saoud *et al* (1988), Hermas *et al* (1990) and Bakheit *et al* (2017) which were 36.1 and 35.0; 35.9 and 34.01 and 38.85 and 36.20 kg, respectively. The estimated Wt6 was similar to the estimates of 96.42-123.40 kg, that decided by Bakheit *et al* (2017). But Wt6 was lower than the estimates of 170.6, 150.0 and 150.8 kg which were recorded by Umesh (1996), Bissa *et al* (1998) and Sallal *et al* (2010), respectively. The calves' weight estimate Wt12 was within 159.70-221.04 kg which was reported by Bakheit *et al* (2017). On the other hand, Wt12 was lower than the estimate of 211.02, 247.37 and 295.89 which were reported by Bissa *et al* (1998), Sallal *et al* (2010) and Salehi *et al* (2013), respectively. The calves Wt18 was within the range reported by Bakheit *et al* (2017) which were 208.62-326.26 kg. Also, the calves Wt24 was lower than of estimates of 290.23 and 356.27 kg which were reported by Bissa *et al* (1998) and Salehi *et al* (2013). Estimate of calves Wt30, Wt36, Wt42 and W48 were lower than 328.44, 373.23, 435.13, 486.95 kg, reported by Bissa *et al* (1998).

Table 2 showed that most of the studied weight traits were non-significantly affected by birth year (BY) except WtB. The difference in birth weight may be due to the weights of females. The effect of calves sex (S) on weight was non-significant (P>0.05) in most studied weight traits except Wt18. This difference might be due to the fact that male calves were more nervous for females at Wt18. The effect of S on WtB was similar to the results of Sallal *et al* (2010), while El-bashir *et al* (2012) found significant on WtB. The effect of S on Wt6 was consistent with the results of Sallal *et al* (2010), who determined that there were no significant sex differences Wt6. The effect of S on Wt12 was agreed with Sallal *et al* (2010) and Salehi *et al* (2013), where there was non-significant effect of S on Wt12. The difference between sex was determined as a significant effect on Wt24 according to Salehi

Table 2. Least square means±standard errors (LSM±SE) and variance analysis of weights traits at different ages/ kg of Shami camel.

Factors	Least Squares Means±Standard Errors								
	WtB	**Wt6**	**Wt12**	**Wt18**	**Wt24**	**Wt30**	**Wt36**	**Wt42**	**Wt48**
μ	32.39± 0.39	121.97± 4.10	178.53± 6.94	230.72± 9.76	259.76± 9.97	12.53± 309.91	340.18± 13.12	354.48± 8.39	426.31± 13.05
BY	*	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**
2002	29.95± 0.66^{b}	120.06± 9.41	187.55± 17.74	208.33± 26.57	241.80± 26.75	29.96± 319.15	361.28± 26.63	361.33± 20.60	435.03± 26.97
2003	32.49± 0.79^{ab}	141.29± 8.16	197.14± 14.17	248.88± 19.15	282.07± 19.65	328.05± 26.42	324.06± 41.84	370.78± 23.78	433.85± 38.74
2004	32.58± 0.84^{ab}	125.98± 7.65	186.36± 12.80	241.25± 17.31	277.63± 17.43	297.00± 25.79	356.80± 25.85	348.47± 17.64	462.17± 24.62
2005	32.69± 0.94^{a}	119.83± 10.32	200.25± 16.92	251.63± 25.57	290.68± 26.75	374.33± 29.96	365.10± 33.21	361.25± 22.56	420.75± 36.94
2006	33.50± 1.20^{a}	119.84± 12.33	161.02± 20.22	251.50± 27.33	253.08± 28.20	274.33± 36.47	325.33± 32.41	340.83± 20.60	404.17± 33.72
2007	33.12± 1.13^{a}	104.83± 11.54	138.88± 18.91	182.71± 25.57	213.29± 25.74	266.46± 34.11	308.50± 30.31	344.21± 17.23	401.88± 28.21
Sex	**NS**	**NS**	**NS**	*	**NS**	**NS**	**NS**	**NS**	**NS**
Male	32.93± 0.51	114.90± 5.37	173.57± 9.36	206.71± 13.54^{b}	240.69± 13.47	291.43± 18.73	324.29± 21.60	350.01± 12.85	425.11± 19.92
Female	31.84± 0.59	129.04± 6.20	183.50± 10.24	255.16± 14..05^{a}	278.83± 14.71	328.40± 16.65	356.06± 14.90	358.94± 10.78	427.51± 16.87
YP × Sex	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**
MSE	26.81	1277.35	3434.12	6274.93	6361.62	7979.85	6301.43	2036.53	5457.10
CV%	16.19	28.77	32.00	33.68	30.41	28.13	22.80	12.63	17.15

WtB: Birth weight, Wt6: Weight at 6 month old, Wt12: Yearling weight, Wt18: Weight at 1.5 years old, Wt24: Weight at 2 years old, Wt30: Weight at 2.5 years old, Wt36: Weight at 3 years old, Wt42: Weight at 3.5 years old, Wt48: Weight at 4 years old. * : p<0.05. ** : p<0.01. ns: insignificant effect. μ: Overall mean. MSE: Mean Square Error. BY : birth year. Sex : Calf Sex. $^{abc...}$Means in the same column without common letter are different at p<0.05.

et al (2013). The interaction effect (BY by S) was insignificant on all studied weights. These confirm that the differences between sex within birth years were homogeneous but according to El-bashir *et al* (2012) a significant effect of sex by age on WtB was seen.

Conclusions

The year of production and parity affected significantly the daily milk yield, milk fat%, milk protein%, lactose%, non-fat solid% and total solids%, hence, improving environmental conditions could maximise the benefits of such investment. Also, the birth year significantly affected birth weight. Male's weights were higher than females at the age of 18 months in the Shami camel.

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Agreement between the International Camel Organisation (ICO) and the King Abdulaziz Public Library

A joint agreement was signed by the Secretary-General of the International Camel Organisation (ICO), and the Director General of the King Abdulaziz Public Library, Bandar bin Abdullah Al-Mubarak, at the library's main headquarters in Riyadh with the aim of strengthening cooperation between the two parties and presenting a number of initiatives and projects to serve the spread of the camel's cultural heritage around the world.

This agreement seeks to achieve a number of goals, most notably supporting projects and initiatives to spread camel culture globally, organising international conferences and seminars, and building an interactive database on camels worldwide from In order to review this great legacy, build an international conceptual map on camels, hold a specialised exhibition on camel collections, support research and studies related to camels, pay attention to the culture and development of children, link children with the heritage of camels, worldwide, print an encyclopedia of camels through the ages and publish it on paper and electronically, and support initiatives Related to the translation of camel works from and into Arabic.

Inauguration of the Kumbhalgarh Camel Dairy

On 24 February 2020, the Kumbhalgarh Camel dairy that has been established by an organisation Lokhit Pashu-Palak Sansthan (LPPS) on its campus in Sadri (Rajasthan) with substantial support of LPP was formally inaugurated in a festive event with about 300 guests. Among them were the local MLA (Member of Legislative Assembly) and former Energy Minister Pushhpendra Singh, Sarita Kumari of Ghanerao, IFAD country director Rasha Omar, German benefactor Bettina Bock, and various other dignitaries, foreign guests, friends, Startup Oasis, mentors, women groups, etc. The MLA promised support for setting up a solar energy unit to make the dairy independent of the main power supply. The local hoteliers served a "Camel Conservation Lunch" featuring various novel menu items prepared with camel milk. A range of camel cheeses were also showcased. The celebration was preceded by a two-day training programme in hygienic milk collection for camel breeders from all over Rajasthan.

Saudi Arabia and India scraps camel festival

The festival for camels is stopped by the authorities in a central Saudi over organisers' failure to comply with precautions to curb the spread of the novel coronavirus. Governor of Qassim Prince Faisal bin Meshal bin Saud directed stopping the event in the governorate of Deria, a local official said. In view of non-compliance with applying precautionary measures and preventive protocols ordered by the leadership at festivals and public events and out of his concern for safety of citizens and residents, the governor of Qassim ordered the immediate halt to the festival. In recent weeks, Saudi Arabia has seen a significant drop in new coronavirus cases, allowing relaxed restrictions earlier imposed on different activities. Camels are a popular animal closely linked to heritage in the Arabian Peninsula country.

Department of Tourism, Government of Rajasthan, India has cancelled all the events till March 2021 and this includes International Camel Festival as well. Its happening after 26 years of regular hosting of this festival in Bikaner where large number of tourist come. This is a precautionary step to prevent Covid 19 infection in the city.

(Courtesy: Gulf News 4th November 2020 and Dainik Bhasker January 2nd 2021)

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MORPHOLOGICAL OBSERVATION OF THE *Wohlfahrtia magnifica* IN MONGOLIA PLATEAU

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ABSTRACT

The *Wohlfahrtia magnifica* living in Mongolia plateau was the only pathogen of Bactrian camel vaginal myiasis. In this paper, the head, thorax, abdomen, wing, leg and male and female identification of *W. magnifica* were observed and described by ultra depth imager. The *W. magnifica* was a kind of medium and large flies and covered with silver-gray pollen. The head had compound eyes, mouthparts, antenna, and a large amount of bristles. The dorsal of thorax was covered with silver-gray pollen and the mesothorax was developed, the legs, wings and halter were attached to thorax. The abdomen was divided into five abdominal segments, its dorsal had symmetrical black patch and the ventral was gray-black.The wings were composed of veins and cells. The leg was divided into propodium, mesopodium and metapodium. The structure of terminalia was difference between male and female, the female flies had serration end edges in the caudal end of abdominal segment and its terminalia was concave and the anal papilla was grayish white. To the male flies, its end edges of the caudal end of abdominal segment was smooth and its terminalia was convex, the dark yellow saccular process was shaped like a spoon swollen.This results would enrich people's understanding of the biological characteristics of *W. magnifica* in the Mongolia Plateau.

Key words: Morphology, ultra depth imager, *Wohlfahrtia magnifica*

Bactrian camels usually suffer from *W. magnifica* in summer and autumn. *W. magnifica* belongs to the order Diptera, family Sarcophagidae, genus *Wohlfahrtia Brauer*. The larvae of *W. magnifica* mature in the vaginal tissues of Bactrian camels, also known as the Mediterranean spiral worm, which is specialised parasite of warm blooded vertebrates (Tóth *et al,* 2006; Pirali *et al,* 2014; Wangchao, 2019). After mating, when the eggs develop until the 1st instar larvae in the female flies, the female fly looks for the host to deposit larvae, thus the healthy Bactrian camel becomes infected with vagina myiasis or the diseased camel to be re-infected (Xiwen *et al,* 2019). The 3rd instar larvae automatically falls off from its host, quickly burrows into the soil and metamorphosis into pupae, the pupae becomes imagoes in the dry season after about 20 days (Xiwen, 2018). Mongolia Plateau has little rain falls and hot in summer which is beneficial to the metamorphosis and development of *W. magnifica*, hence Bactrian camels are more predisposed.

There are few reports on morphological characteristics of *W. magnifica* but its occurrence in Mongolia Plateau has not been reported. In this study, we describe the morphological characteristics of *W. magnifica* in the Mongolia Plateau.

Materials and Methods

Sample collection

The wild *W. magnifica* were caught in the field and others were those hatched in laboratory.

Ultra depth imager sample preparation

The samples with structural integrity were placed in the room for observation and photographed using a KEYENCE-1000 Ultra Depth Imager.

Results

Observation on the dorsal, ventral and lateral of W. magnifica

Observation on the head of W. magnifica

The compound eyes of *W. magnifica* were red and yellow, naked, and the size of ommatidium was equal. There were 3 ocellus on the ocellar triangle

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and red coloured, the bristles on the ocellar triangle was divided into two pairs, the one pair of which was called ocellar bristles, leaned forward and bifurcated left and right, and the another pair of which was called postocellar bristles, leaned backward and parallel. The parafrontalia and parafacialia were covered by the silver gray pollen and under the parafrontalia had small black hairs, silver gray pollen of interfrontalia was slightly thin and small black hairs on its both sides. The face was composed of lunule, mid-facial plate, epistoma and a pair of facialia, the lower end of facialia has small black hair and developed vibrissae. The frontal bristles in the both side of interfrontalia were arranged in parallel and symmetrically, 8 bristles on each side. The vertex had prevertical bristles, inner vertical bristles, outer vertical bristles and post vertical bristles, the outer vertical bristles and post-compound eyes bristles were differentiated obviously. The gena was located under the compound eyes, parafacialia and intermedian triangle, the bottom colour of single gena was black and covered with black hair, it has a small amount of silver gray pollen. The mouthparts, namely proboscis was located under distal of head and brown coloured, a pair of maxillary palpi were located on the baisproboscis, brownish black and expansion of its distal end was not obviously. The peristoma bristles was one row and developed, the oral disc was composed of a pair of semicircular labellum. The antennae has three segments, the 1st and 2nd segments were short and small, the 3rd segment was significantly longer than the other two segments, the 2nd segment was dull yellow, the 3rd segment was brown, naked and with aristate, the antennae was also covered with a large number of bristle (Fig 2).

Observation on the lateral of the thorax and abdomen of W. magnifica

Bottom colour of the thorax was black, the notopleural bristles, front-mesopleura bristles and post- mesopleura bristles were developed, and there was clearance bristles between the post-mesopleura bristles. There were pteropleura, pteropleural bristles and the coxa of front, middle and hind legs in the thorax. Two pairs of sternopleural bristles were located in sternopleura. The halter was a small rod-like structure and it's a vestigial structure of rear wing. There were prostigma and post-stigma in the thorax and which were covered with bristles (Fig 3).

Observation on the dorsal of the thorax of W. magnifica

The dorsal of the thorax was covered with silver gray pollen and the mesothorax was well developed. The mesonotum consist of scutum, scutellum, postscutellum and mesophragma. The scutum was divided into prescutum and postscutum by the scutal sulcus. The scutum had three light black longitudinal grooves and two ridges. There were acrostichal bristles, dorsocentral bristles, intra-alar bristles, presulcal bristles, supra-alar bristles, prealar bristles in the scutum. The bristles on the scutellum was well developed and including discal scutellar bristles and margin scutellar bristles (Fig 4).

Observation on the abdomen of W. magnifica

Abdomen of *W. magnifica* was gray pollen and divided into five abdominal segments. On the dorsal, there were symmetrical black patch distributed in sagittal median line. The 3rd and 5th tergites has middle marginal bristles, respectively. Black patch in the 1st and 2nd abdominal segments were shaped like "m". There were three black patch in the 3rd to 5th abdominal segments, respectively. Middle black patch in the 3rd and 4th abdominal segments were large and they were connected with each other from 1st to 4th abdominal segments, black patch in the 5th abdominal segments were relatively small. On the ventral, each pair of abdominal segments was bilateral symmetry and separated by six sternitein the middle, the stigma were distributed in each segment, and the anus was located in terminalia (Fig 5).

Observation on the wings of W. magnifica

The wing was transparent, and there were two types of veins including longitudinal vein and cross- vein. The radial stem vein was thick and it was divided into the 1st radial vein and radius sector. The radial node which was located in the distal end of the radius sector divided into the 2nd and 3rd radial vein and the 4th and 5th radial vein. The medial vein was divided into the 1st and 2nd medial vein and the 3rd and 4th medial vein. The 1st cubital vein had a right angle bend at the wing base and was combined with the 1st anal vein to form the 1st cubital and 1st anal vein. The 2nd cubital vein was degenerated to form a shallow vestige, the 1st and 2nd medial vein (m1 + 2) had a right-angle bend and the 4th and 5th radial vein (r4 + 5) was open cell. The cross-veins included humeral cross vein, radio medial cross-vein, inter-medial cross- vein, and medial-cubital cross- vein, while the radio medial cross-vein lost its dark halo. The veins divided the wing membrane into form cells (Szpila *et al,* 2010), there were mainly basal costal cell, distal costal cell, subcostal cell, 1st radial cell, 3rd radial cell, basal 5th radial cell, distal 5th radial cell, basal 2nd medial cell, distal 2nd medial cell, basal

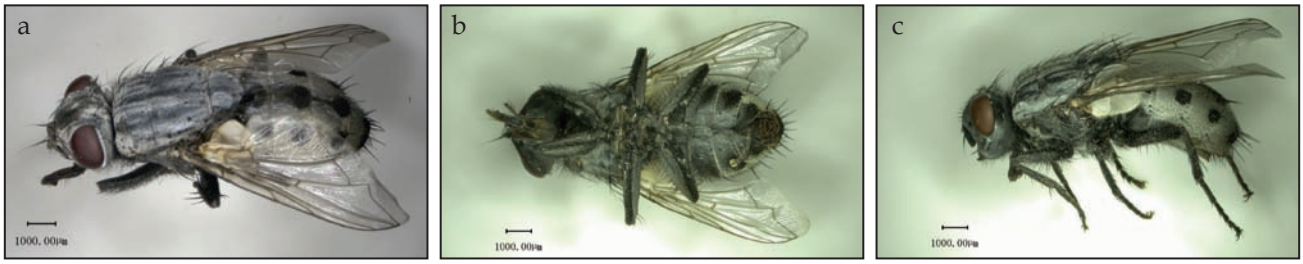

Fig 1. Observation on the whole body of *Wohlfahrtia magnifica* a: dorsal view; b: ventral view; c: lateral view

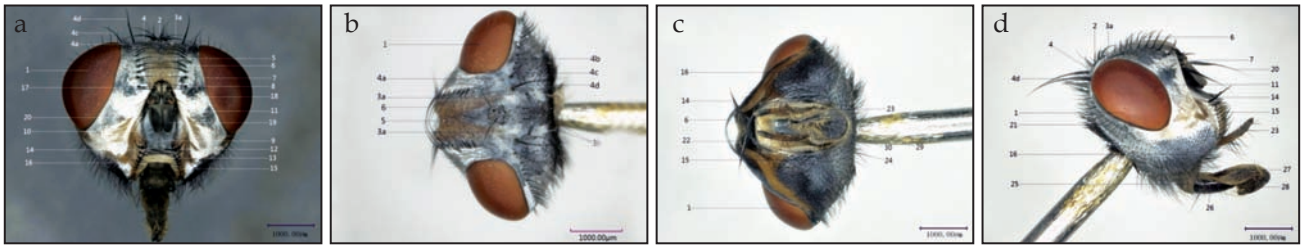

Fig 2. The observation of *Wohlfahrtia magnifica's* head

a: anterior view; b: dorsal view; c: ventral view; d: lateral view

1 - compound eyes; 2–ocellar triangle; 3a - ocellar bristles; 3b - post ocellar bristles; 4 - vertex; 4a - prevertical bristles; 4b - outer vertical bristles; 4c - inner vertical bristles; 4d - postvertical bristles; 5 - interfrontalia; 6 - frontal bristles; 7 - lateral bristles; 8 - lunule; 9 - mid-facial plate; 10 - facialia; 11 - parafacialia; 12 - facies bristles; 13 - epistoma; 14 - vibrissae; 15 - peristoma bristles; 16 - gena; 17 - the first segments of antenna; 18 - the second segments of antenna; 19 - the third segments of antenna; 20 - arista; 21 - post-compound eyes bristles; 22 - peristoma; 23– maxillary palpus; 24 - mouthparts fossa ; 25 -baisproboscis; 26 - haustellum; 27 - prementum; 28 - oral disc; 29 - labellum; 30 - labellum port

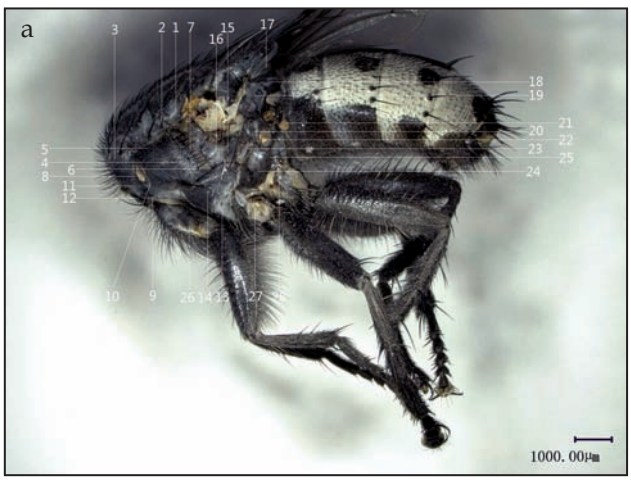

Fig 3. Lateral view of *Wohlfahrtia magnifica's* thorax and abdomen

1 - notopleura; 2 - notopleural bristles; 3 - humeral callus; 4 - mesopleura; 5 - front- mesopleura bristles; 6 - post-mesopleura bristles; 7 - clearance bristles; 8 - prostigma; 9 - prostigmal bristles; 10 - auxillary prostigmal seta; 11 - propleuron; 12 - episternum bristles; 13 - sternopleura; 14 - sternopleural bristles; 15 - pteropleura; 16 - pteropleural bristles; 17 - postscutellum; 18 - postnotum of mesothorax; 19 - superior pleurotergite; 20 - inferior pleurotergite; 21 - halter; 22 - post-stigma; 23 - beret; 24 - hypopleura; 25 - hypopleural bristles; 26 - front legs coxa; 27 - middle legs coxa; 28 - hind legs coxa

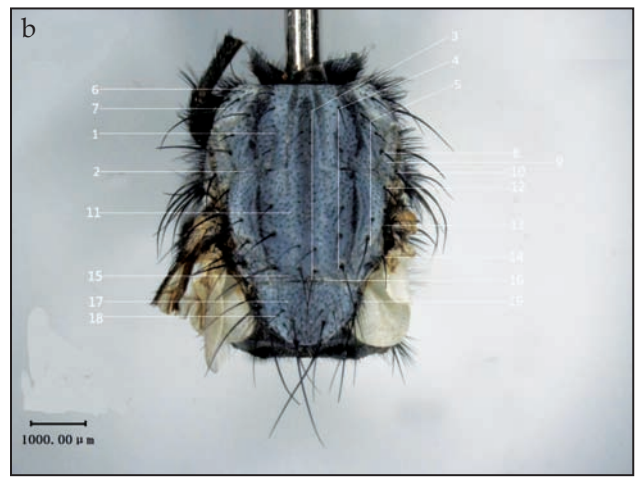

Fig 4. Dorsal view of *Wohlfahrtia magnifica's* thorax

1 - mesothorax prescutum; 2 - scutal sulcus; 3 - acrostichal bristles; 4 - dorsocentral bristles; 5 - intra-alar bristles; 6 - humeral callus; 7 - humeral bristles; 8 - presulcal bristle; 9 - notopleura; 10 - notopleural bristles; 11 - mesothorax postscutum; 12 - prealar bristles; 13 - supra-alar bristles; 14 - postalar bristles; 15 - postsulcal dorsocentral bristles; 16 - scutellar suture; 17 - scutellum; 18- discal scutellar bristles; 19 - margin scutellar bristles

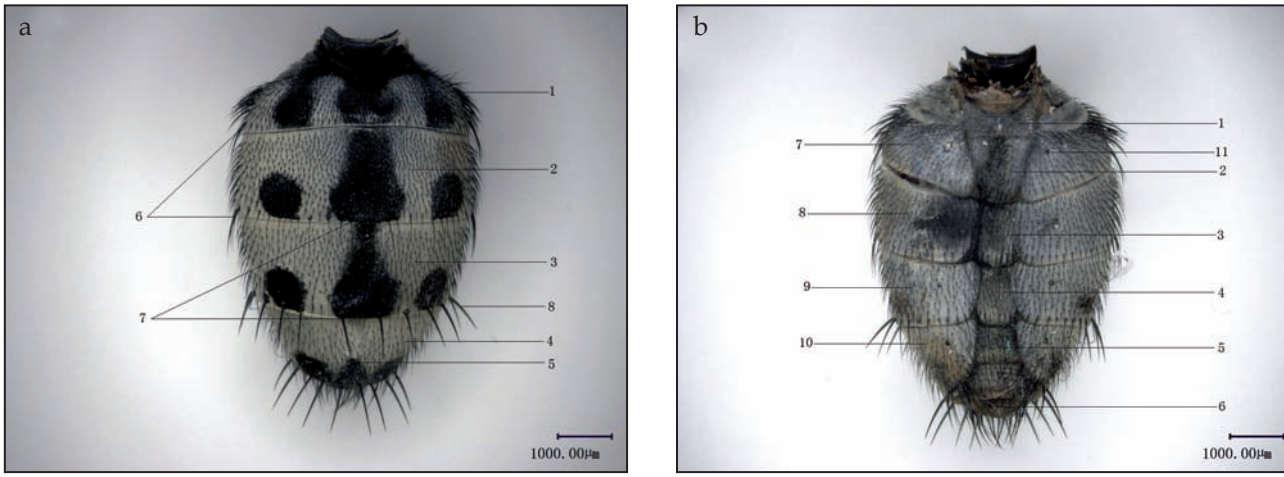

Fig 5. The observation of *Wohlfahrtia magnifica's* abdomen

a: 1 - 1st and 2nd abdominal segment; 2 - 3rd abdominal segment; 3 - 4th abdominal segment; 4 - 5th abdominal segment; 5 - discal bristles; 6 - lateral bristles; 7 - middle marginal bristles; 8 - lateral marginal bristles

b: 1 - 1st sternite; 2 - 2nd sternite; 3 - 3rd sternite; 4 - 4th sternite; 5 - 5th sternite; 6 - anus; 7 - 1st and 2nd abdominal segment; 8 - 3rd abdominal segment; 9 - 4th abdominal segment; 10 - 5th abdominal segment; 11 – stigma

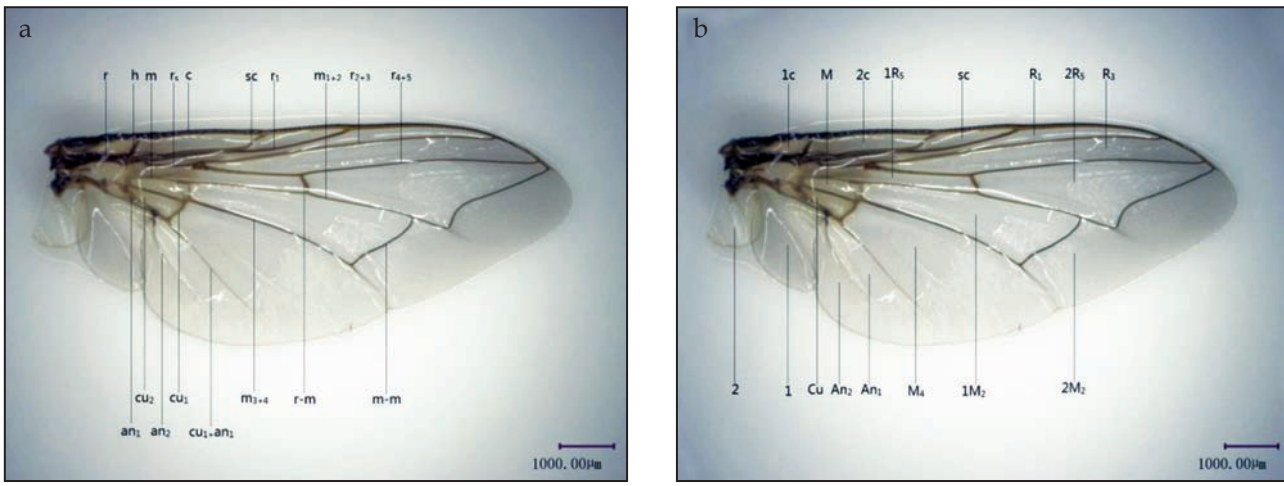

Fig 6. The observation of *Wohlfahrtia magnifica's* wing

a: veins of wing membrane c - costa; sc - subcosta; r - radial stem vein; r1 - 1st radial vein; rs -radius sector; r2+3 - 2nd and 3rd radial vein; r4+5 -4th and 5th radial vein; m - medial vein; m1+2 - 1st and 2nd medial vein; m3+4 -3rd and 4th medial vein; cu1 - 1st cubital vein; cu2 -2nd cubital vein; an1 -1st anal vein; an2 - 2nd anal vein; h - humeral cross-vein; r-m - radio-medial cross-vein; m-m - inter-medial cross-vein; cu1+an1- 1st cubital and 1st anal vein

b: cells of wing membrane 1c - basal costal cell; 2c - distal costal cell; sc - subcostal cell; R1 - 1st radial cell; R3 -3rd radial cell 1R5 -basal 5th radial cell; 2R5 -distal 5th radial cell; M - basal 4th medial cell; 1M2 - basal 2nd medial cell; 2M2 -distal 2nd medial cell; M4 - distal 4th medial cell; cu - cubital cell; An1 - 1st anal cell; An2 - 2nd anal cell; 1 – upper squama; 2 – lower squama

4th medial cell, distal 4th medial cell, cubital cell, 1st anal cell, 2nd anal cell. The squamae were white and attached with thorax, they were connected in the front of upper squama and lower squama, the lower squama was about twice the size of the upper squama (Fig 6).

Observation on the leg of W. magnifica

The *W. magnifica* leg consisted of a pair of propodium, a pair of mesopodium, and a pair of metapodium, each of which was divided into coxa, trochanter, femur, tibia and tarsus. The tarsus was composed of 5 tarsomeres, the 1st tarsomere was the longest, and the pretarsus had the claw and pulvilli, a large amount of bristles were covered around the legs (Fig 7).

Comparison of male and female flies

Observing on the male and female of *Wohlfahrtia magnifica*, the male abdomen was long oval and

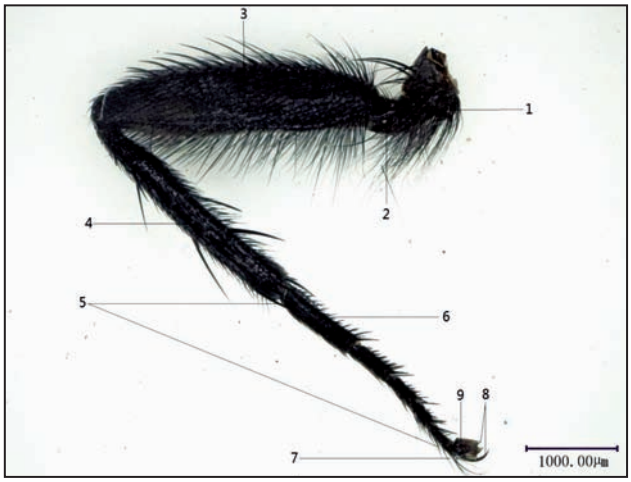

Fig 7. The observation of *Wohlfahrtia Magnifica's* leg

1 - coxa; 2 - trochanter; 3 - femur; 4 - tibia; 5 - tarsus; 6 - 1sttarsomere; 7 - pretarsus; 8 - claws ; 9 - pulvilli.

female abdomen was oval, abdomen of male flies was longer and narrow than that of female flies. Difference of the terminalia between male and female were the most obviously, caudal end of abdominal segments in male flies was smooth and its terminalia was convex, tergites of the last abdominal segments formed a dark yellow eminentia which extended downward oblique to form saccular process, a spoon–like expansion. Female flies had serration in the caudal end of abdominal segments and its terminalia was concave and anal papilla was grayish white (Fig 8 and Fig 9).

Measurement of W. magnifica

20 adult flies were randomly taken to measure each part of body with vernier caliper (Fig 10 and table 1)

According to the data analysis, the adult length of *Wohlfahrtia magnifica* was about 1.423 cm and belong to a medium and large fly. The length of head was about 0.280 cm and its width was about 0.437 cm, the length was about half of width of the head, the length of wing was about 1.005 cm and its width was about 0.401 cm, the width was about half of the length of wing; length of the thorax and abdomen was slightly larger than its width, respectively, the propodium and mesopodium were similar in length and the metapodium was longer than the other two legs.

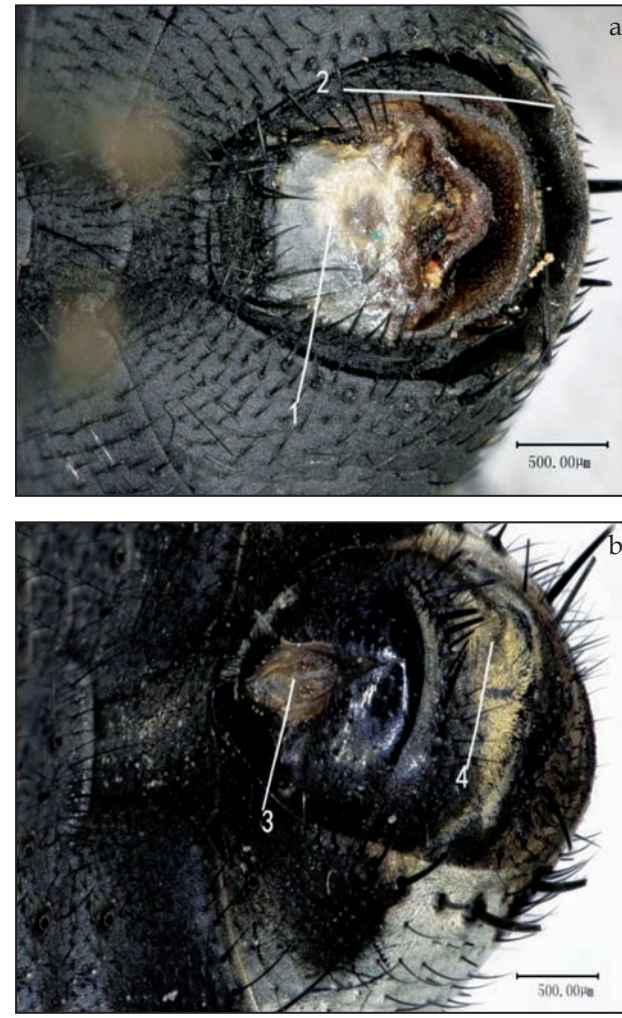

Fig 8. Comparison of terminalia for male and female of *Wohlfahrtia magnifica*

a: Female b: male 1 - anal papilla; 2 - serration; 3 - anus; 4- saccular process

Discussion

The *W. magnifica* is widely distributed in the Mongolia plateau, causing myiasis of many kinds of livestock. Vaginal myiasis occurs from the end of May to the beginning of October in the Bactrian camel.

Lunule in the head of *W. magnifica* were located between the antennae and interfrontalia, and it's extending downward to separate the facial and parafacialia, the gena height was obviously a half high than that of eye, occiput was backward expansion

Table 1. Size measurement of various parts of *Wohlfahrtia magnifica* (unit: cm)

hl	0.280±0.004	tl	0.500±0.009	al	0.641±0.013	wl	1.005±0.018	pl	1.076±0.031	mpl	1.143±0.026
hw	0.437±0.005	tw	0.473±0.010	aw	0.449±0.008	ww	0.401±0.009	ml	1.056±0.057	bl	1.423±0.016

pl - propodium length ; ml - mesopodium length; mpl - metapodium length
Note: the measured data was the average value ± standard error

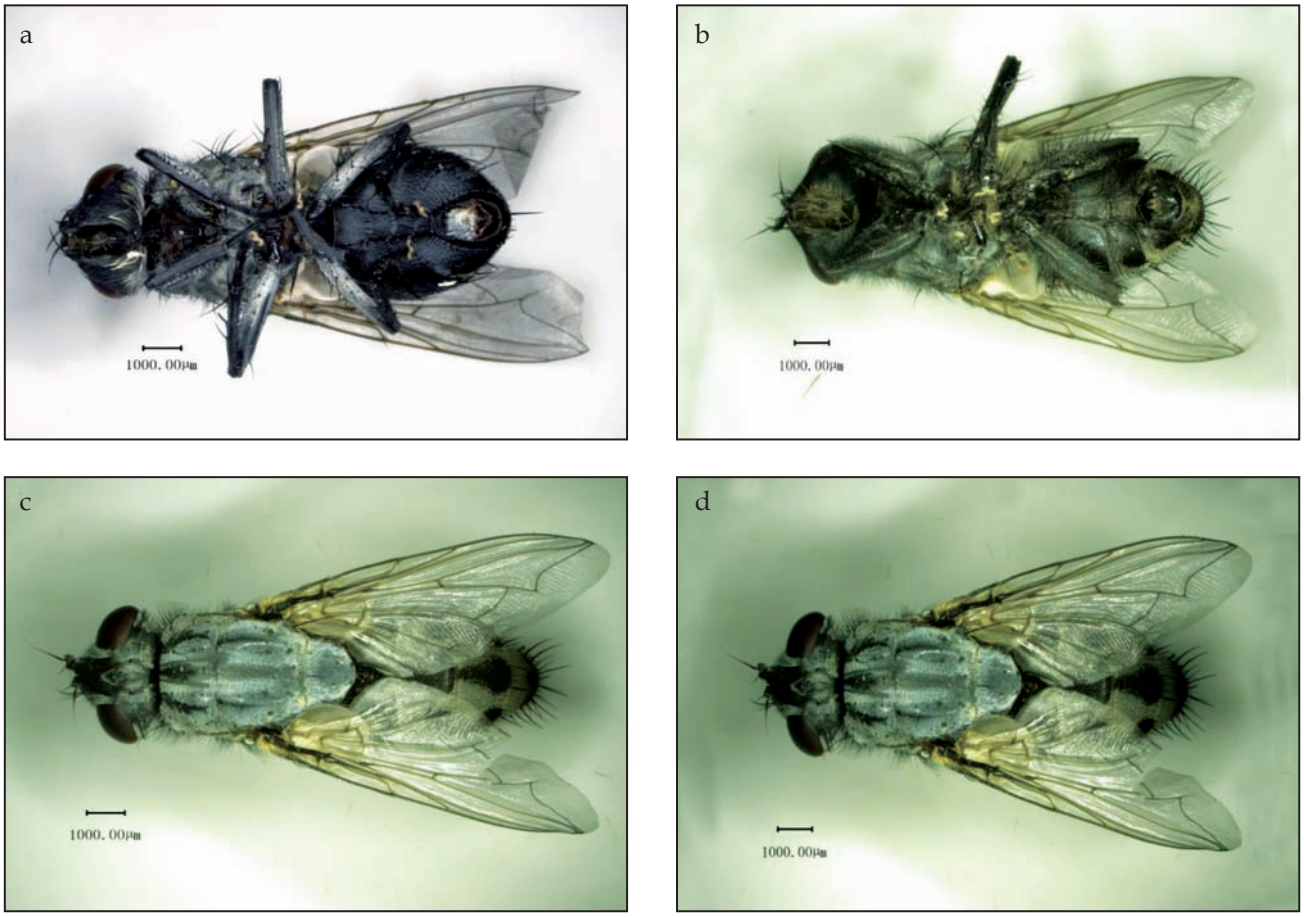

Fig 9. Morphological Comparison between male and female of *Wohlfahrtia magnifica*

a: female ventral view; b: male ventral view; c: female dorsal view; d: male dorsal view

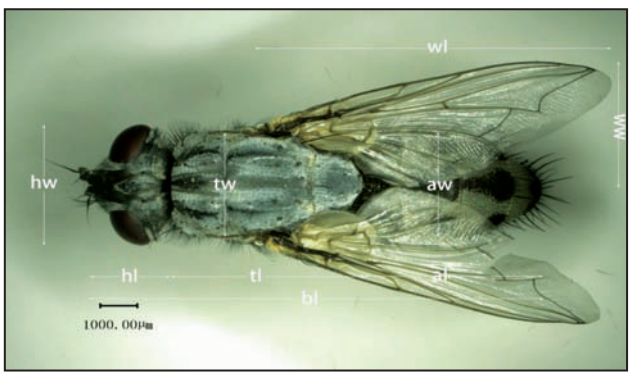

Fig 10. Dimension measurement and marking of various parts of *Wohlfahrtia magnifica*

hw - head width; hl - head length; tw - thorax width; tl - thorax length; aw - abdomen width; al - abdomen length; ww - wing width; wl- wing length; bl - body length.

(Baohai, 2001). The distal of maxillary palpi were black and maxillary palpi had olfactory and gustatory functions (Zhongcheng, 2005). The antennae of *W. magnifica* had three segments, arista was located in the base of the 3rd segment and covered by feathery cilia (Mengyu and Xinsheng, 1990). The antennae was slightly short, the length of the 3rd segment was 1.5 times as long as the 2nd segment of antennae (Zhang Ming, 2016). Thousands of olfactory receptors were distributed on the antennae, which was of great significance in host, spawning, mate seeking and other behaviours of insects (Szyszka and Galizia, 2015; Tan Jing *et al,* 2016).

The anal-cell of wing was short and closed away from the wing margin, the squamae and alulae were differentiated obviously. The wings used for flying by insects were made up of membranous and there were crisscross veins on the wing surface, actually veins were formed by thickening parts of the wing surface at the trachea, which acted as a skeleton to support and strengthen the wing surface, and was also related to the twisting movement of the wing during flight. The halter play a balancing role in flight (Jinping, 2017). The wings comprised of nonsmooth microstructures, this kind of structure not only enhanced the hydrophobic performance of wing surface and provideed mass stabilisation for flight,

but also effectively reduced the reflectivity of wing surface (Yanling *et al,* 2014).

The base of legs was close to each other, and located in the ventral of thorax, the scutal sulcus of mesothorax was obviously integrity (Whitmore *et al,* 2013). The three longitudinal grooves and two ridges on the dorsal of thorax were obviously and increased the contact area with environment. There were bristles arranged like arc in the hypopleura which located in the under of the anterior of post-stigma. The acrostichal bristles on the dorsal of thorax had five bristle sites, the anterior three were tiny and posterior two were developed. "Vase- like shaped" black patch were on the dorsal of abdomen, and black patch on dorsal of the 5^{th} abdominal segments were clearly, the abdomen was covered with silver gray pollen.

According to observation, the *W. magnifica* was belong to medium and large flies and with darker colour. Morphological difference was obvious in the terminalia between male and female. With the naked eye, the terminalia of male was bulge outward to form convex and the terminalia of female was sunken inward to form concavity. Under anatomical microscope, we can see the dark yellow saccular process in the male flies and the white anal papillae in the female flies; anal papillae were used to hold the maggots and help the maggots adhere to object. The external genitalia of insects play an important role in courtship and mating behaviour (Eberhard, 2010). Through this study, we had a comprehensive understanding of the morphological structure of *W. magnifica* living in the Mongolia Plateau, which would enrich people's knowledge of the biological characteristics of *W. magnifica*.

Acknowledgements

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ICO Signs Cooperation Agreement with International Organization to Protect Camels during Transport

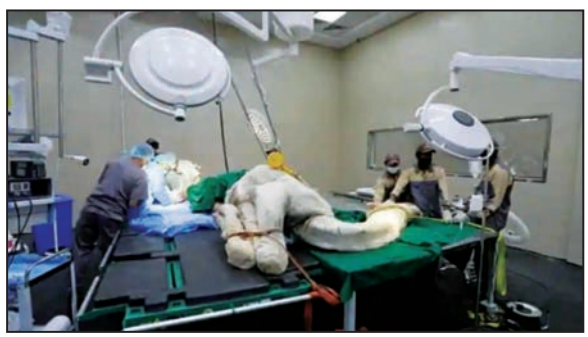
The International Camel Organization (ICO) has signed a cooperation agreement with the international Angels of Animals organisation to improve the levels of camel breeding and transport from one place to another around the world, and to support scientific and applied research in this field. This agreement aims to overcome all challenges that camels face in their transport all around the world. The ICO seeks through this agreement to develop camel transport operations over long distances through multiple means of transport, in addition to spreading awareness of the importance of adhering to animals welfare regulations and the safety of their transportation at the global level.

Active Camel Research Scientist Award 2020

In this Covid 19 pandemic year 2020 when everything was stand still, laboratories and institutions were closed, we were quite apprehensive about receiving manuscripts for various issues of JCPR. Astonishingly, there was an overwhelming response from the camel scientists who contributed excellent manuscripts for JCPR. As a reciprocating gratitude gesture Camel Publishing House will issue an award certificate bearing a title, "ACTIVE CAMEL RESEARCH SCIENTIST AWARD 2020" to all the authors of volume 27 of JCPR. This award is dedicated to those camel research scientists who continued to work in the year 2020 during Covid 19 pandemic despite of several constraints and preferred to get their work published in the Journal of Camel Practice and Research. All authors will receive a PDF of this award by email in the first week of January 2021. Congratulations to all.

Largest camel hospital in the world opens in Saudi Arabia

The world's largest camel treatment and research facility opened in Saudi Arabia's Qassim Region. The more-than 100 million riyal ($26.7 million) facility is one of only a few in the world dedicated to camel care and spans 70,000 square meters, SPA reported. Governor of Qassim Region Prince Dr. Faisal bin Mishaal bin Saud bin Abdulaziz inaugurated Salam Veterinary Group's camel hospital and toured the facility upon its opening. It provides basic medical care but also treatment for chronic and infectious diseases, surgery and radiology services to the Kingdom's beloved desert animal. It also focuses on the research and development of camels' breeding and fertilisation processes, with a specialist center for embryonic transfer technology. Previously, camel owners and traders in the Qassim region, also known for hosting the world's largest camel market, used to drive 655 kilometers to treat their animals. The Salam Veterinary Camel Hospital can handle nearly 145 camels and the stables can hold upto 400 camels. The research centre is working on mitigating diseases and other risks to the animals. The facility will be generating employment for nearly 300 people across six different units, namely surgery, medical treatment, camel accommodation, laboratories for testing and research and a calf unit. The local authorities have also urged the employees to take care of proper sanitisation while interacting with each other and the camels.



(Courtesy: Lauren Holtmeier, Al Arabiya English Wednesday 08 July 2020)

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DISTAL LIMB LAMENESS IN DRAUGHT DROMEDARY CAMELS

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ABSTRACT

This study was conducted to study the prevalence and incidence of lameness in draught camels in private camel farms. The incidence and prevalence of the distal limb lameness was 28.22% out of 450 draught camels. Lameness was more frequently observed in the distal forelimbs (53.5%) than hind limbs (46.6%). The foot disorders were the most common disorders causing lameness (59.05%) and the fetlock and metacarpus (MC)/metatarsal (MT) disorders were 40.94%. The distal forelimb affections had a higher incidence of foot disorders (31.5%) and fetlock and metacarpus (MC)/metatarsal (MT) disorders (22.0%) compared with the hind limbs. The prevalent distal limb disorders were the interdigital fibroma (17.3%), foot wounds (14.66%), foot abscess (14.66%), swollen digit (13.3%), the sole ulcer (9.3%), incomplete separation of the nail (6.67%) and septic distal interphalangeal joint arthritis (6.65%). Degenerative joint diseases of fetlock were (21.15%), pastern (6.65%) and coffin joint (9.33%). The MC/MT osteoperiosteal reactions (25.00%), angular fetlock deformity either valgus or varus deformities with toe-out or toe-in (21.24% and 11.53%, respectively) and septic tenosynovitis (21.15%) showed the highest causes of lameness in draught camels. The abnormal camel limb conformations, excessive workloads, trauma, and the camels used for long hours in harsh and unsuitable sandy and rocky ground conditions were considered the causes of lameness. This study will provide an assessment of the incidence of distal limb disorders in working camels.

Key words: Distal limb, draught camel, lameness, foot disorders, fetlock disorders

Draught camels are often overburdened and used for long hours in harsh conditions and suffer from a variety of musculoskeletal disorders and lameness. Camel can carry a load up to 300 kilograms over long distances and more than 450 kilograms over a short distance (Gahlot, 2007). Lameness in camel has widely different etiology which includes direct trauma, nutrition fractures, punctured foot (Gahlot, 2007 and Al-Juboori, 2013) and abnormal conformation of the limbs (Mostafa and Khalil, 2018). The commonly reported surgical musculoskeletal affections were phalangeal and foot affections, punctured foot, foot abscess, avulsion of the toe nail, severed flexors and congenital flexion of the fetlock, medial deviation of the fetlock, laceration at metacarpus (Qauzi, 2010 and Gharu, 2014); fracture, sprain, subluxation and punctured foot (Ramadan, 1994).

Camel lameness was represented through Partial or non-weight bearing by one or more limbs, swelling over joint, pain on palpation, toe-out postures, shivering while sitting, semi flexed hock in sitting postures and asymmetry of the pelvis (Gahlot, 2007). The objective of this study was to identify the range and prevalence of distal limb pathological conditions contributing to lameness in draught camels.

Materials and Methods

This study was carried out on 450 camels belonging to private camel farms from March 2016 to November 2019. The camels were of both sex (112 males and 15 females) with a mean age of 6.65 ± 3.88 years. These animals were examined for various surgical affections of fore and hind limbs

Lameness recorded in the present studies were based on history and clinical examination. The occurrence of lameness and surgical affections of the distal fore and hind limbs were examined clinically and radiographically. Clinical examination included a gross examination of the distal fore and hind limbs of a camel in standing position and palpation.

Animals were observed in standing from front, sides, and rear positions and during a motion for diagnosis of lameness and surgical conditions. The supportive, swinging, or complementary type of lameness were determined. Camel progression in a straight line, in a circle (clock and anticlockwise),

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and by observing gait on sandy, hard tracks (Gahlot, 2007). The solar and dorsal surface of foot was examined after removing the dirt by washing and cleaning for swelling, embedded foreign body, growths, exuberant granulation, sinus tracts, and others. The site of pain was located by physical examination, thumb palpation, or hoof tester. Exploratory puncture of the swelling was done to aspirate the contents in order to differentiate abscess from other swellings.

Clinical examination was performed under xylazine (Xylaject, Adwia Co., Cairo, Egypt.) at a dose rate of 0.25 mg/kg after securing camel by rope halters and taken into lateral recumbency. A distal limb radiographic study was taken in routine dorso/palmar/planter and lateromedial projections. For foot radiography dorso/palmar/planter projections were made. The radiographic factors used were 55-65 Kvp and 10mAs and 80 cm FFD (Mostafa *et al,* 1993).

Incidence/ occurrence of clinical distal limb surgical conditions causing lameness was recorded on either the fore and hind limbs. The incidence and percentage were represented as the mean ± standard error (±SD).

Results and Discussion

The number of camels with the distal limb lameness was 127 (28.22%) out of 450 examined camels. Gahlot (2013) found that the incidence of lameness in camels was 24.61% followed by arthritis (16.92%) and wound (16.92%). The incidence of lameness in racing camel was found to be 9.39% in acute and 2.50% in chronic lameness (Al Juboori, 2013). However, the incidence of lameness in draught camels in the present study was higher compared with racing camels. Furthermore, the incidence of musculoskeletal disorders causing lameness in camels was 24.63% in forelimbs and 26.15% in hind limbs (Quazi, 2010). In addition, Singh and Gahlot (1997) reported the incidence of musculoskeletal lameness in the clinical (10.10%) and in the field (55.62%).

Reix *et al* (2014) and Broster *et al* (2009) reported that the incidence of lameness in working donkeys and horses was 96%. The differences in incidence of lameness between camels and equine could be attributed in the present study to the distinguishing features of the camel's limbs as being relatively long and slender strong to raise the body away from hot sand and have a broad flat leathery pad to adapt to the desert environment (Soliman, 2015).

The current study revealed the incidence of the forelimbs was 53.5% and 46.6% in the hind limbs. Similar results coincided with Al-Juboori (2013) who diagnosed the incidence of camel forelimb lameness (62.45%) and hind limbs (37.55%). In this respect, Stashak and Hill (2002) reported that 95 % of lameness was found in the distal forelimb of the horse due to the forelimbs receive the shock of landing and subjected to a great concussion. Contrary to our findings, draught donkeys and horses had a greater significant hind limb lameness than forelimbs due to the propulsion required for draught traction work (Maranhão *et al,* 2006).

Accordingly, in the present study, the distal forelimb affections had higher incidence of foot disorders (31.5%) and fetlock and metacarpus (MC)/metatarsal (MT) disorders (22.0%) compared with the hind limb (Fig 1). Table (1) revealed that foot disorders were the most common distal fore and hind limb disorders causing lameness (59.05%) and the fetlock and metacarpus/metatarsal disorders were 40.94%. The similar findings were also diagnosed in foot disorders in camels (59.37%) (Gahlot, 2011).

The interdigital fibroma (17.3%) appeared as a firm mass in the middle of interdigital space covering the dorsal aspect of both digits and the skin covering of the fibroma was ulcerated (Fig 2). In bovine, Amstel and Shearer (2008) pointed out chronic skin irritation from grazing stubble or rocky pastures or poor limb conformations were the common cause of interdigital hyperplasia. Consequently, draught camel fibroma in the present study might be attributed to hard chronic irritation of unsuitable working surfaces such as sharp rocky stones and thrones

Foot wounds (14.6%) were mainly punctured wounds. The wound had swollen, hot, painful resulted from sharp pointed foreign bodies at the heel or lateral aspect of the footpad or at the abaxial aspect of the 4th digit (Fig 3). In previous study Gahlot and Chouhan (1992), Purohit and Chouhan (1992), Ramadan (1994), and Gahlot (2011) reported that punctured foot had the highest incidence in camels. The penetration of the sole by glass pieces, thrones, nail, and sharp foreign bodies was considered the main cause of punctured foot in camels (Jhirwal *et al,* 2007). Therefore, draught camels in the present study have a high incidence of the punctured foot. This suggests that the road surfaces predisposes foot for the injuries like punctured foot. Furthermore, secondary complications were recorded in draught camels such as septic distal interphalangeal joints and phalangeal phlegmon.

Swollen digit (13.3%) clinically appeared as painless, diffuse firm swelling involving one

Table 1. The distribution of the most common distal limb disorders in the fore and hind limbs in draught camels.

Surgical affections	Limb		Total	Percentage
	Fore	Hind		
1. Foot disorders				
Interdigital fibroma	8	5	13	17.34%
Localised foot abscess	7	4	11	14.66%
Foot wounds	5	6	11	14.66%
Swollen digits	6	4	10	13.33
Sole ulcer	3	4	7	9.34%
DJD coffin joint	2	5	7	9.33%
Nail affections	2	3	5	6.67%
DJD pastern joint	3	2	5	6.65%
Septic coffin joint arthritis	3	2	5	6.65%
Ankylosing arthritis	1	0	1	1,30%
Total foot disorders	40	35	75	59.05%
2. Fetlock disorders				
Osteoperiosteal reactions(MC/MT)	9	4	13	25.00%
Angular medial deviation	6	5	11	21.24
Septic tenosynovitis	4	7	11	21.15%
DJD fetlock	5	6	11	21.15%
Angular lateral deviation	4	2	6	11.53%
Total fetlock disorders	28	24	52	40.94%
Total fore and hind limbs	68	59	127	28.2%

DJD: Degenerative joint disease. MC/MT: metacarpal/metatarsal.

digit extending to the phalangeal areas (Fig 4). Radiographically, increased thickness of the soft tissue of the affected digits was found to be associated with osteoperiosteal reactions on the PII and PIII. This could be attributed to the continuous repeated traumatic injuries of these digits during walking on rocky, stony and uneven surfaces. In addition, Ramadan (1994) diagnosed cases of diffuse swelling of the digits and termed an elephant foot due to the sand flies biting.

The sole ulcer (9.3%) appeared an irregular small circumscribed loss of the keratinised layer of the footpad (Fig 5) with a severe loss in the surrounding keratinised pad at the solar aspect. Weaver (1975) attributed ulceration of the sole in cattle to bad housing and husbandry conditions such as short, rough standings, gross neglect of hoof trimming, and the excess feeding of concentrates. On the other hand, Greenough (2015) reported sole ulcer results from activation of metalloproteinase in subclinical laminitis and crushed and ischemia of the corium associated with the unhygienic condition. The camel sole ulcer might be due to overworking in unsuitable roads.

In the present study, draught camels have a high incidence of foot disorders, i.e. foot abscess (14.66%), incomplete separation of the nail (6.67%), and septic distal interphalangeal joint arthritis (6.65%). Foot abscess either interdigital or localised (Fig 6) were considered as a common sequel to traumatic pain penetration and punctured wound caused by rocky stone and throne planet (Dioli and Stimmelmayr, 1992). The observed causes in the present study were excessive workload and long draught hours on unsuitable road surfaces.

Degenerative joint disease (DJD) of the proximal (Fig 7a, b) and distal interphalanageal (Fig 8a, b) joints were seen and radiographic pictures showed uneven joint spaces and periarticular bone growths. Ramadan (1994) and Gahlot (2007) reported that camel arthritis and punctured foot were the main causes of lameness in camels. In addition, ankylosing arthritis of the PIP joint associated with osteoperosteal reaction covering the distal half of the PI and proximal 2/3 of the PII was seen in aged camel more than 10 years old. This might be attributed to traumatic injuries or long harsh hours working in a rocky stony and rough or uneven grounds. The same has been reported in camels by

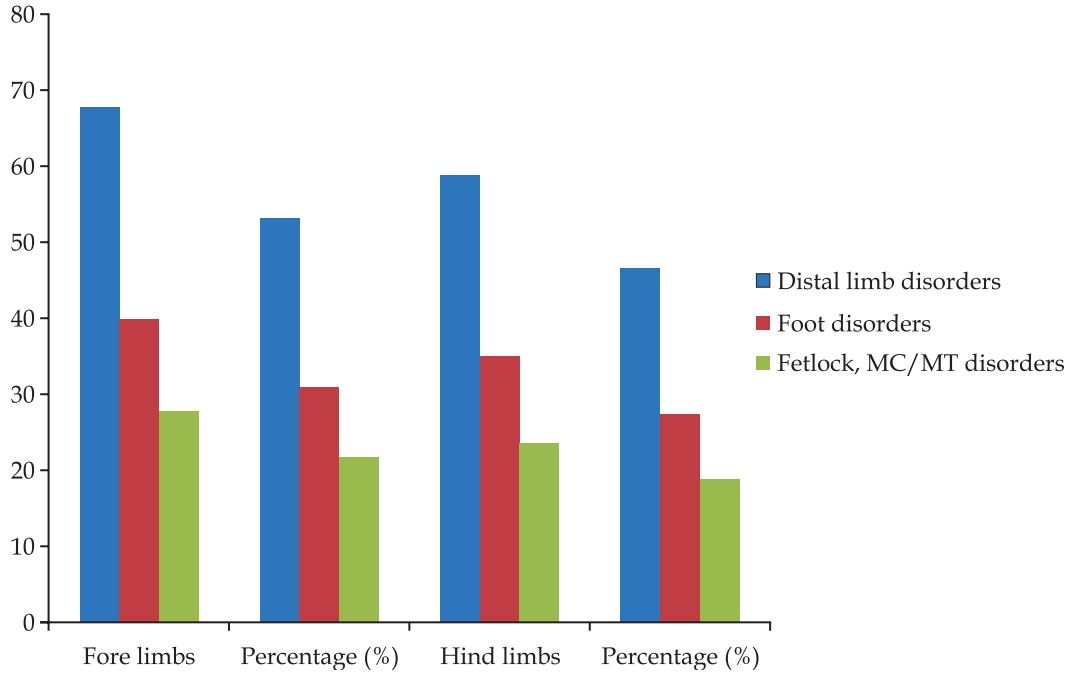

Fig 1. Histogram for distribution of distal limb lameness in fore and hind limb of draught camels.

Fig 2. Interdigital fibroma at the proximal part of the interdigital space with phlegmon of the foot (arrow).

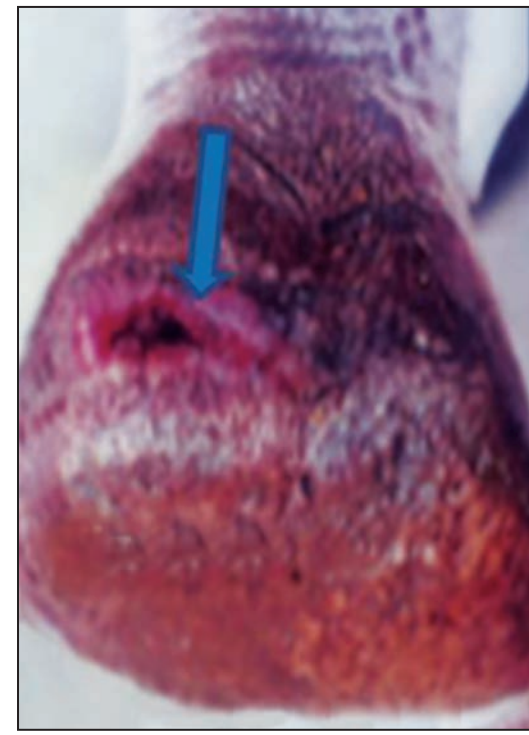

Fig 3. Punctured foot wound at the heel (arrow).

Ramadan (1994). Similar findings have been reported in draught horses predisposed to OA of the PIP and DIP joints with short, upright pastern and other factors such as angular limb deformities toe-in or toe- out conformation could play a substantial role (Goble, 2011).

Mostafa and Khalil (2018) reported that the most common camel limb conformations predisposed to fore and hind limb lameness were base-wide (26.9%), base narrow (10.1%), toe out (15.4%), upright pastern (13.9%) and sloppy pastern (18.2). Stashak and Hill (2002) reported that upright pastern in

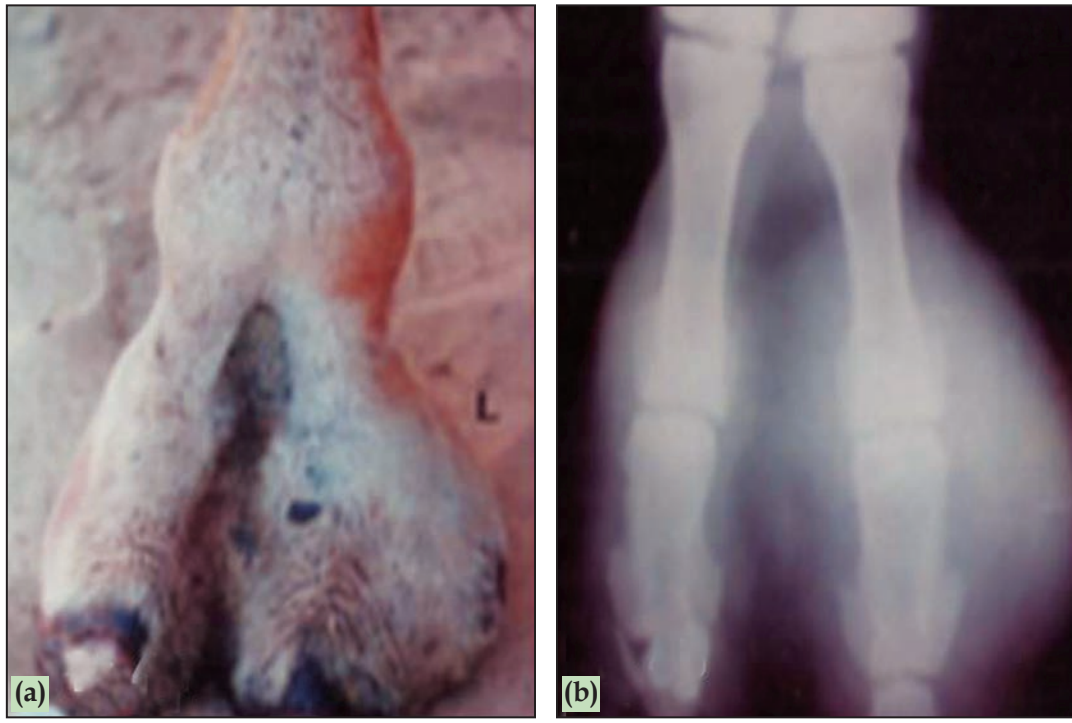

Fig 4. **(a)** Some degree of digital swelling at the 4th left hind digit, **(b)** Radiography showed severe soft tissue thickening at abaxial digit.

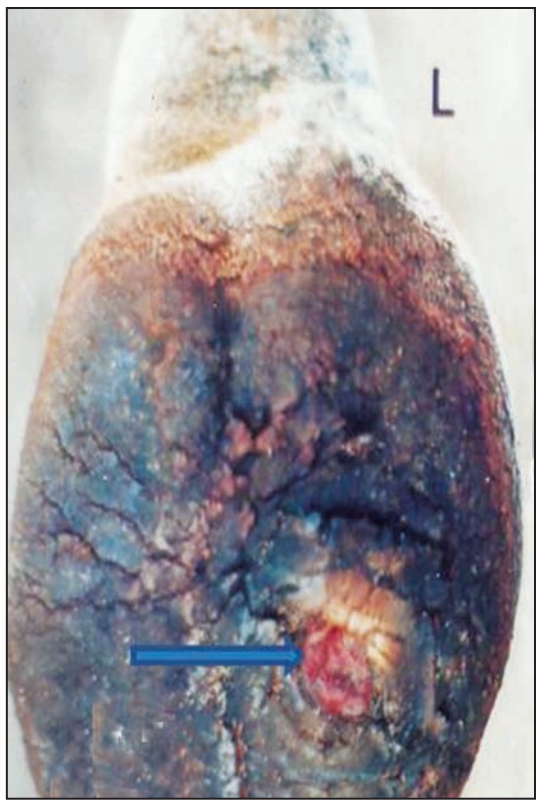

Fig 5. Superficial circumscribed sole ulcer with excessive wearing of the surrounding keratinised foot pad (arrow).

Fig 6. Open interdigital abscess discharging pus (arrow).

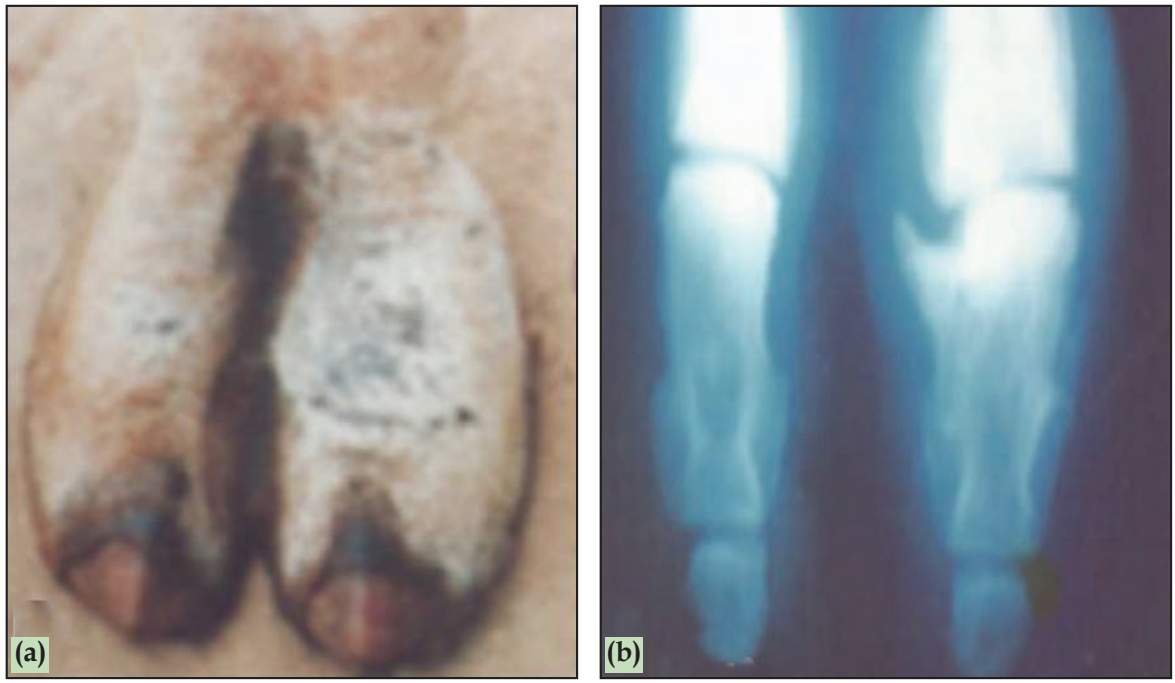

Fig 7. **(a)** Degenerative Joint Disease (DJD) in form of osteoarthritis of the proximal interphalangeal joint (PIP) with marked swelling at the abaxial digit. **(b)** Radiography revealed osteosclerotic bone reaction, osteoperiosteal reaction at the distal articular surface of PI and proximal articular surface of PII. Marked lipping on the axial border of PII.

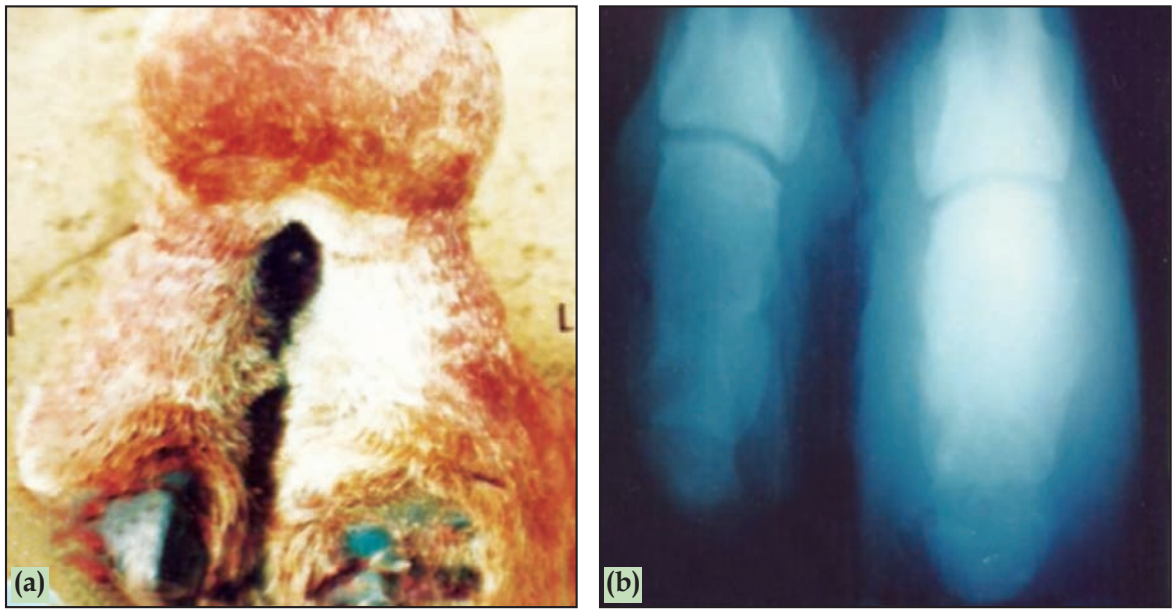

Fig 8. (a, b): DJD in form of osteoarthritis of the distal interphalangeal joints (DIP) or coffin joint . Notice osteoperiosteal reaction covering the PII and PIII and involving the DIP.

horses predisposes to concussion and injuries to the fetlock, phalangeal, and soft tissue structures. Moreover, Anderson *et al* (2004) reported that toe out creates excessive strain in the inner side of the hoof, pastern, and fetlock predisposing to DJD, ring bone and SDFT tendonitis, and suspensory desmitis in horses. Therefore, the observed DJD of the fetlock, pastern and coffin joints in the present study could be attributed to abnormal camel limb conformations associated with excessive workloads and the camels used for long hours in harsh unsuitable ground conditions. Similar findings have been observed in working horses (Broster *et al,* 2009) and donkeys (Reix *et al,* 2014).

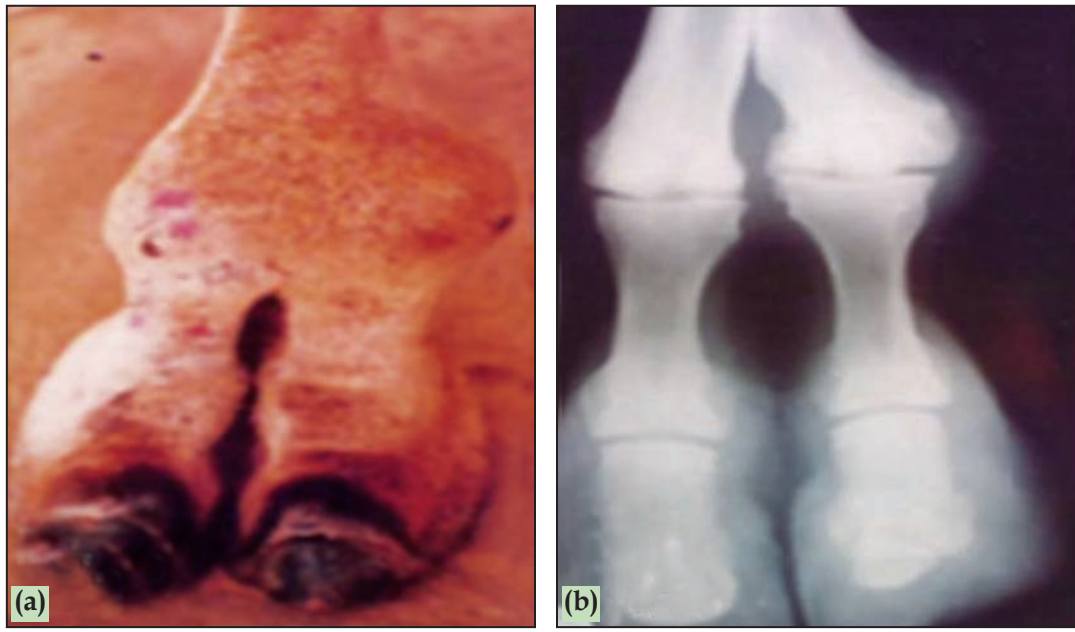

Fig 9. **(a)** Angular limb deformities with valgus and toe out conformation and **(b)** Varus with toe in conformation.

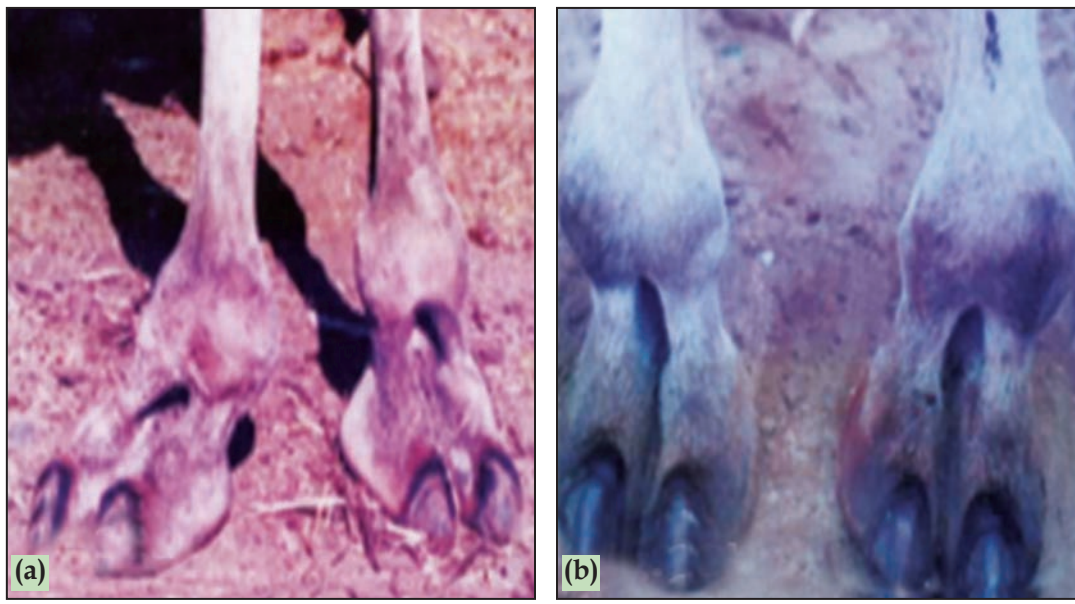

Fig 10. **(a)** Angular fetlock deformity with marked swelling, medial fetlock deviation, **(b)** Dorso-palmar radiograph showed broadening and irregularity of the articular surface of the distal end of the metacarpal.

The current study revealed that the metacarpal (MC)/metatarsal region and fetlock joint region had 40.94% of the distal limb disorders. Stashak and Hill (2002) reported that the superficial situation of the horse distal limb structures were accounted for high pathological disorders

Angular fetlock deformity either valgus or varus deformities with toe-out or toe-in were 21.24% and 11.53%, respectively (Fig 9a, b). Fahmy *et al* (2006) diagnosed angular fetlock deformity of the hind limbs in 6929 camels suffering from different degrees of valgus or varus deformities. Moreover, toe out conformation induced excess strain on the inner side of the pastern, and fetlock predisposes DJD of the fetlock and pastern, ring bone and foot soreness in the horses (Thomas, 2005). Similar results have been observed in camels fetlock DJD (Fig 10 a, b) and foot disorders in the present study.

In the present study, the osteoperiosteal (25%) reactions of both MC/MT regions were observed most commonly in the palmar/planter or dorsal aspects as single or multiple osteophyte reactions or exostosis. In addition, open traumatic septic tenosynovitis for flexor tendons (21.15%) was also noted. Aljaboori (2013) diagnosed sore shin with osteophyte formation (10.64%) in racing camels due to the traumatic opening of soft tissue structures. Goble (2011) attributed exostosis of MT/MC in the horse to direct trauma or instability due to abnormal limb conformation. In conclusion, this study reveals a high prevalence incidence of distal limb lameness in draught camels.

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DERMATOPHYTOSIS IN A NOMADIC CIRCUS CAMEL AND ITS MANAGEMENT WITH MICONAZOLE THERAPY

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ABSTRACT

The present study reports the incidence of dermatophytosis in a circus dromedary aged about 2.5 years with emphasis on clinical signs, laboratory examination and treatment. Camel had skin problem persisting for a month. Skin scrapings were collected from the affected camel and examined by direct microscopy method and cultural examination for isolation and identification by standard protocols. It documented the presence of superficial dermatophyte - *Trichophyton* spp skin infection in the affected camel. The camel was treated with Miconazole topical therapy combination and it resulted in an initiation of recovery by 7th day and complete recovery was reported by fourth week.

Key words: Dromedary camel, dermatophytosis, miconazole therapy

Skin diseases contribute to one of the major problems in camel, having various causes such as bacteria, viruses, parasites, fungus, yeast, tumour and allergies (Shokri and Khosravi, 2011). Among them fungal, i.e. infection, dermatophytosis caused by dermatophytes, is considered to be one of the most important contagious and zoonotic disease (Ghoke *et al,* 2006) that can spread among animals via direct physical contact with infested animal and indirectly through contaminated fomites (Ganguly *et al,* 2017b). Mohammadpour *et al* (2020) reported that camels are one of the important carriers and source of infection for human, livestock and wildlife in Iran.

Dermatophytes are keratolytic fungi which invade keratinised *Stratum corneum* of the epithelial skin layers and its appendages like hair, feather, horn and nail, causing mild to severe, localised and/or diffuse infections (Sabra and Al-Harbi, 2015). The infected animals usually have roughly circular patch with hair loss and the skin becomes thickened, flaky, crusty and greyish in colour (Wisal *et al,* 2010). Dermatophytes infection is more prone in immune compromised animals where the incidence is high in young camels less than three years of age (Wisal *et al,* 2010, Ganguly *et al,* 2017a). The long haired skin of the camel gives proper habitat for the growth of dermatophytes (Baghza *et al,* 2016). Though camels are often potential carriers of these microorganisms, for which little information has been documented (Ganguly *et al,* 2017a). The present study documented the incidence of dermatophytosis in nomadic circus camel and its effective management using Miconazole topical therapy.

Materials and Methods

Case history and clinical examination

A male nomadic circus camel aged about 2.5 years was presented to the Large Animal Medicine Referral Clinic of the Veterinary College and Research Institute, Orathanadu, with the history of skin disease since last 30 days. There were crusty hairless areas distributed over the head, neck, shoulder, flank, limbs and perineal region. The camel was having its normal feed intake, rumination, defecation, and urination. Dermatological examination revealed dry, odourless, greyish-white, round, circular to irregular patches of hair less areas, which were little raised above the skin and were seen with powdery scales

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on the skin (Fig 1). Cotton swabs soaked in 70% alcohol were used to sterile the infected area prior to sample collection. Skin scrapings were collected from the lesions by using sterile scalpel blade in Petri dish. Blood samples were collected through jugular vein in vials containing EDTA as an anticoagulant. Haematological parameters were assessed as per the standard methods (Coles, 1986).

Mycological Evaluation

a. Direct microscopic examination

A part of the collected skin scrapings in 20% Potassium Hydroxide (KOH) was placed on a clean glass slide to digest the keratin material. This was then covered with a clean glass and gently heated for one minute without boiling. This was stained with lactophenol-cotton blue and microscopically examined under a light microscope using low and high power magnifications, for the presence of fungal elements of arthropores and hyphae.

b. Mycological culture

A part of the skin scrapings was inoculated with Sabouraud's dextrose broth and incubated at 35°C for 72 hours and then streaked onto Sabouraud's dextrose agar containing Chloramphenicol (0.05mg/ml) and Actidione (0.5mg/ml) by spread plate method of culturing, to obtain colonical structure (Weitzman and Summerbell, 1995; Enany *et al,* 2013). Isolated dermatophytes were identified on the basis of the rate of growth and gross morphology of the colony and on the microscopic features of the fungal isolates as per standard protocols (Frey *et al*, 1979). Briefly, a drop of lactophenol cotton blue stain was placed on a clean glass slide with a portion of mycelium and the cover slip was placed and examined under low and high power magnifications. The culture plates were discarded after incubation at 27°C for four weeks as described by Robert and Pihet (2008).

Clinical Management

The camel was treated with the topical application of a cream containing 2% of Miconazole nitrate (Globe Miconozole nitrate 2% cream Antifungal, Hargraves online Health care), 10% Iodine ointment along with Zinc oxide ointment on alternate days for four weeks. These were applied after removal of skin crusts in the affected areas. Additional supplementation of Vitamin A and mineral mixtures were administered orally daily for four weeks. The animal showed uneventful recovery after four weeks.

Results and Discussion

In present study the direct examination of clinical specimens in 20% potassium hydroxide (KOH) under light microscope revealed delicate blue hyphae with fruiting structure in pale blue background morphologically to dermatophytes (Fig. 2). The fungal colonies were obtained on Sabouraud's dextrose agar with Chloramphenicol and Actidione, and it revealed the presence of characteristic colonies of *Trichophyton* spp. with white to gray coloured, small and button shaped colonies with incubation at 35°C for 72 hours (Ganguly *et al,* 2017a). This confirmed the diagnosis in this study. Haematology values showed no significant abnormalities. The results of this study found that the *Trichophyton* fungus is the causative agent of this camel's skin problem. This was in accordance with the previous reports which observed that the Trychophyton spp. is the most common dermatophyte that affects camels (Wisal *et al,* 2010; Baghza *et al,* 2016;

Fig 1. Greyish-white, round, circular to irregular patches on the neck region of nomadic camel.

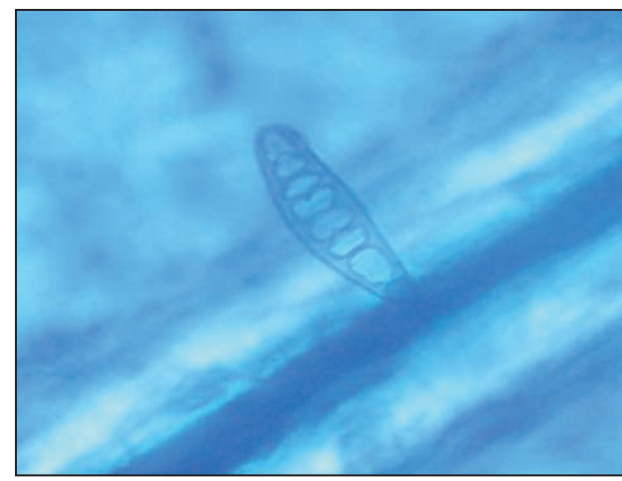

Fig 2. Hyphae of *Trichophyton* spp.

Almuzaini *et al* (2016)). Kuttin *et al* (1986) reported that *Trichophyton* fungi were responsible for the majority of camel's infection since the camels' hair are presented with a suitable substrate for the growth of Trychophyton spp. as compared with human and bovine hair. The main clinical symptoms observed in the affected camel were dry, grayish-white, thick crusty lesions perceptibly raised above the skin on head, neck shoulder, flank and perineal regions. These observations were in accordance with Kuttin *et al* (1986) and Baghza *et al* (2016) who observed that majority of the animals had rounded dark skin lesions on the head, neck and shoulder of the body. Tuteja *et al* (2013) reported the lesions of ring worm with *Trichophyton* spp. which were comparatively dry, hard, crusty, granulomatous and larger in size.

The treatment of this camel with Miconozole cream, 10% Iodine ointment and Zinc oxide ointment along with nutritional supplementation with vitamin A and minerals resulted in early recovery by 7th day of therapy and an uneventful recovery was observed by fourth week. Similar treatment strategies were recorded earlier by Almuzaini *et al* (2016) who reported that *Trichophyton* infected camels were effectively treated with 10% topical iodine ointment and vitamin A supplement. Fowler (2010) also reported that mineral and vitamin supplementation could be administered for effective treatment of ringworm infection. However, use of Miconazole in camels was not reported so far. Possibly the early recovery observed by 7th day of therapy could possibly be due to the inclusion of Miconazole in the treatment. In previous studies the recovery was reported to occur from 20 to 40 days.

In this study the *Trichophyton* was identified from a young camel which was below three years of age. This finding was in concurrence with Al-Ani *et al* (1995), Wisal *et al* (2010) and Ganguly *et al* (2017a) who isolated *Trichophyton* from young camels and observed that they were at greater risk for dermatophytosis. Abdella (2019) reported *Trichophyton verrucosum* is the most common dermatophytes species isolated from camel. Abbas and Omer (2005) quoted that dermatophytosis occurs commonly in young camels while camels above four years of age are apparently immune. The susceptibility of this young camel is probably due to the low immunity of young camels coupled with stress due to its nomadic lifestyle based survival. Naturally camels are exposed to severe stress conditions which render them susceptible to many diseases including fungal disease (Almuzaini *et al,* 2015) and hence in this study, the stress due to continuous usage for performing in circus, frequent transportations and environmental stress, all could have added it. The warm and humid climate prevailing in the study area could also have favoured the growth of these fungal spores. This was in accordance with Shokri and Khosravi (2011) who reported that warm-humid climates are good conditions for sporulation of the fungi and their consequent spread in the environment. Mixed infection with sarcoptic mange is also a predisposing factor for occurrence of dermatophytosis in camel (Al-Salihi *et al,* 2013).

There is very limited number of studies available on dermatophytosis in nomadic camels (Pal, 2016). As dermatophytes has been confirmed as a common cause of ringworm infection in human (Weitzman and Summerbell, 1995), animal handlers may be infected through a direct contact with such infected camel or indirectly through contaminated materials. This needs to be effectively addressed while handling such affected camels.

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INSTRUCTIONS TO CONTRIBUTORS

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Each issue of the Journal of Camel Practice and Research will contain some titbits like My Camel Memories, Clinical Camelids, `**from the old literature', `cartoons' and interesting news** items'. Readers are welcome to contribute for these and due credit lines will suitably be included. However, all these are subject to scrutiny by members of the editorial board.

News of any International Association of Camel or Camelids will be included as and when necessary. `**Research in progress'**, is a special feature we intend to incorporate in the Journal of Camel Practice and Research. In this column researchers can report initial findings of their work in advance, so that others engaged in similar pursuit can exchange views about it. However, such short communications will be entertained on understanding that full article will also appear in this journal.

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For edited symposium/congress/proceedings: Abdalla HS (1992). Camel trypanosomiasis in the Sudan. Proceedings First International Camel Conference, Dubai (UAE), February 2-6, pp 401-403.

Books (Personal authors): Gahlot TK and Chouhan DS (1992). Camel Surgery, Ist Edn. Gyan Prakashan Mandir, Gauri Niwas, 2b5, Pawanpuri, Bikaner, India. pp 37-50.

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Thesis: Rathod Avni (2006).Therapeutic studies on sarcopticosis in camels (*Camelus dromedarius*). Unpublished Masters Thesis (MVSc), Rajasthan Agricultural University, Bikaner, Rajasthan, India.

Commercial booklets: Anonymous/Name (1967). Conray-Contrast Media. IIIrd Edn., 12-15, May and Baker Ltd., Dagenham, Essex, England.

Magazine articles: Taylor D (1985). The Constipated Camel. Reader's Digest. Indian Edn. RDI Print & Publishing (P) Ltd., Mehra House, 250-C, New Cross Road, Worli, Bombay, India. 126:60-64

News paper articles: Anonymous or name of correspondent (1985). Bright Sunlight causes Cataract. Times of India, New Delhi, City-1, India October-9 pp 3, Col 3-5.

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