INFECTIONOUS AGENTS AND NUTRITIONAL FACTORS ASSOCIATED IN INFERTILITY IN SHE-CAMELS

BY

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DEDICATON

This Humble Effort is
Dedicated to:

My Late Supervisor
Dr. Abdu Eldawi Abdalla
Who passed away during last stages of this
Study,
for his genuine guidance and constructive
criticism „God bless his sole„

my late father and mother Asma Awad
and younger brother Al Rasheed
they did their best to lift me up to this stage and
enable me to attend heights of an ideal life and to
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ABSTRACT

During the period from April 2001 to June 2002, Schowak area, east of the Sudan was repeatedly visited and vaginal swabs and blood samples were collected from 55 she-camels suspect to be infertile.

Different bacteria were isolated from 49 (89.1%) of the 55 vaginal swabs. Pure cultures were observed in 46.9 % of the positive samples while 53.1 % gave mixed cultures.

The predominant bacteria isolated were staphylococci (34.5 %) followed by streptococci (25 %), Corynebacterium ( 15.5 %), Bacillus ( 8.3 %), Micrococcus ( 4.8 %), Vibrio ( 3.6 %) and Stomatococcus spp ( 2.4 %). Aerococcus, Clostridium, E.coli, enterococcus and Morganella were isolated from one swab each (1.2 %).

Two species of fungi were isolated from the 55 vaginal swabs (3.6 %). Aspergillus was isolated twice and Mucor once.

Using the Rose Bengal Plate test, Brucella antibodies were detected in the sera of 58.1% of the 55 she – camels investigated.

The RIA of progesterone hormone revealed that a low percentage of the 54 she- camels were pregnant (11.1 %).
Analysis of blood proteins revealed that three animals (5.5 %) had normal values and 52 animals had high values (94.5 %).

Analysis of blood Calcium revealed that of 54 animals (98.2 %) had high values and one animal (1.8 %) had a low value.

Analysis of blood in-organic phosphorus revealed that of the 55 animal, one animal (1.9 %) had a normal value, 96.2 % had high values and only one animal (1.9 %) had a low value and two samples were not suitable for test (3.6%).

Analysis of blood iron revealed 8 % of the samples had normal values, 6 % of the animal examined had high values and 86 % of the animals had low blood iron concentration and five samples were not suitable for test (9.1%).

The causes of infertility are likely to be due to bacterial infections as the major cause followed by protein and minerals excesses or deficiencies and fungal infection.
ملخص الأطروحة
خلال الفترة من أبريل 2001 وحتى يونيو 2002 تمت عدة زيارات لمنطقة الشوك بشرق السودان. أخذت خلالها مسحات مهبلية وعينات دم من 55 ناقة متهمة بانعدام الخصوبة. تم عزل أنواع مختلفة للبكتريا من (89.1%) من المسحات. لوحظت عزلات نقص لبعض البكتريا (46.9%) من العينات الموجبة بينما (53.1%) كانت عزلات مختلطة.
أغلب البكتريا التي تم عزلها من جنس المكورات العنقودية (34.5%) ثم المكورات السببية (25%) ثم البكتريا الودية (15.5%) والمكورات الدقيقة (8.3%) والمكورات الدقيقة (4.8%) والمكورات الضمائية (3.6%) والأنواع (2.4%) والأيروسوكس والملوثات والمورقنيلا والإسكيشياكولاي والمكورات المعوية عزلت مرة واحدة (12%) تم عزل جنسي من الفطر من الـ 55 مسحة مهبلية، الرشاشيات عزلت مرتين (8.8%) بينما فطر الميكور عزل مرة واحدة.
باشر لون الرياح وجدت 58.1% غشاء مصل تحتوي على أجسام مضادة لبكتريا البيروسيلا.
تم تحليل درجة تركيز هرمون البروجسترون في الدم، وجدت نسبة منخفضة من الـ 55 ناقة (11.1%).
تم فحص تركيز البروتين في الدم ووجدت في 3 من النباتات (5.4%) نسبة عادية و في 52 ناقة (94.5%) نسبة مرتفعة.
فحص تركيز الكالسيوم في الدم أفاد بـ 54 ناقة (98.2%) لديها نسبة مرتفعة، بينما حيوان واحد (1.8%) لديه نسبة منخفضة.
تحليل الفسفر الغير عضوي في الدم أفاد بأن حيوان واحد لديه تركيز عادي (1.9%). 98.2% لديها تركيز مرتفع، وحيوان لديه تركيز منخفض (1.9%) ووجدت عينات غير صالحة للفحص (3.6%).
تم تحليل تركيز الحديد في الدم ووجدت 8% من الـ 55 عينة ذات تركيز عادي، 6% ذات تركيز أعلى و 86% ذات تركيز منخفض و وجدت 5 عينات تالفة (9.1%).
أسباب ضعف وانعدام الخصوبة في 88.9% من النباتات التي تم فحصها، أهمها العناوين بالبكتريا يتبعها زيادة أو نقص البروتين والكالسيوم والفسفور وال الحديد ثم العيد أو الفطريات.
INTRODUCTION

The reference to camels in the Old Testament is a clear indication of the normal presence of domesticated camels in the Arabian Peninsula before the time of prophet Abraham, i.e. approximately 1800 B.C. (Gauthier-Pilters, and Dagg, 1981). The camel in the Holy Kur'an is referred to in a number of verses. In Sura vi, the Arabic word “An’am” is used for the cud-chewing animals, were “ibil” (i.e camels) are mentioned along with sheep, goats and cattle. Sura (Al-A’raf) verse 73 refers to the wonderful she-camel belonging to Saleh (or rather Shelah), the son of Arphaxad the son of Sam (or rather Shem), son of Noah. The verse reads:” to the Thamud people we send Saleh, one of their own brethren. He said: O, my people! Worship God, ye have no other god but him. Now has come into you a clear sign from your Lord. This she-camel of God is a sign into you: so leave it to graze on God earth and let it come no harm (The Glorious Kur’an).

In the past two decades and at the onset of desertification and lack of rain-fall most of the traditional livestock in the Sudan suffered or died. The camel was the least to be affected and it survived through the crises.
without many losses which were recorded in other species (Musa et al., 1980).

With increasing human population pressure and declining per capita production of food in Africa there is an urgent need to develop previously marginal resources such as the semi–arid and arid range lands, and optimize their utilization through appropriate livestock production systems of which camel production is certainly the most suitable one (Wilson et al., 1990).

The camel (Camelus dromedarius; one–humped camel dromedary) is an important livestock species uniquely adapted to hot and arid environments. It produces milk, meat, wool, hair and hides, serves for riding, as a beast of burden and as a draft animal for agriculture and short distance transport. It is most numerous in the arid areas of Africa, particularly in the arid lowlands of Eastern Africa, i.e in Somalia, Sudan, Ethiopia, Kenya and Djibouti. Approximately 11.5 million camels exist in this region representing over 80% of Africa and two thirds of the world population of this animal (FAO, 1980).
Estimates of the camel population in Sudan vary between 2.7 million (MAR, 1989) and 3.1 million (FAO, 1988). Abdul Majid (1998), stated that, the total number of camel dromedaries in Sudan is about 2.9 million, that is 16% of one humped camels in the world, 20% of Africa and 24.2% of the Arab world camels. He recorded 30 as the average number of a camel herd; 36% of which are usually males and 64% females, the nomads usually keep 86% of animals in a herd for calf rearing and milking, 8.2% for racing, 2.5% for packing and 3.1% for sale. The bulk of camels are found mainly in Kordofan, Darfur and Butana i.e in the Northern dry lands of the country. Sudan has a large experience in many aspects of camel research, particularly in the field of nutrition, reproductive physiology, meat quality and processing, camel diseases and feeding. Furthermore there is considerable allocation of manpower as well as physical and financial resources to support camel production (Farid, 1981).

The last two decades also saw an increased scientific interest in all matters pertaining to utilization and management of arid lands with substantial number of scientific publications on the camel. According
to Willison et al., (1990) most of these publications covered diseases and veterinary aspects (34%), anatomy and morphology (16%), general physiology (12%) and reproductive physiology (10%). Studies on

feeding and nutrition, Productivity, production systems, camel management and economics are very rare (Ahmed, 1987).

Production is very closely related and integrated with the process of reproduction. It has been widely accepted that without a sound system of production, the reproduction of any species would be jeopardized and arrested.

Diseases are regarded as one of the major constraints which limit camel production and infertility is thought to be an essential and major component of this limitation. Nevertheless, general field observation and a few available surveys indicate that slow reproduction, low lifetime performance of breeding females and high calf mortality are the major constraints of camel Production (Mukasa, 1981).

However, to date there is still not a single project or program in the field focusing mainly on the improvement of practical camel
production in Eastern Africa (Saint-Martin et al., 1990). Despite the fact that no systematic work has been done on the factors involved in she-camel infertility in Sudan, they are considered to be important causes of high economic losses.

**Objectives of the study:**

1- To determine prevalence of infertility in free-range she-camels.

2- To identify the infectious agents associated with infertility.

3- To recognize the nutritional and or hormonal levels associated with infertility.
Infertility could be defined as failure to breed which causes great economical losses. The immediate impact to the individual owner of livestock may not be so apparent compared to certain epizootic diseases, but its effects are still great which include loss due to keeping of barren animals, the absence of offspring's and decline in the milk production beside the cost of treatment (Merck's, 1986).

Generally fertility level of camels is good. According to Bedouin breeders, of every 100 she-camels mated 80 – 90 bring calves and about 1% are sterile. Poor nutrition in seasons of low rainfall and resultant poor grazing is a cause of reduced sexual activity in both sexes. Abortion is rarely seen. Unthriftness due to diseases leads to infertility and may result in abortion (Chen et al., 1985). Musa et al. (1992) observed that fertility of she-camels is maintained throughout life and that through breeding in alternate years, which is the usual practice, a female can yield a total of 12 offspring. However, an average of less than eight seems to be more likely. They recorded that one mating per estrous is usually common and it is possible for a male to serve five or six females in a day. It is said that one
male could suffice 200 females per year, with controlled breeding, but a much smaller number is customary (Elias et al., 1985).

1.1 Physiological aspects of camel reproduction:

Veterinary literature contains many references to the structure and function of the female genital organs of domestic animals, the camel is the only exception and very little attention has been given to the study of its genital tract (Moniem, 1968).

El Wishy (1990) recorded that puberty in female camels is influenced by several factors such as body weight, breed and animal health status. Females become sexually mature around the age of 4 years, therefore giving their first birth around 5 years of age. Sieme et al. (1990) observed that she-camels reach puberty at 2 years but are not usually mated until 3 years of age. Males are sexually active at 3 years. Both sexes breed throughout their lives (Agarwal et al., 1987). In the male, full reproductive power is not developed until six years (Novoa, 1970) or even seven years (El Wishy, 1988) of age. When sexually mature, male camels show an annual rut, roughly from November to July, after which they are sexually quiescent. During the rut the temperament changes towards aggressiveness, less tractile nature, including a predisposition to fight other males and inclination to bite other animals as well as human beings (Xu et al., 1985).
1.1.1 Breeding season and oestrus cycle:

A zootechnical short term survey carried out in Butana (Sudan) has shown six types of camel herds defined with a multivariable analysis of the principal component of the livestock farming systems. Their reproductive performance was compared. The average age at first parturition was six years and eight months and the fertility rate was approximately 35% and 70% of the births occurred from May to August. The average interval between births was 28.5 months and deceased with increase of rank of gestation. The typology was analyzed with a view to identify the factors influencing reproductive performance (Saint-Martin et al., 1990).

Many investigators consider the dromedary camel as a seasonal polyoestrus animal (Shalash, 1965 and Musa and Abu Sineina, 1978). The period of the oestrus in she-camel is easily recognizable by general restlessness, often aggressiveness in manner, and by swelling and discharges from the vulva. The length of oestrous cycle is normally 2 – 3 weeks, although in the Bactrian camel the period can extend to 30 - 40 days (Yasin and Wahid, 1957).

Rai et al. (1990) initiated breeding at an early age using FSH as the predominant hormone. About 65-80% of the adult body weight was achieved by the Bikaneri heifers at 2-3 years of age. Nine prepubertal
animals were injected with 1000 and 2000 IU of folligon for 2 or 3 consecutive days while three saline treated animals served as control. Oestrus was induced in all the treated animals 3 to 5 days post-treatment and all were successfully mated. Four animals in the treated group settled after first service while the rest of the animals conceived pregnancy up to the full term. The elevated progesterone level (8-12 ng) in the conceived animals was maintained up to 22 days post mating. The progesterone level remained low throughout the cycle in anovulatory heats (<= 1 µg) however in the controls progesterone persisted at the basal level. The gestation period for the treated animal was 382.6 ± 6.31 days against the herd average of 379.3 ± 3.46 days. The average birth weights of the calves born out of the treated heifers was lower (27.2 ± 0.58 kg) as compared to the herd average 37.0 ± 1.34 kg).

With regard to oestrus cycle, Leese (1927), pointed out that the female camel comes on heat 3 – 4 week after calving. He described the external signs of heat as restlessness, bleating, swelling of the vulva and vulva discharge. Barmintsev (1951) described the reproduction of Bactrian camel and pointed the lack of precise information on the exact moment of ovulation during the rut heat, and on the life span of the spermatozoa in the genital passage of the female. He outlined also the differences of opinion on
whether ovulation is induced by the act of mating. Mares (1954) observed that the male camel mates the female whether the later is on heat or not. Yasin and Abdel Wahid (1957) stated that camels of Pakistan breed from December to March and that oestrus is expressed by restlessness, swelling and discharge from vulva with a bleating noise. Abdalla (1960) reported that the dromedary is a ploy – oestrous animal with a tendency to breed in the raining season. He also stated that in the oestrous cycle in the camel there is a luteal phase in which there is a corpus luteum, which is smaller than that of pregnancy. Marshall (1962) described the camel as a polyoestrous animal with a cycle of 2 – 3 weeks and that heat lasts 3 – 7 days. Marshal pointed out that the foal heat takes place 1 - 3 days after parturition.

Although the oestrus cycle has been studied in many species, including cow (Quinland et al., 1939; Nalbandov and Casida, 1942; Brown, 1944; Hancock, 1948; Trum, 1950 and Arthur, 1964), bitch (Evans and Cole, 1931), cat (Liche, 1939), rabbit (Hafez, 1962), the camel seems to have received little or no attention. Ovarian cycle activity was fully described by Musa, (1980). He reported a 28-day cycles in which follicles matured in six days maintained this size for 13 days, then regressed in eight days, but there was no spontaneous ovulation in the camel (Chen Yuen and Pan 1979., Musa, 1980., Shalash, 1965). Thus, without mating there will be
no luteal phase. The highest ovarian activity of camel in central Sudan occurs from March to August. During the rest of the year the ovaries remain inactive or show a limited number of small follicles (Musa and Abusinea, 1978). They also recorded that manual stimulation of the cervix for 15 minuets did not induce ovulation, although lutenization of mature graafian follicle occurred.

Ovulation occurs 30-48 hours following copulation (Chen Yuen and Pan, 1979). Shalash (1965) stated that, without programming there was no formation of corpus luteum. The size of the corpus luteum depends on the ovarian activity (Musa and Abusinea, 1978). The corpus luteum becomes large and lasts longer when mating occurs at the time of maximum follicular development. When mating takes place later, the corpus luteum becomes smaller and disappears in a short period of time. In Beersheva (Israel) research was carried out using radioimmunoassay of sexual hormones in the blood of the female. There was an increase in oestradiol activity from the beginning of December, which ended toward the beginning of April. Surprisingly enough, there were also peaks of progesterone activity, although no male was present. The peaks in hormone activity were 23 days apart. From the middle of January there were peaks of oestradiol activity every 7 days accompanied by peaks, of progesterone 2 days later. Twenty-
four to 48 hours following mating, the luteanizing hormone (LH) appeared. The LH then declined steadily for 6 days and a second peak, even greater than the first, was found almost two weeks later. At that time, the progesterone level was extremely high.

Nalbandov and Casida (1942) stated that the oestrogen levels were low, but both hormones showed peaks in activity every 4 days even when LH activity was non-existent. These 4 days fluctuations continued almost right through the pregnancy. While undertaking physiological research both on body fluids and renal function, two camels aborted. In these camels the hormone activity declined to base-line levels.

Oestrus has been known to re-occur a day after calving (Barmintsev, 1951). If the camel is well fed, oestrous can occur within one-month post-partum (Mares, 1954; Yasin and Wahid, 1957). If the camel has no milk, then oestrus occurs within 28 days (Evans and Powys, 1979). This means that in good feeding conditions camels can be mated as soon as the young calves start grazing.

Yasin and Wahid (1957) reported a gestation of 365 to 395 days whereas Evans and Powys (1979) recorded a gestation period of 373 to 393 days. The gestation period of Bactrian camel was 402 days according to Chen Yuen and Pan (1979).
1.2 Factors involved and responsible for infertility:

Black’s (1976) classified the causes of infertility into five categories, which include: Nutritional level, environmental and managemental conditions, disease of the genital tract of both male and female, hereditary (e.g. congenital deformities) and physical and psychological inability or disturbances.

Over the last 40 – 50 years there has been a noticeable change in the causes of infertility in cattle in many parts of the world. The recognition of *Trichomonas fetus* infection (Stableforth et al., 1937) and *Compylobacter fetus* infection (Sjollema et al., 1949) as causes of widespread infertility constituted major advances. Control measures, in particular the widespread use of artificial insemination, have largely eliminated these diseases from the U.K although world – wide they are both important causes of infertility (Phillipo et al., 1982). The eradication programmes for bovine tuberculosis and brucellosis have reduced the importance of both of these diseases as causes of reproduction loss (Reid et al., 1979). Although non – specific infection due to opportunist pathogens are still important, by far the greatest cause of infertility is poor management of herds. At the same time, the demands put upon the dairy cows to produce more milk and the genetic selection for high yield have inevitably resulted in functional aberrations of
the reproductive and endocrine systems, therefore, the various causes of cattle infertility can be discussed under these main heading: anatomical factors, functional abnormalities and managemental problems (Butler and Smith, 1989). These causes also apply to camels.

1.2.1 Anatomical factors affecting fertility:

1.2.1.1 Congenital abnormalities:

Both congenital and acquired abnormalities of the genital system can influence fertility. The latter types are more frequently encountered, as demonstrated in a survey by Kessy (1978) who found that in 2000 genital tracts that were examined from abattoirs only six specimen (0.3%) had evidence of congenital abnormalities compared with 194 (9.65%) with acquired lesions. Since most of the latter were identified in the tracts from parous specimens the importance of conditions that might occur during pregnancy and especially at parturition and during the puerperium, are demonstrated (Afiefy et al., 1973).

EI-Wishy (1990) observed gross abnormalities in she-camels in 2.7%, 30 out of 1120 genital organs. They included ovarian cysts (1.0%), teratomas (0.4%), ovarian hypoplasia (0.1%), mucometra (0.8%) and pyometra (0.5%). The non-functional ovaries observed in most (61.0%) of
the other organs might account for delayed puberty, long post-partum anoestrus and protracted calving intervals in dromedary camels of Egypt.

Ovarian agenesis in which one or both ovaries may be absent, in these cases the genital tract is infantile and cyclical behavior is absent. An apparently hereditary condition of "virtual absence of ovaries" was seen by Fincher (1946) in three maternal half–sister heifers. When one or both ovaries are small, narrow and functionless this condition is called ovarian hypoplasia. Gonadal hypoplasia has been shown to affect both males and females of the Swedish highland cattle breed, and among 8145 cows Lagerlof (1939) found an incidence of 13.1% of the affected cows, 87.1% had hypoplasia of the left every, 4.3% of the right and 8.6% of both ovaries. Where both ovaries were hypoplastic the genital tract was infantile and oestrous cycle did not occur. Eriksson (1938) attributed this affection to an inherited autosomal recessive gene.

In Kessy's survey (1978) the uterine tube was the most frequent site identified as having congenital defects. Unilateral aplasia was identified in 0.1% of the specimens, duplication of the tube in 0.05% and segmental aplasia in 0.05%. Several other abnormalities were also identified which could not definitely be assumed to be congenital; 0.3% of the genital tracts from multiparous individuals had unilaterally or bilaterally occluded uterine
tubes whilst one specimen showed evidence of hydrosalpinx (Hafez, 1975). Freemartinism is a distinct form of intersexuality, which arises as a result of vascular anastomosis of the adjacent chorio-allantoic sacs of heterozygous fetuses in multiple pregnancies (Lillie, 1916). As a result, although the external genitalia of freemartin heifers appear normal the internal genitalia frequently show masculinization. In extreme cases the gonads resemble testes, though spermatogenesis is not apparent and there are well-developed epididymides, vasa deferentia and vesicular glands (Short et al., 1969). However in the least affected cases the female genital tract may be small, with a persistent hymen and hypoplastic ovaries (Wijeratre et al., 1977), whilst there are also reports of one freemartin heifers which showed signs of oestrus and the presence of a corpus luteum on the ovary (Wilkes et al., 1981), although there are doubts whether this was a true freemartin, or rather a congenitally malformed tubular genial tract (Arthur, 1959). It is generally assumed that 92% of heifers, which are born as co – twins to bulls, are sterile (Biggers and McFeely, 1966). Vascular anastomosis occurs as early as 30 days of gestation; thus if there is death of the male twin of heterozygous pair after this time with the other being carried to term, it is possible for a single – born free martin to occur. This has been demonstrated
as a cause of infertility in heifers with apparently normal external genitalia but with sex chromosomes chimerism (Wijeratre et al., 1977).

### 1.2.1.2 Acquired abnormalities:

Since the early survey conducted by Carpenter et al. (1921), which showed that 15.3% of cows, which were examined at routine clinical work, had lesions of the uterine tubes and adnexa, many similar studies have confirmed their high frequency of occurrence. The percentage incidence ranged from 0.95% in an abattoir study in Australia (summer, 1974) to 100% in a similar study in Egypt (Afiefy et al., 1973). Adhesion of the uterus a troublesome sequel to the caesarean operation is adhesion of the uterus to the omentum, intestines or abdominal wall.

A similar lesion may follow uterine rupture. Such lesions may accompany ovariobursal disease and may follow tardy involution of the uterus and metritis. They are frequently associated with sterility (Andreson and Davis, 1958).

Parturient trauma of the tubular genital tracts occurs due to dystocia due to fetal over-size which commonly occurs in cattle, particularly in the Friesian breed. Delivery of large calves by heavy traction frequently damages the birth canal to such an extent that the animal is rendered sterile (Mcleod, 1990). A third degree perineal rupture may occur at calving,
usually as a result of dystocia; the whole thickness of the vagina and rectal wall ruptures so that the rectum and vagina are confluent. The cow thus has a cloaca. This lesion does not heals; thus air and faces are aspirated into the vagina causing vaginitis and metritis. Affected cows have a chronic mucopurulent vulval discharge but the general health is not impaired; normal cycling resumes but conception does not occur because of the metritis (Lamming, 1978).

Granulosa cell tumors are the commonest neoplasms of the bovine ovary but carcinomas, fibromas, thecomas and sarcomas have also been described (AL – Dahash and David, 1977). Most of the large and cystic neoplasms of the bovine ovary are granulosa cell tumors. They have been seen in pregnant as well as non-pregnant cattle. In the early stage of tumor it presumably secretes oestrogens, for the affected animal is often nymphomaniacal (McGilliard and Polan, 1983). The non-affected ovary is usually of the anoestrus type. Arthur (1998) saw a Friesian cow with a granulosa cell tumor, which weighted 24 kg; the cow showed successive phases of nymphomania, anoestrous and virilism.

Tumors of the uterus are rare in cattle, although in the USA lymphosarcoma of the uterus is not uncommon. In all countries leiomyomata are sometimes seen; pregnancy may occur in the neoplastic uterus. The
larger uterine tumor may be confused on rectal palpation with a mummified fetus. A 2-year abattoir survey in Denver by Anderson and Davis (1958) revealed 24% of the cattle tumors (excluding cancer of the eye) to be in the genitalia; the latter were classified as follow; adeno-carcinoma of the uterus, 26 cases, lymphosarcoma of the uterus six; leiomyoma of the uterus, four; granulosa cell tumors of the ovary, six; cystadenoma of the ovary, one; squamous epithelioma of the vulva, one.

The relative frequency of the uterine adenocarcinoma in this series and its rarity in Britain are noteworthy (Ball et al., 1983). Tumors of the cervix are rare in cattle and the few recorded have been benign. Fibropapillomata of the vagina and vulva of cattle are not uncommon, they do not cause infertility but may interfere with birth. They are usually pedunculated and may be removed surgically. There is a possibility that one form of vaginal fibropapilloma is of viral origin and that is transmitted venerealy. It occurs in young cattle and undergoes spontaneous resolution (Beynon, 1978).

1.2.2 Functional forms of infertility:

As a rule the functional forms of infertility tend to affect individual animals within a herd but in the aggregate they constitute an important cause of infertility; and when they affect a large number of a particular subgroup in
a herd they frequently effect some other problems especially nutrition (Drew, 1978). Most functional aberrations occur because of some endocrinological abnormalities that are frequently difficult to specify even with current methods of hormone assay, particularly when single, spot samples of blood or milk are examined. The abnormalities occur as a result of inherited factors; nutritional deficiencies or excesses; social influences, which may arise from modern husbandry methods, for instance the grouping of large numbers of cows thus interfering with establishment of stable social hierarchy; and the stress of production (Morris, 1976).

Ali et al. (1992) detected infundibular cysts in she-camels by using the ultrasound probe scanner (WIC 50, with 5m Hz Scan head, USA) coupled with rectal palpation. Their morphological, histological, physiochemical and bacteriological study of the infundibular cysts revealed, funnel-shaped infundibulum 10-28 cm in width, 18-50 cm in length, the content was fluid and had different colors (clear, yellowish, light or dark brown). The amount of fluid collected from each cyst ranged between 300 and 3.800ml. Twenty four out of 30 cyst-fluids (80%) gave pure growth of a single type of bacterial colonies. The organism were identified an Aeromonas hydrophila according to their biochemical properties.
Only little information is available on the incidence of *Aeromonas hydrophila* in mammals, except in man (Gray, 1984). The organism isolated from faeces of some domestic animals: cow, buffalo, Goat and chickens. The only single report in the literature concerning the isolation of *A. hydrophila* from camels is that of Gameel *et al.* (1986). The organism was isolated in association with *Clostridium perfringens* type A and *Clostridium sordelli*. The histological and morphological changes of the cyst proved that there were dilatations of the cranial part of the infundibular wall hence designated as "infundibular cysts". The prevalence of such cysts was fairly high in the camels examined and coincided with the increased incidence of infertility observed in the camels brought to the Veterinary Teaching Hospital in Saudi Arabia (Annual report, 1989).

**1.2.3 Hormonal factors involved in infertility:**

Cristofori and Quaranto (1990) evaluated oestradiol –17B and progesterone levels in 197 female camels (66 were pregnant). They found significant difference (P< 0.01) in progesterone levels between pregnant and non-pregnant she-camels. Significant differences (P< 0.01) were also recorded in oestradiol -17B levels between anoestrous and cyclic subjects and between she-camels in different cycle stages.
Factors affecting reproductive performance were studied by Abdel Rahim and Al-Nazier (1990) in three camel farms under semi-intensive system of management. Records were kept for 150 breeding she-camels during three consecutive breeding seasons. Progesterone hormone was measured in milk samples taken during mating and for 5 days. Nutritional status of the animals was scored at calving using external body features as condition scores I, II, III and IV. First post-partum oestrus was noticed within 2 weeks after calving with a mean of 29±12 days. Conception rate was high (85%). Mating was always successful when females accepted the male willingly. This was clearly shown by low progesterone values and indicated the presence of mature follicle. There was also a significant relationship between body condition at calving and conception rate. Animals above condition score III had higher conception rates. Accordingly organization of feeding is recommended so that animals are in the proper condition at calving.

There is much conflicting evidence concerning the relationship between hormonal imbalance and infertility, probably influenced to some extent by the errors inherent in measuring hormone concentration in a limited numbers of peripheral blood samples. It is known that the rate of transport of the oocyte and zygote along the uterine tube is under the
influence of oestrogen and progesterone (Whitney and Burdick, 1936, 1938). Thus if there is an imbalance of these hormones there may be accelerated or retarded passage of the zygote, so that it reaches the uterus at a time when the environment is hostile to its survival. It has also been known for some time that in order to have good embryo survival after embryo transfer the recipients and donors oestrous cycles must be synchronized within one day of each other (Rowson et al., 1969; Newcombe and Rowson, 1975).

The development of radioimmunoassay (RIA) techniques recently has enabled analyst's to detect and measure analytes present in very low concentration in body fluids (Mile et al., 1974). This technique has been adopted in Sudan as a diagnostic tool since 1968. The first and the most important reagent in RIA to be considered is the antibody production, as high affinity and specificity antibodies are the backbone of RIA technique (Ali, 2001).

Progesterone is one of the female principle sex hormones normally secreted by the ovaries and placenta. The adrenal cortex also secretes small amounts of estrogens and progesterone. The metabolite of progestrone is pregnanediol, which is conjugated as glucouronates prior to excretion in the urine (Abdulla and Abbas, 2000).
Rivarola (1973) stated that the daily progesterone levels are considered the most accurate means for documenting a defective luteal phase. Measurements of serum progesterone have also been used to check the effectiveness of ovulation induction, to monitor progesterone replacement therapy and to detect and evaluate patients at risk for abortion during the early weeks of pregnancy.

Determination of plasma or salivary (progesterone) has almost completely replaced measurement of urinary pregnanediol. They may be used for the following purposes:

1. To indicate the development of functional corpus luteum, after spontaneous ovulation or occurring as a result of ovarian stimulation, nevertheless in the she-camel there is complete absence of the luteal phase and progertrone can not be detected unless pregnancy occurs (Arthur, 1998). This difficulty was overcome through special forms designed for this study (Appendix 1).

2. To investigate recurrent abortion.

3. To investigate diseases of the adrenal cortex.

Excessive formation of progesterone and excretion of preganaediol occurs in a small percentage of adrenal tumors (Walton et al., 1984).
Khanna, (1988) analyzed sera of Bikaneri breed of camels for testosterone, progesterone, estradiol, thyroxine (T₄) and triiodothyronine (T₃) using RIA. The results indicated that the breeding season had a marked influence on the hormonal level. He observed that the steroid hormones were significantly higher during the rutting season whereas thyroid hormones were higher during the non-rutting season, but T₄:T₃ were almost double during the rutting season. The hormonal studies on pregnant females revealed that the females bearing male calve had relatively lower estradiol (76.5 ± 10.8 pg/ml) than those carrying a female fetus (112.3 ± 19.6 pg/ml). On the other hand, he stated that progesterone level was slightly higher in dams carrying a male fetus (5.13 ± 0.69 ng/ml) than in those carrying a female fetus (4.45 ± 0.28 µg/ml).

An experiment was made on nine camel heifers, aged 2-3 years, concerning the effect of FSH on lowering the age at first calving. It was revealed that, after injection of FSH, all heifers exhibited oestrus and were successfully mated. However, full-term pregnancy was carried in only five animals (Benirschke, 1967).

Corah et al. (1974) recorded elevation of plasma progesterone concentration in beef heifers that failed to conceive, in cows that were infused with ACTH (Gwazdauskas et al., 1979) and in Guernsey cows
subjected to high environmental temperature (Abilay et al., 1975). Abnormal plasma oestrogen concentrations on days 3 and 4 of the oestrous cycle have been observed in cows with abnormal embryos (Ayalon, 1973).

Erb et al. (1976) found higher plasma oestrogen concentration 8 and 12 hours after oestrous in fertile cows compared with others. Some workers have found a positive relationship between luteal phase progesterone concentration in the peripheral blood or milk and embryo survival, whilst others have found no such relationship. Kubasik (1984) recorded that progesterone is secreted by the ovaries in very small amounts during the follicular phase of the menstrual cycle. Progesterone levels increase sharply during the luteal phase of menstrual cycle, reaching a maximum some 5 to 10 days after the midcycle LH peak. If fertilization does not occur, a steep decline to follicular levels sets in about 4 days before the next menstrual period. If implantation of fertilized ovum occurs, the progesterone concentration continues to rise.

Azouz et al. (1992) measured serum FSH, LH concentration, serum prolactin, serum cortisol and serum testosterone concentrations in male camels. They observed a significant decrease in Prolactin levels compared to large rise in gonadotropin (FSH and LH), testosterone and cortisol levels, in male camels during the rutting season. Hyper- prolactinaemia was found to
decrease testosterone in male rats (Ahmed et al., 1989) and to produce degenerative changes in their testes and secondary sex glands (Taha, 1988; Seifelnasr et al., 1989). Further, McNeilly, (1978) observed that induced hyperprolactinaemia in male rats caused a prolonged suppression of FSH and LH concentration in the serum and in pituitary tissue. Ismail et al. (1984) found high levels of prolactin in the blood of male dromedary camels in the nonbreeding season, it seems that hyperprolactinaemia, which is accompanied by decreased serum levels of FSH, LH, testosterone and cortisol, is likely to be the main cause of low fertility during the non-breeding season.

1.2.4 Managemental factors involved in infertility:

1.2.4.1 Nutrition:

The usual habitat of the camel is not only characterized by high temperatures and scarcity of water, but in consequence of these environmental conditions also by a considerable seasonal variation in available forage quantity and forage quality. Herbivores can adapt to such fluctuations in forage quality by either increased selectivity for high quality plant material or by more efficient digestion for poor quality materials. The camel can do both (Mason, 1984).
In a series of comparative studies carried out with domestic ruminants, camels and donkeys on a semi-arid thornbush savannah in Isiolo District in Kenya various adaptive mechanisms to fluctuating forage supply were investigated (Engelhardt et al., 1988). Feeding behavior and dietary preferences were the most important adaptive mechanisms investigated. All livestock species ingest higher numbers of forage species during the growing season than during the dry season. Goats, followed by camels accept the highest number of forage species resulting in a more even and thus benign utilization of the available vegetations. Local zebus and donkeys in comparison specialize on a very limited range which can easily lead to overgrazing of certain species (King, 1983). The protein content of the diets selected by all animals was higher, and correspondingly the crude fiber content was lower, during the green season than during the dry season. Cattle were consistently selecting the poorest quality with a marked quality change between seasons. Camels were consistently able to select best qualities with minor differences between seasons.

Donkeys, sheep and goats, in that order, took intermediate positions. Camels, but also goats, can be classified as concentrate selector according to Hofmann (1988), if the pasture permits such feeding behaviour. However, studies carried out with camels in the Butana grasslands in Sudan showed
that camels adopt feeding preference almost identical to those of cattle, if
grasses are the only forage available (Zeuner, 1963).

Rossiter (1970) observed that inadequate diet delays puberty, causes
unoestrus and depresses ovarian function in older animals. In rural areas the
cyclic gain of weight with onset of rain and its loss in dry season continues
to be a major limitation to livestock production. He also recorded especial
form of infertility in she-camel, which graze on pasture dominated by
subterranean clover. The infertility is caused by the high content of
oestrogenic substances in the leaves of that plant. He recorded that the
isoflavones or phytoestrogens, which occur, it is formononetin, which is the
most biologically active, and the risk of a pasture can be determined by its
chemical assay. The various strains of clover vary greatly in their
oestrogenic activity. Thus Dinninup, Dwalganup and Yarloop are very active
while Clare, Mt Barker, Bacchus Marsh, Daliak, Northam A and
Woogenellup are poorly active and Geraldton occupies an intermediate
position (Abu Sin, 1982).

1.2.4.2 Deficiency of energy and protein:

Protein and energy deficiencies usually occur concurrently in under
fed livestock and often cannot be strictly separated (Jacobson, 1978).
Deficiency of energy is the most common nutrient factor, which limits
The clinical finding in an energy deficient animal will depend on the age of the animal, whether or not it is pregnant or in lactation, the presence of concurrent deficiencies of other nutrients, and environmental influences. Insufficient supply of energy in the young animal’s results in retarded growth and delay in onset of puberty. In mature animal there will be a marked decline in milk production and a shortened lactation.

Oetzel (1986) recorded that prolonged energy deficiency in pregnant beef heifers resulted in a failure to produce adequate quantities of colostrum at parturition. In mature animals there was also a marked loss of body weight especially during high demand for energy as in late pregnancy and early lactation. There was prolonged period of anoestrum lasting up to several months which had a marked effect on reproductive performance in the herd. Prolonged deficiency of energy during late gestation may result in under-sized, weak neonates with a high mortality rate (Oetzel, 1985). The normal value of blood protein in camels is $5.72 \pm 0.026$ g/dl (Sawsan, 1995).

1.2.5 Mineral deficiencies:

Little work has been done on mineral supplementation in camels. Copper is deficient in animal tissue, soil and pasture in Western Sudan except Bahar El Arab area. Phosphorus becomes a limiting nutrient with
advance of dry season in Baggara region in western Sudan (Ahmed, 1976). Deficiency of any dietary constituent which is necessary for normal metabolism such as iron, copper, cobalt, manganese, iodine, phosphorus and Vit. A is a possible cause of unthriftiness and hence anoestrus. Under British farming conditions, deficiency of copper is the most likely dietary cause of anoestrus (Arthur, 1964).

A number of environmental factors affect the concentration of the important isoflavones in the pasture. They are much higher when the soil is deficient in phosphorus. In the search for other factors affecting oestrogenic potency, it has been observed that clover leaves that are entirely red or have red margins have much higher content of oestrogenic isoflavones than green leaves (Thain and Robinson, 1968). It is assumed that the leaf redness is due to viral infection.

Clover is most potent oestrogenically in spring, and sheep eating a lot of the plant at this time can become temporarily infertile, but are normally fertile again by the usual breeding season in autumn. However, ingestion of the plant in several successive years causes permanent clover disease – infertility from which ewes do not recover (Ludewig, 1973). The disease is important only in sheep. Cattle are generally considered to be unaffected but
the subject is still controversial as evidence is against cattle being affected. Horses appear to be able to graze the toxic pastures without ill-effects.

1.2.5.1 Manganese deficiency:

Dietary deficiency of manganese is thought to cause infertility and skeletal deformities both congenitally and acquired. In cattle the common syndromes are infertility, calves with congenital limb deformities (Dyers and Rojas, 1965) and calves, which manifest poor growth, dry coat and loss of coat colour. Egan (1972) described a manganese responsive infertility in ewes, as it is well known in cattle. In cattle it is manifested by slowness to exhibit oestrus, and failure to conceive, often accompanied by subnormal size of one or both ovaries. Sub-oestrus and weak oestrus have also been observed. In pigs experimental diets low in manganese cause reduction in skeletal growth, muscle weakness, obesity, and irregular, diminished or absent oestrus, agalactia and resorption of fetuses or the birth of still born piglets (Plumlee, 1956).

1.2.5.2 Calcium deficiency:

Hignett (1956) proposed that functional infertility occurred in cattle on diets with calcium to phosphorous ration outside the range of 1:2 to 2:1 this might have been correct if high calcium to phosphorous intakes dietary reduced manganese (copper or iodine) availability in diets marginally
deficient in one or other of these elements. He recorded that; pigs fed on heavy concentrate rations might have developed a hypocalcemic tetany, which responded to treatment with calcium salts. Tetany may also occur in young growing cattle in some circumstance. Inappetence, stiffness, tendency of bones to fracture, disinclination to stand, difficult parturition, reduction of milk flow, loss of condition and reduced fertility are all non-specific signs recorded in adults. The normal value of blood calcium in camels is $2.64 \pm 0.08 \text{ mmol/l}$ (Swasan, 1995).

1.2.5.3 Phosphorus deficiency:

It is characterized by pica, poor growth, and infertility and in the later stages, oesteodystrophy. Phosphorus deficiency under field conditions may be exacerbated by deficiency of Vit. D and possibly by an excess of calcium. Experimentally large doses of vitamin A decrease the absorption of phosphors in cattle, and this may contribute to the development of nutritional oesteodystrophies (Manston, 1964).

Retarded growth, low milk yield and reduced fertility are the earliest sign of phosphors deficiency. For example, severe phosphors deficiency in range beef cattle dropped the calving percentage from 70% to 20%. However, it was claimed that relative infertility occurred in dairy heifers on daily intakes of less than 40 g of phosphate. The infertility was accompanied
by anoestrus and irregular oestrus and delayed sexual maturity (Morrow, 1967). This was not in agreement with Littlejohn and Lewis, (1960) who showed that fertility was independent of the calcium or phosphorus content or the calcium, phosphorus ratio in the diet given to cattle.

Hart and Mitchell (1965) stated that, the effects of mal-nutrition on fertility were likely to be general and that infertility might often be related to lack of total average intake rather than to specific deficiency. The experimental production of phosphorus deficiency in beef cows indicated that several months on a deficient diet were necessary before clinical signs developed. The clinical signs included general unthriftness, marked body weight loss, reduced feed consumption reluctance to move, abnormal stance, bone fractures and finally impaired reproduction (Call, 1986). The normal value of blood inorganic phosphorus in camels is $1.78 \pm 0.07$ m mol/l (Sawsan., 1995).

1.2.6. Infectious forms of infertility:

The uterus must be essentially sterile for optimal conception to occur (Fitch and Bishop, 1932). (Harden, 1958). The general concession is that the presence of microorganisms in the uterine mucosa alters the uterine pH and is inimical for the survival of spermatozoan in the uterine lumen. If the organisms or their metabolites are present for a long time they cause
endometritis or cervicitis and prevent the implantation of the fertilized ovum in the uterine bed. It has been reported that bacteria contributed to a great extent to reproductive failure (Fitch and Bishop, 1932). Dowson (1960), stated that the organisms isolated from healthy cows, in a descending order of frequency were; *staphylococcus spp, Escherichia coli, streptococcus spp.*, *Anthracoids (bacillus), Proteus, Pseudomonas* and *Neisseria spp*. They reported that cervical mucus was collected from 106 repeat breeder cows during oestrus and were subjected to bacteriological examination; out of which 86 yielded 199 species of bacteria (81.13%). Bacteria which were isolated frequently and which were of significance included; *Actinomyces pyogenes, Staph. aureus, Pseudomonas aeruginosa* and haemolytic *E. Coli*. These organisms were incriminated as potential pathogens which render female genital tract harmful to the viability of sperms (Gunter et al., 1955). In a quantitative study of bacteria from normal sterile and repeat breeding cows, he observed that 22 out of 25 types of bacterial isolates were common for both groups of animals.

Al Sayed (1994) reported a single isolate of *Haemophilus somnus*, two isolates of *Staph. aureus* and *Bacillus subtilis* in the repeat breeder group, the exceptional organisms were from the family *Enterobacteriae* and the genus *Corynebacterium (Actinomyces)* that occurred in a significantly
higher rate in the repeat breeder group. Predominant among both groups were organisms of the genera, *Micrococcus, Staphylococcus, Escherichia, Bacillus* and *Actinomyces (Corynebacterium)*. Organism of the genera *Branhamella, Actinobacillus, Pasteurella, klebsiella, Haemo-phillus, Proteus* and *Kurthia* constituted a minority of the isolates.

Al Sayed *et al.* (1994) studied the relationship between the presence of certain microorganisms in the uterus; cervix and placenta of cows and subsequent infertility problems, using cervical swabs, uterine discharges and washes as specimen, they isolated *E. coli, Staph. aureus, Rhizopus spp., Aspergillus niger, Actinomyces pyogenes, Staph. epidermidis, Listeria monocytogenes, Aspergillus fumigatus, Aspergillus flavus, Mucor* and *Candida albicans*.

Wernery and Wernery (1992) studied the uterine bacterial flora of 80 barren camels in two herds bred for racing in Dubai – some with and some without endometritis. Their main findings were: (i) the range of organisms isolated was very similar to those obtained from equine and bovine uteri except that *Streptococcus zooepidemicus* which is the commonest pathogen recovered from equine uteri and the organism of contagious equine metritis - *Taylorella equigeitalis*- were not found. *Trichomonas* and *Campylobacter*
fetus were both isolated from infertile camels (Yagil and Van Creveld, 1990).

1.2.6.1 Puerperal metritis:

Albrechtsein (1917) and Nielsen (1924) believed that the primary cause of infertility in camels was infections of the tubular portion of the genital tract, particularly the uterus with pyogenic organisms and that ovarian changes were secondary. Consequently, the main feature of their treatment comprised medication of the uterus. Puerperal metritis occurs within a few days of parturition. It usually follows an abnormal first or second stage of labour. It is associated with uterine inertia and is frequently accompanied by retention of the fetal membranes. The infecting organisms are *A. pyogenes*, group *C streptococci*, hemolytic *Staphylococcus*, *coliforms*, and Gram-negative anaerobes particularly *Bacteroides spp*. exceptionally, *clostridia* may be present and soon they produce serious, and often fatal disease. The bacterial invaders colonize the non-involuted uterus whence their toxins are absorbed and cause severe conditions, like septicemia and pyaemia. The uterus may contain a large volume of toxic exudates (Ahktar *et al.*, 1993).
**1.2.6.2 Endometritis:**

In 1941 the Survey Committee of the National Veterinary Medical Association of Great Britain and Ireland (now the British Veterinary Association) came to the conclusion that 90% of infertility affecting dairy cows in Great Britain was due to varying degrees of endometritis. Wright (1945) working at the Liverpool school, did not subscribe to this view and later research supported their contentions that chronic catarrhal or mucopurulent endometritis is an uncommon cause of infertility. Also in a bacteriological study of the uteri of infertile cows, Keisal and Dacres (1959) isolated bacteria from the uteri of infertile cows in 28.1 percent of 1359 attempts and were of the opinion that many of these bacteria were opportunists and transient in nature.

Hess and his colleagues (1909) believed that ovarian abnormalities were the primary cause of common acquired bovine infertility. In 1972 Hammond made an outstanding contribution to the science of cattle breeding by his analysis of normal reproduction in the cow. This fundamental study provided satisfactory basis for subsequent investigation of infertility out of which came considerable support to the Swiss view that many common abnormalities of reproduction were primarily due to ovarian dysfunction.
Andriamanga et al. (1984) stated that, inflammation of the endometrim is a common condition in the cow, and it has a profound effect upon the fertility of the animal but does not affect its general health. They observed that the causal organism usually reaches the uterus from the vagina at coitus, insemination, parturition or postpartum, although it is possible in some circumstances for an infection to arrive by the circulation. Specific pathogens such as *C. fetus* and *Trichomonas fætus* affect fertility because, having entered the genital tract at coitus, they cause endometritis. In the U.K and Northern Europe the non–specific, opportunist pathogens are the most important causes of endometritis and have a significant effect upon fertility (Anon, 1992).

Endometritis influences fertility in two ways, first in the short term, it reduces fertility by extending the calving to conception interval and increasing the number of services per pregnancy and, secondly, in the long term, it can result in sterility due to irreversible changes of the genital tract. With regard to the short term influence, Studer and Morrow (1978) found a significant correlation between the state of the uterus, as determined by rectal palpation, and the calving – conception interval, especially, in relation to the amount of pus in the discharges.
In Egypt Hegazy, Youseff and Selim (1979) made a bacteriological and histopathological study of cases of endometritis. The range of bacteria they found included the following *Proteus* spp., *Serratia* spp., *Enterobacter* spp., *Klebsiella* spp., *E. coli*, enterococci, *Bacillus* spp., *C. renale*, *A. pyogenes*, *Staph aureus*, *Micrococcus* spp. and *Strept pyogenes*. It was observed that the most severe purulent endometritis was associated with the presence of *A. pyogenes*.

Domenech *et al.* (1977) described a condition in Ethiopian camels characterized by enlargement and caseation of internal and external lymph glands, locally called "mala". He examined 59 cases of the condition and recovered streptococci alone, or in association with other bacteria, from 13 cases (57%), *Corynebacterium pseudotuberculosis* was isolated from 37% of cases, *Staphylococcus* spp. from 10% and *A. pyogenes* from 6.7% of cases. The *Streptococcus* isolates belonged to lancefield's group B.

**1.2.6.3 Pyometra:**

Since progesterone domination of the genital system increases its susceptibility to infection, any condition which results in prolongation of the luteal phase in cattle can enable non-specific opportunistic organism to become pathogenic. A persistent corpus luteum either of dioestrus or a degenerate pregnancy, and luteal cysts, can sometimes result in pyometra
(Baker, 1987). Ball et al. (1983) observed that, in cattle, with persistent corpus luteum of dioestrous, the genital tract was under the continuous influence of progesterone and without the intervening oestrus, the infective process progressed. He added that in a small number of cases pyometra resulted from embryonic or fetal death in which the corpus luteum of pregnancy persisted, with subsequent invasion with *A. pyogenes* and the production of purulent exudates. In some of these instances it was possible to identify the remains of the embryo.

**1.2.6.4 Salpingitis, cervicitis and vaginitis:**

Chronic salpingitis, invariably resulting from ascending infection from the uterus, can result in infertility if unilateral and sterility if bilateral (Barlow et al., 1986). He isolated from the vagina and cervix of a cow following obstetric trauma incurred during the relief of difficult dystocia, *E. coli*, streptococci, staphylococci and *A. pyogenes*, the latter organism being most prominent in established infections. They also observed that, there was a change in the importance of different specific infectious agents in causing infertility and believed that adequate attention must be paid to all infectious agents. Those, which were incriminated, could cause catastrophic effects if they gained entry to a herd with a low immune status.
1.2.6.5 Genital vibriosis:

Infection due to *C. fetus* (formerly *Vibrio fetus*) has long been recognized as a cause of abortion in sheep and cattle (McFadyean and Stockman, 1913). Although its importance has declined with the use of A.I and because of bull screening at A.I studs and antibiotic addition in semen extenders. It must always be considered as a cause of infertility where natural service is used since it is most frequently transmitted venerally from carrier bulls to susceptible cows and heifers. In many countries *Compylobacter* is still a major cause of reproductive disease.

In a 15 year study in Argentine involving over 11300 bulls, 22% were found to be immuno-fluorescent -positive for *C. fetus* (Villar and Spina, 1982); whilst in 400 cows, in three dairy herds, in California 47% were seropositive (Ahktar *et al.*, 1993). About 90% of infertility in cattle was found to be due to *C. fetus* subspecies *C. veneralis* (Esslemont and Spincer, 1992).

Phillip *et al.* (1959) in a generalized survey in UK of repeat breeder cows showed that 75% had a low grade infection of the uterus and it had been estimated that 40% of infertility were due to vibriosis and advocated that uterine infection was an area problem, varied from place to place. They emphasized that infection usually took place at or soon after parturition.
1.2.6.6 Brucellosis (contagious abortion):

Brucellosis in Sudan was first reported as early as 1908 (Haseeb, 1950). *Br. abortus* was first isolated from a farm in Khartoum (Bennett, 1943), while *Br.meletensis* was isolated from British residents in Gezira area who drank goat milk (Dafalla and Khan, 1958). Brucellosis was reported from different parts of Sudan (Dafalla, 1962, Shigidi and Razig, 1971, and Ibrahim, 1975). Musa (1990) reported prevalence of the disease in man, cattle, camels, sheep, goats and equines and concluded that the highest prevalence rate was encountered in intensive farming systems and under nomadic conditions.

Brucellosis of camels is quite prevalent in Sudan and has been reported from many areas throughout the country. Abu Damir *et al.* (1943) tested sera from 740 camels of both sexes and found that 4.9% of the sera tested by the Rose Bengal Plate test, complement fixation and serum agglutination tests showed titers greater than 80 i.u. Both sexes were found to show high antibody levels. Tibin (1987) observed that brucellosis was also found widely spread in other species mixed with cattle like camels, horses and donkeys.
To date, only *Br. abortus* has been isolated from infected camels in the Sudan. Agab *et al.* (1995) isolated *Br. abortus* from camels serologically positive for brucellosis.

The incidence of brucellosis in camel populations seems to be related to breeding and husbandry practices (Richard, 1980). In the USSR where camels are kept in close proximity on large farms, there was serological evidence of a 15% infection rate (Palgov and Zhslobovski, 1964), where as in intensive systems, as in Chad, of 316 serum samples tested 13 (3.8%) showed positive agglutination reactions (Graber, 1986) and a reactor rate of 5.5% was reported in Ethiopia (Richard, 1980). Zaki (1948) showed the presence of *Br. abortus* agglutinins in 14% of male and 26% of female Egyptian camels. Hamada *et al.* (1963) examined 175 camels slaughtered in Cairo and Giza abattoirs for *Brucella* agglutinins and recorded an infection rate of 10.3%, most of these camels had originated from Sudan. In the Sudan itself, an infection rate of between 1.75 and 5.75% was reported by Mustafa and Awad Elkarim (1971) and 14% of camels in North Eastern Kenya were shown to be serologically positive by the Rose Bengal Plate test, serum agglutination test and complement fixation test (Waghela *et al.*, 1978). In Nigeria, Okoh (1979) found a reactor rate of approximately 1% of 31232 slaughtered animals reported to have originated from the Republic of Niger.
In India, Kulshrestha et al. (1975) using the tube agglutination test, reported a prevalence of 1.8% of 61315 apparently normal camels. Burgemeister (1975), using the milk ring test, examined 135 samples from Tunisian camels and found all were negative for Brucella antibodies, although 5.8% (3/52) of camels serum samples had complement fixing antibodies to Br. Abortus and 3.8% (2/52) to Br. melitensis.

Rutter and Mack (1963) stated that any of the three species of Brucella may infect camels, but that in Africa Br. melitensis was the causative species, whereas in the USSR infection was usually due to Br. abortus. Although camels are considered relatively highly susceptible to Brucella infection, attempts for isolation of the organism were met with variable success. In Egypt, Zaki (1948) failed to demonstrate Brucella spp. by direct culture or guinea-pig inoculation; in milk samples from 10 camels, one of which had a high agglutination titre. In an outbreak in the USSR in which 2% of animals aborted, Br. abortus was recovered from feutses and 15% were bacteriologically and serologically positive. In Kenya, Evans and Powys (1979) suspected brucellosis to be prevalent on a coastal camel ranch; however, laboratory tests were inconclusive and they failed to isolate the organism.
1.2.6.7 Tuberculosis of the genitalia:

Tuberculosis is not a common ailment of camels probably because most herds are husbanded in such a way as to avoid the closely-housed conditions which predispose to the disease. The exceptions are on camel farms in the USSR and in some countries of the Middle East where draught camels are often kept indoors in close association with cattle. In Egypt, tuberculosis has long been known as a disease of camels. As early as 1900, 3% of camels slaughtered at Cairo abattoirs were reported to have tuberculosis although imported Syrian and Arabian camels were free of the disease. Typical tubercle bacilli were demonstrated in lesions of Egyptian camels (Mason, 1917). He was of the opinion that the bacilli responsible for tuberculosis in camels were of the bovine type and that while camels were relatively resistant to the disease, their resistance broke down under stress and wasting was rapid.

Camel meat and milk constitute an important source of animal protein, especially for the majority of lower income groups of people. Camels living in open areas are seldom infected with tuberculosis, unlike those living indoor in close contact with other animal species (Rutter and Mack, 1963).
El. Mossalami, Siam and El-Sergani (1971) studied tuberculosis like lesions in slaughtered camels and found the percentage of tuberculosis to be 0.33% of the slaughtered camels. Old camels showed a higher frequency than young ones. They also found more lesions in males than in she-camels. Only the pulmonary form was observed.

In 1985, Chamoiseau, Bah, and Ahmed reported the first case of pulmonary tuberculosis was reported in camels in Mauritania. The disease was very rare on that country, but conditions for its extension were existing and infected milking females had appeared to transmit the infection via milk. At post-mortem examination, the most frequently affected organs in tuberculous camels were found to be the lungs, bronchial and mediastinal lymph nodes, pleura and livers. Lesions might also be found in the spleens, kidneys and trachea (Higgins, 1983).

1.2.6.8 Abortion and sporadic abortion:

Palgov and Zhslobovski (1964) reported seven cases of abortion in camels, from which Brucella spp. could not be isolated, but Strepto-coccus spp., pathogenic for mice and causing abortion in rabbits were recovered.

Sporadic abortion due to E.coli has been reported (Rowe and Smithies, 1978). It has been suggested that, following stress, the organism
reaches the fetus and placenta via haematogenous spread or ascending through the genital tract.

**1.2.6.9 Genital mycoplasmosis:**

There has been much controversy concerning the relationship between mycoplasmas and genital diseases in cattle ever since Mycoplasma bovigenitalium was demonstrated in the genital tract of infertile cows and semen of bulls (Edward et al., 1947; Blom and Erno, 1967). Evidence of pathogenicity has mainly been indirect, based frequently on the isolation of mycoplasmas from diseased rather than healthy tissue, and with limited experimental studies (Eaglesome and Garcia, 1992).

**1.2.7 Mycotic abortion:**

Mycotic agents also cause pre- or post calving problems and infertility. In this context Aspergillus spp. (Ainsworth and Austwick, 1973; Adam and Bada, 1975). Mucor spp., Rhizopus (Njoku, 1972) Candida spp. (Reyes et al., 1975, Cuci and Nika, 1983), Cladosporium spp., Pencillium spp. and other fungi were isolated from cows with reproductive problems.

In the Sudan the incidence and the role of fungi causing abortion and/or endometritis of repeat breeding condition have not been thoroughly investigated. Dafalla et al. (1984) found a correlation between mycotic infection in cows and infertility. They investigated 132 uterine washes from
fertile and repeat breeder cows and isolated *A. fumigatus, A. flavus, Penicillium* spp, *Rhodotorula* spp. and *C. albicans*.

Fungi were found to have a prolonged duration of infection and more adverse effects on fertility parameters than bacteria. It is most probable that this situation might have been due to inadequate attention paid to mycotic infection and in most cases to the indiscriminate use of antibiotics which suppress bacteria and thus indirectly favors the growth and propagation of fungi (Al Sayed *et al.*, 1994).

Fungal invasion of the placenta and fetus is a frequent and consistent cause of abortion. In cattle fungal abortion are normally sporadic; although in some herds the incidence may be as high as 5-10%. Elsewhere where the frequency of diagnosis is high; in the northeastern states of the USA mycotic abortion accounted for 22% of all infectious abortions and 5.1% of all abortions investigated (Hubert *et al.*, 1973). Kirkbride *et al.* (1973) stated that in South Dakota, USA in a survey over 5 years period, about 14.6% of all infectious abortions were due to fungi; and reached 4.8% of the total number of abortions.

Mycotic abortion in cattle is much more prevalent during the months of December, January, February and March in the U.K compared with the rest of the year. The fungi that are more frequently isolated following
abortion as *Absidia* spp, *Rhizopus* spp, *Mucor* spp and *Aspergillus* spp., particularly *A. fumigatus*. Other fungi such as *Mortiella wolfii* and *Petriellidium boydii* have also been implicated (Pepin, 1983). There are no reports about the isolation of fungi from cases of abortion in she-camels.
MATERIALS AND METHODS

This study was conducted between April 2000 and June 2001. Five camel herds of one-humped (*Camelus dromedarius*) in the vicinity of Schowak in the Botana area East of Sudan, consisting of 50 - 60 each were included in this study. The animals were 6 - 15 years of age and belonged to two breeds; Rashaidi and Anafi. Suspected she-camels were divided into three groups: Group A of repeat breeders’ heifer. Group B of first calvers with reported abortion and then became infertile. Group C of Adult multiparous which became infertile. Hundred and ten samples were collected from 55 she-camels suspected to be infertile. The number and types of samples collected are depicted in Table (1).

Table 1: A summary of the types and number of samples collected from each group of camels.

<table>
<thead>
<tr>
<th>Sample Type</th>
<th>Group A</th>
<th>Group B</th>
<th>Group C</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vaginal swabs</td>
<td>17</td>
<td>20</td>
<td>18</td>
<td>55</td>
</tr>
<tr>
<td>Blood</td>
<td>17</td>
<td>20</td>
<td>18</td>
<td>55</td>
</tr>
<tr>
<td>Total</td>
<td>34</td>
<td>40</td>
<td>36</td>
<td>110</td>
</tr>
</tbody>
</table>
2.1 Clinical examination:

1. History taking: The history was taken from the herd owners in special forms designed for this study (Appendix No.1).

2. General Examination.

The preliminary general inspection was carried out at some distance away from the camels. Since the study involved only reproductive disorders, attention was focused mainly on reproductive functions.

Other examinations, included temperature, pulse, palpation, percussion auscultation etc, to detect any abnormalities in the physical parameters of the suspected camels. Any animals showing discharges from the vulva-vagina received special attention.

2.2 Collection of samples:

2.2.1 Vaginal swabs:

Swabs were made of small pieces of cotton wool wrapped tightly around the ends of thin steel wires each was approximately 15 cm long. The swabs were inserted in test tubes which were then plugged with cotton wool and sterilized in a hot air oven at 180°C for 1 hr. The collection of vaginal swabs was made according to Ellaithy (1983).
Procedure:

She-camels were restrained in the sitting position and the tails were secured. The perineal area, skin, vulva were cleaned with soap and water and disinfected with 70% methyl alcohol. Sterile swabs were then removed from the tubes and gently inserted into the vagina. Care was taken not to touch any part of the surrounding while removing the swabs in order to avoid contamination. The swabs were then returned to the test tubes and sent to the laboratory in thermos flasks filled with ice cubes.

2.2.2 Blood:

For biochemical studies, 10 ml of blood was collected from the jugular vein using sterile syringes. Care was taken to avoid contamination either from the site of collection or from the system. Blood was left at room temperature for six hours and the sera were then separated and stored at -20°C till analyzed.

Preparation of plasma for biochemical studies, the blood was collected in a plastic heparinized container, shacked and rotated genitally then centrifuged in 3000 rpm for 5 minuets, the plasma will separated in the upper or clear layer then aspirated and preserved in refrigerator 4°C until used.
2.3 Sterilization of glassware:

New batches of glassware, Petri dishes, pipettes flasks etc were immersed in N HCl overnight to remove the surface alkali and were then rinsed with normal tap water 6 - 8 times, followed by four changes of distilled water, then dried in a hot air oven at 65°C before sterilization at 180 °C for 1 hr.

2.4 Media/s and reagent used for isolation and identification of bacteria:

(a) MacConkey agar (Oxoid CM7b):

<table>
<thead>
<tr>
<th>Contents</th>
<th>g/ Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peptone</td>
<td>20.0</td>
</tr>
<tr>
<td>Lactose</td>
<td>10.0</td>
</tr>
<tr>
<td>Bile salts</td>
<td>05.0</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>0.50</td>
</tr>
<tr>
<td>Neutral red</td>
<td>0.075</td>
</tr>
<tr>
<td>Agar No. 3</td>
<td>12.00</td>
</tr>
</tbody>
</table>

pH= 7-4 (approx.)

Procedures:

Fifty two grams were suspended in 1 litre of distilled water, boiled until dissolved completely and sterilized by autoclaving at 121°C for 15 min., then poured into sterile Petri dishes in 15 ml portions, allowed to dry and then stored at 4°C until used.

(b) Nutrient agar (Oxoid CM3):

<table>
<thead>
<tr>
<th>Contents</th>
<th>g/Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lab- lemco powder</td>
<td>1.0</td>
</tr>
<tr>
<td>Yeast extract</td>
<td>2.0</td>
</tr>
<tr>
<td>Peptone</td>
<td>5.0</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>0.5</td>
</tr>
<tr>
<td>Agar 3</td>
<td>15</td>
</tr>
</tbody>
</table>

pH= 7.4 (approx.)
Procedure:

Twenty eight grams were suspended in 1 litre of distilled water and brought to the boil to dissolve completely, then sterilized by autoclaving at 121°C for 15 minutes, cooled to 45 – 50°C and distributed into sterile Petri dishes in 15 ml portions for each plate. After drying, the medium was kept at 4°C until used.

(C) Blood agar (Oxoid CM55):

<table>
<thead>
<tr>
<th>Contents</th>
<th>g/Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lab-lemco powder</td>
<td>10</td>
</tr>
<tr>
<td>Peptone</td>
<td>10</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>05</td>
</tr>
<tr>
<td>Agar No. 3</td>
<td>15</td>
</tr>
</tbody>
</table>

pH 7.3 (approx.)

Procedure:

The contents were dissolved in 1 litre of distilled water. This was sterilized by autoclaving at 121°C for 15 minutes and cooled to 50°C. Defibrinated blood was allowed to warm at room temperature before being added to the molten agar, thoroughly mixed and the mixture was dispensed aseptically in 15 ml amounts in sterile Petri dishes. Depending upon the agar base used, the pH was within the range of 7.2 – 7.6 at room temperature. The prepared medium was kept at 4°C in sealed plastic bags until used.
(d) **Horse serum agar (Oxoid, Cruickshank et al., 1975):**

```
<table>
<thead>
<tr>
<th>Contents</th>
<th>g/Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peptone</td>
<td>10</td>
</tr>
<tr>
<td>Lab–Lemco powder</td>
<td>05</td>
</tr>
<tr>
<td>Dextrose</td>
<td>10</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>05</td>
</tr>
<tr>
<td>Agar</td>
<td>15</td>
</tr>
</tbody>
</table>
```

**Procedure:**

The ingredients were dissolved in distilled water and sterilized by autoclaving. 10 ml of sterile horse serum were added to 90 ml of the medium. Then mixed and distributed in Petri dishes.

(e) **Edward’s medium (Oxoid CM127):**

```
<table>
<thead>
<tr>
<th>Contents</th>
<th>g/Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Meat extract agar</td>
<td>01.12</td>
</tr>
<tr>
<td>Crystal violet sol</td>
<td>02.00</td>
</tr>
<tr>
<td>Aesculin</td>
<td>01.00</td>
</tr>
<tr>
<td>Sterile bovine blood</td>
<td>50.00</td>
</tr>
</tbody>
</table>
```

**Procedure:**

Aesculin was made into solution by boiling it in 5 ml of distilled water. It was added to the melted cooled agar at the same time the blood was added.

(f) **Motility medium (Barrow and Feltham, 1993):**

```
<table>
<thead>
<tr>
<th>Contents</th>
<th>g/Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peptone</td>
<td>10.0</td>
</tr>
<tr>
<td>Meat extract</td>
<td>3.0</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>5.0</td>
</tr>
<tr>
<td>Agar</td>
<td>4.0</td>
</tr>
<tr>
<td>Gelatin</td>
<td>80</td>
</tr>
</tbody>
</table>
```
Procedure:

The gelatin was soaked in 1 litre of distilled water for 30 minutes then the other ingredients were added and heated to dissolved and then sterilized at 115°C for 20 minutes and distributed in sterile test tubes 5 ml amounts. The prepared medium was kept at 4°C until used.

(g) MR–VP medium (Oxoid CM 43):

<table>
<thead>
<tr>
<th>Contents</th>
<th>g/Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peptone</td>
<td>5.0</td>
</tr>
<tr>
<td>Dextrose</td>
<td>5.0</td>
</tr>
<tr>
<td>Phosphate buffer</td>
<td>5.0</td>
</tr>
</tbody>
</table>

pH 7-5 (approx.)

Procedure:

Fifteen grams were added to 1 litre of distilled water mixed well then distributed into test tubes 5 ml amounts and sterilized by autoclaving at 121°C for 15 minutes. The prepared medium was kept at 4°C until used.

(h) Meat infusion broth:

<table>
<thead>
<tr>
<th>Contents</th>
<th>g/Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lean meat</td>
<td>500</td>
</tr>
<tr>
<td>Peptone</td>
<td>20</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>5</td>
</tr>
</tbody>
</table>

PH = 7-4 (approx.)

Procedure:

Watery extract of meat was made by extracting lean meat for 24 hours at 2°C. The remaining juice was squeezed from the meat. The extract was simmered for 15 minutes and filtered then 1% peptone and 0.5% sodium
chloride were added, before autoclaving at 121°C for 15min. The content were dissolved in 1 litre of distilled water and distributed in sterile test tubes 5 ml amounts.

2.5. Media used for isolation of fungi:

(a) Sabourauds dextrose agar (SDA) with chloramphenicol:

<table>
<thead>
<tr>
<th>Contents</th>
<th>g/Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mycological peptone</td>
<td>10</td>
</tr>
<tr>
<td>Dextrose</td>
<td>40</td>
</tr>
<tr>
<td>Agar No.1</td>
<td>15</td>
</tr>
<tr>
<td>Chloramphenicol</td>
<td>0.05</td>
</tr>
</tbody>
</table>

pH 5.6 (approx.)

Procedure:

An amount of 65 grams of the dehydrated medium were suspended in 1 litre of distilled water boiled and autoclaved at 121°C for 15 minutes then aseptically chloramphenicol (0.05g/litre) was added. The medium was distributed into sterile Petri dishes or sterile bottles to make slants. The prepared medium was kept at 4°C until used.

(b) Corn meal agar (Oxoid):

<table>
<thead>
<tr>
<th>Contents</th>
<th>g/Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Corn meal extract</td>
<td>2.0</td>
</tr>
<tr>
<td>Agar No. 3</td>
<td>15</td>
</tr>
</tbody>
</table>

pH. 6.0 (approx.)
Procedure:

An amount of 17 grams was suspended in 1 litre of distilled water, boiled to dissolve and sterilized by autoclaving at 121°C for 15 minutes. Then the media was poured aseptically into sterile Petri dishes or bottles and allowed to solidify, then stored at 4°C until used.

(c) Malt extract agar (Oxoid):

<table>
<thead>
<tr>
<th>Contents</th>
<th>g/Litre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Malt extract</td>
<td>30</td>
</tr>
<tr>
<td>Mycological peptone</td>
<td>5.0</td>
</tr>
<tr>
<td>Agar</td>
<td>15</td>
</tr>
</tbody>
</table>

pH = 7-4 (approx.)

Procedure:

The contents were dissolved in 1 litre of distilled water and sterilized by autoclaving at 121°C for 15 minutes. The melted media was dispensed in 15 ml amounts in universal bottles and solidified in slopes.

2.6. Methods of isolation and identification of microorganisms:

All vaginal swabs from the three groups of she-camels were cultured on bacteriological media: MacConkey agar, Blood agar, nutrient agar, horse serum agar and Edward's medium, and mycological media, Sabouraud's dextrose agar, Corn meal agar and malt extract agar. Bacteriological media were incubated at 37°C for 24-48 hrs aerobically, under 10% CO₂ tension and anaerobically. Microaerophilic and anaerobic condition were provided
using commercial gas packs. Plates with negative growth were further incubated for 7 days before being discarded as negative.

Samples cultured for fungi were incubated on to slopes of different mycological medias and incubated at 27ºC. The plates were incubated at room temperature and examined daily for 4 weeks before they were discarded as negative. Stained by gram methods and examined under 100X lens.

2.6.1 Gram's stain

Required solutions

- Two grams of crystal violet or methyl violet were dissolved in 20 ml methyl alcohol.
- 0.8 gram of ammonium oxalate was dissolve in 80 ml distilled water.
- The two solutions were mixed and filtered through two layers of filter paper and stored in tightly capped bottle at room temperature.
Procedure:

- A thin smear was prepared, air dried and fixed by passing through Bunsen burner's flame three times, or fixed with methanol or ethanol for 5 minutes.
- Crystal violet stain was applied for 0.5 minutes and washed with water.
- Lugol’s iodine solution was applied and drained for 0.5 minutes.
- Decolourization was done with few drops of acetone which was washed thoroughly with water.
- Counter staining was done with weak carbol fuchsin for 0.5 min; the smear was thoroughly washed, drained, blotted and dried.

2.7 Biochemical tests used for identification of bacteria:

The following tests were performed to identify and differentiate the various organisms.

(a) Aesculin hydrolysis:

Aesculin agar was used to differentiate sterptococci. A pure culture agar plate was prepared and inoculated with the bacterium or the Aesculin agar slopes were inoculated with a pure culture of the media then incubated at 37°C for up to 7 days and examined daily for dark brown or black
precipitation. Black precipitates indicated hydrolysis of aesculin to 6-7 dihydroxycoumarin.

(b) Catalase test:

This test was used to detect the ability of the organism to produce the enzyme catalase which catalyzes the release of oxygen from hydrogen peroxide giving rise to small bubbles from the bacterial suspension. A bulk of bacterial colonies was placed on a clean slide using a wire loop and a drop of 3% H₂O₂ was poured over the colonies, the appearance of immediate bubbling indicated a positive result.

(c) Oxidase test:

This test was conducted to examine the ability of the bacterium to produce the enzyme oxidase. A filter paper soaked with oxidase reagent strip was placed on a sterile Petri dish or an a clean microscope slide. Using a bent glass rod, a generous amount of bacterial culture was streaked on the moist part of the filter paper. Appearance of blue coloration within 10 seconds indicated a positive reaction.

(d) Indole test:

Peptone water was inoculated with the bacterium using a wire loop and was incubated for 24 hours. Approximately 0.5 ml of culture was removed in a narrow test tube and 0.2 ml of Kovac’s reagent (appendix2) was added
and gently shacked, and observed for the appearance of a red colour which indicates the break down of tryptophane to indole.

(e) Urease production test:

This test was used for the detection of the ability of the organism to produce urease which breaks down the urea incorporated in the medium thus liberating ammonia which in turn increases the pH of the medium to alkalinity. A loop full of bacterium under test was inoculated on the urea slope and incubated at 37°C over night. The positive change showed change of colour from light yellow to pink.

(f) Nitrate reduction test: (Cook, 1950)

A blotting paper strip (about 16 ×10 mm) containing potassium nitrate was placed in the centre of blood agar plate at the time of inoculation. The strips were papered by soaking bloting paper in a warm solution of 40% potassium nitrate and then dried. Stab-inoculants test and control organisms were placed near the periphery of the plate and incubated for 24-48 hrs. A positive reaction was indicated by a dark green-brown zone of methaemoglobin formed by the reduction of hemoglobin by nitrite.
(g) **Gelatin liquefaction:**

Charcoal gelatin discs were used for gelatin liquefactions reaction, which is shown by the release of carbon particles in the medium, with eventual complete disintegration of the disc. Preserved gelatin discs were washed in sterile peptone water and then placed in universal bottles containing bacterial suspension in peptone water and incubated for 24 hours at 37°C. Intact discs indicated negative results.

(h) **Motility test:**

A direct examination method was used to determine motility of the organisms. The organism was grown in peptone water and incubated for 4-6 hours. A small amount of white petroleum jelly was applied at the corners of a clean cover slip. A drop of bacterial culture was placed on the middle of the cover slip the slide was then examined under x<sup>10</sup> objective to focus the suspension and x<sup>40</sup> to a certain the motility.

(i) **Sugar fermentation test:**

The test organism was inoculated into peptone water containing different sugars. The media was then incubated at 37°C and examined daily for up to 7 days. Acid production was indicated by the development of a red colour in the media, whereas gas production was shown by development of an empty space in the Durham tubes.
(j) Oxidation fermentation (O/F) test:

Two tubes of Hugh and Leifson's (Barrow and Feltham, 1993) medium were inoculated with the test culture. One of the tubes was covered with a layer of sterile paraffin oil to about 3cm above the surface of the medium, the other was left unsealed. Both were then incubated at 37°C and examined daily up to two weeks. Fermentative organisms were indicated by change in colour to yellow in both tubes, while oxidative organisms were indicated by change in the unsealed tube only. A blue colour in both tubes was considered negative.

(k) Novobiocin sensitivity test:

A volume of two ml of diluted test culture was spread on the surface of a nutrient agar plate. The excess fluid was discarded and the plate was allowed to dry, then Oxoid disc of novobiocin (5 mg) was applied to the surface of the medium with a sterile forceps and the media was incubated at 37°C for 24 hours. A zone of inhibition around the disc indicated that the organism was sensitive to novobiocin.

(l) Coagulase test:

This test was used to detect the ability of the organism to produce the enzyme coagulase, which is capable of clotting plasma. Positive reaction was indicated by formation of clumps of the plasma.
Two separate drops of sterile distilled water were placed on a clean slide. A portion of culture is added to each drop and mixed to form even suspensions, and then 0.25 ml of rabbit plasma was added to one drop which was mixed and rocked gently for 20 seconds. The formation of clumps in the plasma-containing drop and not in the other drop meant a positive reaction.

2.8 Blood biochemical parameters analysis:

Blood metabolites were analyzed at the Department of Biochemistry, Faculty of Veterinary Medicine, University of Khartoum.

2.8.1 Determination of total blood protein:

Biuret reagent:

This reagent was prepared by dissolving 1.5 g copper sulphate (CuSO₄·5H₂O₂,) and 6 g sodium potassium tartarate (NaKC₄H₄O₆·4H₂O) in 500 ml of water, to which 300 ml carbonate and 10% sodium hydroxide were added with agitation. Then 2 g of potassium iodide was added and the final volume was made up to 2 liters and stored in a polythene bottle.

Peptide structures containing at least two peptide linkages gave positive violet colour reaction, but ammonia salts could interfere with the reaction.
Procedure:

Standard protein solution was prepared by weighing accurately 10 g of bovine albumin and dissolving it in 100 ml distilled water. To three test tubes labeled as, blank, standard and test the following contents were added as illustrated in the table below:

<table>
<thead>
<tr>
<th>Contents</th>
<th>Blank</th>
<th>Standard</th>
<th>Test</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum (ml)</td>
<td>--</td>
<td>--</td>
<td>0.2</td>
</tr>
<tr>
<td>Distilled Water (ml)</td>
<td>0.2</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Standard (ml)</td>
<td>-</td>
<td>0.2</td>
<td>-</td>
</tr>
<tr>
<td>Biuret reagent (ml)</td>
<td>4.8</td>
<td>4.8</td>
<td>4.8</td>
</tr>
</tbody>
</table>

The contents in the three tubes were mixed and then incubated at 37°C for 10 minutes, or left at room temperature for 30 minutes. The readings were made at 540 µm in a spectrophotometer or by using a colorimeter with a green filter (625 µm).

Calculations:

Protein concentration = \( \frac{\text{Reading of test}}{\text{Reading of std}} \times \text{std conc.} = \text{g/100 ml} \)

The concentration of protein in solution was expressed as weight per volume (g/100ml).

2.8.2 Determination of blood inorganic phosphorus:

Test:

One ml of plasma was added to 9 ml of 10% Trichloroacetic Acid (TCA) then filtered. 5 ml were then taken from the supernatant.

Standard:

The standard contained 0.5ml of working standard plus 4.5ml of 10% TCA.
Blank:

The blank was made up of 5 ml of 10% TCA. One ml of ammonium molybdate solution was added to all tubes then mixed and 1 ml of Metol solution was added, mixed and allowed to stand for 30 minutes at room temperature. Reading was taken at 680mµ.

Calculation:

\[
\frac{T}{S} \times 5 = \text{mg/100 ml}
\]

Where

- \(T\) = Reading of test.
- \(S\) = Standard concentration.
- \(S\) = Reading of standard.

Preparation of reagents:

- 10% TCA: 10g trichloroacetic acid in 100 ml distilled water.

- Ammonium molybdate: 15 g of ammonium molybdate in about 400 ml distilled water then 100 ml of N sulphuric were added, then made up to 800 ml with distilled water.

- Metol: One g of p- methyl amino – phenol in 100 ml of 3% solution of sodium bisulphate
**Standard:**

Dissolve 0.2197 g of potassium dihydrogen phosphate in distilled water, made up to one liter with water, then few drops of chloroform were added.

**2.8.3 Determination of blood calcium:**

**Test:**

0.5 ml of serum was mixed with 1.0 ml of 0.5% chloranillic acid, in a centrifuge tube.

**Standard:**

0.5 ml working standard +1.0 ml of chloranillic acid were let to stand for 5 minutes, then centrifuged at 503.1 relative centrifugal forces (RCF) for 5 minutes.

The supernatant was decanted and the tubes were drained on clean filter papers.

The precipitate was washed with 0.5 ml of distilled water then centrifuged, decanted and drained.

The precipitate was dissolved in 4 ml of 4% ferric nitrate and left to stand for 5 minutes.

The test was read at 500 nm using ferric nitrate as a blank.
Calculation:

\[
\frac{T \times B}{S - B} = \text{mg}/100\ \text{ml}
\]

Where
- \( T \) = Reading of test.
- \( B \) = Reading of the Blank.
- \( 10 \) = Concentration 10 mg/100 ml

Reagents:

Chloranillic acid: 0.5%: Without using heat 0.5 g-chloranillic acid was dissolved in 100ml distilled water containing 0.4 ml. Ethanolamine. Then 50/mg sodium cyanide was added, and allowed to stand overnight. The clear upper layer was then removed and stored in the refrigerator.

Feric nitrate: 10g Fe (No₃)₃ was dissolved in 200ml distilled water. Add 30 ml. Of NH₄SO₄ and make up to 250ml with water.

Standard stock solution:

0.25 g of calcium carbonate was dissolved in 0.1 /N HCl and made up to 100 ml. with acid.

Working standard solution:

4 ml of stock in 100ml. distilled water.

Calcium chloride stock solution: 1mg /1ml.

One ml. of stock solution in 10 ml. of distilled water.
2.8.4 Determination of blood iron (Iron Binding Capacity)

**Principle of the method:**

An excess Fe\(^{3+}\) was added to the sample to saturate serum transferring. Uncomplexed Fe\(^{3+}\) was precipitated with magnesium hydroxide carbonate and iron bound to protein in the supernatant was then spectrophotometrically measured.

**Content and composition:**

1. Reagent A. 50 ml iron chloride 0.2 mmoL/L.

2. Reagent B. Magnesium hydroxide carbonates (powder).

To be dispensed in the enclosed plastic spoon.

Additional Reagent -Iron kit.cod.11509.

**Additional Equipment:**

- Desktop centrifuge.

- Spectrophotometers with a 560nm filter (550-570 µm).

**Procedure:**

**Precipitation**

1. Pipetted into labeled tubes:

<p>| | |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample</td>
<td>0.5 ml</td>
</tr>
<tr>
<td>Reagent</td>
<td>1.5 ml</td>
</tr>
</tbody>
</table>

2. Mixed thoroughly and let to stand for 5-30 minutes at room temperature.
3. To each tube one spoonful of Reagent B was added.

4. Mixed thoroughly and then left for 30-60 minutes at room temperature, and during this time thoroughly mixed several times.

5. Then Centrifuged at a minimum of 503.1 RCF. For 10 minutes.

6. Carefully the supernatant was collected.

7. The iron concentration in the supernatant was measured using the kit cod.

**Calculations:**

\[
\text{Total Iron Binding Capacity (TIBC)} = \text{Iron (supernatant)} \times 3
\]

(dilution).

Latent Iron Binding Capacity (LIBC).

\[
\text{LIBC} = \text{TIBC} - \text{serum iron}
\]

**Notes:**

1. Haemolized samples were discarded.

2. The supernatant was stored for up to 1 hour at room temperature. If the supernatant appeared turbid, then it was removed to be centrifuged again.

3. Contamination of glassware with iron will affect the test. Acid washed glassware or plastic tubes were used.
2.9 Rose Bengal Plate test (RBPT):

Collected blood samples obtained from 55 she-camels suspected to be infertile, were centrifuged at 55.9 RCF decanted and stored at -20°C. The serum samples were examined for brucella agglutinins by the Rose Bengal Plate test (RBPT) using the method of Morgan et al. (1978).

Procedure:

Equal volumes of undiluted serum, and stained antigen were placed on a card or glass plate, mixed well with a glass rod, rocked gently back and forth about 5 minutes and then the test was read, in a good light, known positive and negative sera were included in each as test controls.

No agglutination was regarded as negative, while any degree of agglutination (1+, 2+, 3+) was considered a positive reaction.

2.10 Progesterone radioimmunoassay kit (1MK – 458):

The assay was performed in the laboratory of the Sudan Atomic Energy Commission, Khartoum.

Serum from 55 she-camels were separated and stored at – 20°C. The specific radioimmunoassay kits were imported from the Department of Isotopes, Institute of Atomic Energy, in Beijing, China. Serum progesterone concentration was measured according to the method of Homeida (1986).
**Principle of the procedure:**

The radioimmunoassay (RIA) method depends on the competition between progesterone in the specimen and $^{125}\text{I}$– labeled progesterone for a limited number of binding sites on a progesterone specific antibody.

The proportion of the $^{125}\text{I}$– labeled progesterone bound to the antibody is inversely related to the concentration of the progesterone present in the serum. The separating agent is micro particles coupling with a second antibody. Separation of the antibody bound fraction is effected by centrifugation and decantation of the supernatant. By measuring the proportion of $^{125}\text{I}$– labeled progesterone bound in presence of reference standard sera containing known amounts of progesterone; the concentration of progesterone present in the specimens can be determined.

**Materials provided (RIA kits):**

1. $^{125}\text{I}$ – Labeled progesterone solution was provided in a bottle of 22 ml (Red colour).

2. Progesterone antibody solution (raised in rabbits) was provided in a blue bottle of 11 ml.

3. Reference standards (7) of progesterone in lyophilized forms when dissolved they provide the following concentration:

   A: 0 ng/ml   B: 0.2 ng/ml   C: 0.5   D: 2.2 ng/ml
4. The separating agent containing solid phase second antibody micro particles suspension in a bottle of 22 ml.

**Storage instructions:**

1. All kit contents were stored at 2-8°C (not freezeed).
2. All reagents were allowed to come to room temperature prior to use in the assay.

**Assay procedure:**

1. Labeled duplicated tubes were arranged in the assay rack.
2. Each standard was dissolved with 0.5 ml saline solution thoroughly (Except for standard (A) which already contained 1 ml of saline solution).
3. 100µl of each standard, control and she-camel serum was pipetted to the appropriate labeled tubes (see Table 1). Zero standard A was used for non specific binding (NSB) reagent tubes.
4. 200 µl of $^{125}$I- progesterone tracer solution was added to each tube and was mixed briefly using a vortex shaker.
5. 100µl of progesterone antiserum (NSB tubes) was added to the appropriate tubes and was mixed briefly using a vortex shaker.
6. 200µl suspension of separating agent was added to each tube and was mixed thoroughly.

7. All tubes were incubated at 37°C for 3 hours.

8. The tubes were centrifuged for 20 minutes at 1500xg.

9. The supernatant was discarded by decantation.

10. The radioactivity of the precipitate remaining in the tubes was read in an Oak field multi well Gamma counter (PC – RIA. MAS, STRATEC, USA).

### Progesterone RIA kit assay protocol.

<table>
<thead>
<tr>
<th>Total counts</th>
<th>N S B</th>
<th>Standard</th>
<th>Sample</th>
</tr>
</thead>
<tbody>
<tr>
<td>Zero standard</td>
<td>100 µl</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Standards</td>
<td>100 µl</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Samples</td>
<td></td>
<td>100µl</td>
<td></td>
</tr>
<tr>
<td>²⁵³ T-P₄</td>
<td>200 µl</td>
<td>200 µl</td>
<td>200 µl</td>
</tr>
<tr>
<td>Antibody</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
</tr>
<tr>
<td>Saline</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
</tr>
<tr>
<td>Separating agent</td>
<td>200 µl</td>
<td>200 µl</td>
<td>200 µl</td>
</tr>
</tbody>
</table>

**Calculation of results:-**

1. The average cpm for each pair of duplicates tubes was determined.

2. Derive B/B₀ for each standard, control and sample as follow:

   \[
   \frac{B}{B₀} \times \frac{B - NSB}{B₀ - NSB} \times 100
   \]

   Where:
   
   - \( B \) = mean count rate for each pair of tubes.
   - \( B₀ \) = mean count rate for the zero standard or (total binding).
   - NSB = mean count rate for the non – specific binding tubes.
3. A standard curve was constructed by plotting the \( \frac{B}{B_0} \%) \) of each standard against its concentration in \( \eta g/ ml \) (or \( \eta mol/L \)) on logit – 10 g or semi – log graph paper.

4. The value of each serum sample was obtained and controlled by reference to the standard curve.

2.11 Statistical analysis:

Data analysis were performed using statistical package for social science (SPSS)
RESULTS

A total of 110 samples was collected (Table 1). These samples were tested to determine the factors responsible for the infertility.

3.1 Bacteriological results (vaginal swabs):

As depicted in Tables (2, 3, and 4), 49 (89.1%) out of 55 vaginal swabs were positive for bacteriological growth while six (10.9%) were negative. Staphylococci appeared to be the predominant bacteria; 29 isolate (34.5%). *Staph. aureus* was isolated five times (5.1%), whereas of the coagulase-negative Staphylococci, *Staph. schleriferi* was isolated six times (7.1%), *Staph. cohnii* twice (2.4%), and *Staph. warneri, Staph. caseolyticus, Staph. xylosus, Staph. arietae, Staph. chromogenes, Staph. gallinarum, Staph. lentus, Staph. carnosus* and *Staph. caprae* where isolated once (1.2%) each. Staphylococci were followed by Streptococci 21 isolates (25%). *Strept. pygogenes* was isolated five times (5.1%), *Strept. dysgalactiae* four times (4.8%) and *Strept. equimisilis* and *Strept. zooepidemics* were isolated twice each (2.4%). *Strept. porcinus, Strept. suis.* and *Strept. mutans* were isolated once each (1.2%). These were followed by *corynebacteria* which was isolated 13 times (15.5%), *C. pseudodiphtheriticum* four times (4.8%), *C. diphtheriae* three times (3.6%), *C. pseudotuberculosi*s and *C. cystitidis* two
isolates each (2.4%). An unidentified *Corynebacterium spp* and *C. munutissimum* were isolated once each (1.2%).

There were seven isolates (8.3%) of *Bacillus* spp: *Bacillus mycoides* was isolated four times (4.8%), *Bacillus badius* twice (2.4%) and *Bacillus cereus* once (1.2%). *Micrococcus kristinae* and *Micro. sedentarius* were isolated twice (2.4%) each. *Vibrio fluvialis* was isolated three times (3.6%). *Stomatococcus mucilaginosus* was isolated twice (2.4%) and *Aerococcus* spp, *E.coli, Morganella morganii, Enterococcus faecalis* and *Clostridium chauvoei* were isolated once each (1.2%).

Animals in group A- repeat breeder- heifers appeared to be the most infected as bacteria were isolated from all animals (100%), while only 90% of group B animals and 77% of group C animals were infected (Tables, 2, 3 and 4).

Mixed infections were observed in all three groups; 12 samples from group A had more than 1 isolate (70.6%), 11 samples (61.1%) from group B gave more than one isolate and 3 samples (21.4%) from group C had mixed infections (Tables, 2, 3 and 4).

**3.2. Mycological results (vaginal swabs):**

Fifty five vaginal swabs were cultured in different mycological culture media and three fungi were isolated (5.5%); two were *Aspergillus*
spp (66.7%) and one was a *Mucor* spp (33.3%). The remaining 52 swabs (94.6%) were negative (Table 6).

### 3.3. Detection of brucella antibodies:

Serological studies for detection of *Brucella* antibodies were carried out on 55 serum sample from different animals. Thirty two samples (58.2%) were positive, 11 with 1+ (34.4%), 14 with 2+ (43.8%) and seven samples with 3+ (21.9%), while 23 samples (41.8%) were negative (Table 7).

Animals in group C appeared to be the most infected with *Brucella* as 14 animals were positive (43.8%), followed by group B where 10 animals were positive (31.3%) whereas only eight animals in group A (25%) were positive serologically.

### 3.4. Hormone (progesterone) assay:

Blood samples collected from the three groups for progesterone detection showed that some of the animals within each group were pregnant (Table 8) according to high level of progesterone the rest non-pregnant were considered the infertile and were then compared with the pregnant animals for blood biochemical parameters deviation. There was a significant difference (P > 0.05) in the concentration of progesterone between pregnant and non-pregnant (infertile) animals (table 8).
3.5. **Determination of blood biochemical parameters of the plasma of infertile repeat breeders and pregnant heifers (group A):**

Blood biochemical parameters conducted in group A revealed that there was no significant difference in the proteins, calcium, inorganic phosphorus and iron concentration in the infertile heifers as compared to those which became pregnant in the same group (Table 9, Appendix V).

3.6. **Determination of blood biochemical parameters of infertile first calvers with reported abortion then became infertile (group B):**

Blood biochemical parameters conducted in group B revealed that there was no significant difference in the protein, calcium and inorganic phosphorus concentration in the infertile heifers as compared to those which became pregnant in the same group while iron concentration was found to be significantly higher ($P > 0.01$) in infertile animals (Table 10, Appendix VI).

3.7. **Determination of blood biochemical parameters of adult multiparities then became infertile (group C):**

In this group plasma protein concentration was significantly lower in infertile than in pregnant heifers ($P < 0.05$), calcium was significantly higher in the infertile heifers ($P > 0.05$), inorganic phosphorus was significantly lower ($P > 0.01$) and the iron was significantly higher in infertile. (Table 11, Appendix VII)
Table 2: Bacteria Isolated from Vaginal Swabs of Group A She-camels.

<table>
<thead>
<tr>
<th>Vaginal swab No.</th>
<th>Number of isolates</th>
<th>Types of isolates</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1i</td>
<td>2</td>
<td>Aerococcus spp; Streptococcus*</td>
</tr>
<tr>
<td>A2i</td>
<td>2</td>
<td>Micrococcus kristinae Staph. caseolytics</td>
</tr>
<tr>
<td>A3</td>
<td>1</td>
<td>Staph. aureus **</td>
</tr>
<tr>
<td>A4i</td>
<td>2</td>
<td>Staph. schleriferi *** Stomatococcus mucilaginosus.</td>
</tr>
<tr>
<td>A5</td>
<td>1</td>
<td>C. pseudodiphtheriticum</td>
</tr>
<tr>
<td>A6</td>
<td>1</td>
<td>Strept. pyogenes</td>
</tr>
<tr>
<td>A7i</td>
<td>2</td>
<td>Strept. pyogenes Strept. zooepidemics</td>
</tr>
<tr>
<td>A8</td>
<td>1</td>
<td>Staph. xylosus</td>
</tr>
<tr>
<td>A9i</td>
<td>2</td>
<td>Strept. pyogenes Clost. chauvoei</td>
</tr>
<tr>
<td>A10</td>
<td>1</td>
<td>Strept. pyogenes</td>
</tr>
<tr>
<td>A11i</td>
<td>2</td>
<td>Enterococcus. faecalis. C. munutissimum.</td>
</tr>
<tr>
<td>A12i</td>
<td>2</td>
<td>Staph.arlettae *** Staph. schleriferi***</td>
</tr>
<tr>
<td>A13i</td>
<td>2</td>
<td>Staphylococcus* Bacillus badius.</td>
</tr>
<tr>
<td>A14i</td>
<td>2</td>
<td>Staphylococcus* C.pseudotuberculosis</td>
</tr>
<tr>
<td>A16i</td>
<td>2</td>
<td>Bacillus badius. Staph.chromogens ***</td>
</tr>
<tr>
<td>A17i</td>
<td>4</td>
<td>Staphylococcus* C.diphtheriae. Bacillus cereus. C. cystitidis.</td>
</tr>
</tbody>
</table>

* could not be identified to the species level
** Coagulase positive
*** Coagulase negative

Table 3: Bacteria Isolated from Vaginal Swabs of Group B She-camels.

<table>
<thead>
<tr>
<th>Vaginal swab No.</th>
<th>Number of Isolates</th>
<th>Types of Isolates</th>
</tr>
</thead>
<tbody>
<tr>
<td>B1</td>
<td>1</td>
<td><em>Strep. equisimilis.</em></td>
</tr>
<tr>
<td>B2</td>
<td>0</td>
<td>-</td>
</tr>
<tr>
<td>B3i</td>
<td>2</td>
<td><em>Bacillus mycoides.</em></td>
</tr>
<tr>
<td>3ii</td>
<td></td>
<td><em>Staph. aureus.</em> **</td>
</tr>
<tr>
<td>B4</td>
<td>1</td>
<td><em>Strep equisimilis.</em></td>
</tr>
<tr>
<td>B5i</td>
<td></td>
<td><em>Strep. suis.</em></td>
</tr>
<tr>
<td>5ii</td>
<td>2</td>
<td><em>Staph. aureus.</em> **</td>
</tr>
<tr>
<td>B6i</td>
<td>2</td>
<td><em>C. cystitidis.</em></td>
</tr>
<tr>
<td>6ii</td>
<td></td>
<td><em>Strep dysgalactiae.</em></td>
</tr>
<tr>
<td>B7</td>
<td>0</td>
<td>-</td>
</tr>
<tr>
<td>B8</td>
<td>1</td>
<td><em>Strep. dysgalactiae.</em></td>
</tr>
<tr>
<td>B9</td>
<td>1</td>
<td>*** <em>Staph. schleriferi</em></td>
</tr>
<tr>
<td>B10</td>
<td>1</td>
<td><em>Bacillus mycoides</em></td>
</tr>
<tr>
<td>B11i</td>
<td>2</td>
<td><em>Strep. dysgalactiae</em></td>
</tr>
<tr>
<td>B12i</td>
<td></td>
<td><em>Streptococcus</em> *</td>
</tr>
<tr>
<td>12ii</td>
<td>3</td>
<td><em>Staph. gallinarum.</em> ***</td>
</tr>
<tr>
<td>12iii</td>
<td></td>
<td><em>Corynebacterium.</em> *</td>
</tr>
<tr>
<td>B13i</td>
<td>2</td>
<td><em>C. pseudodiphtheriticum</em></td>
</tr>
<tr>
<td>13ii</td>
<td></td>
<td><em>Staphylococcus</em> *</td>
</tr>
<tr>
<td>B14i</td>
<td>4</td>
<td><em>C. diphtheriae</em></td>
</tr>
<tr>
<td>14ii</td>
<td></td>
<td><em>Bacillus mycoides.</em></td>
</tr>
<tr>
<td>14iii</td>
<td></td>
<td><em>Staph. cohnii.</em> ***</td>
</tr>
<tr>
<td>B15i</td>
<td>2</td>
<td><em>Escherichia coli</em></td>
</tr>
<tr>
<td>15ii</td>
<td></td>
<td><em>Morganella morganii</em></td>
</tr>
<tr>
<td>B16i</td>
<td>2</td>
<td>*** <em>Staph. schleriferi</em></td>
</tr>
<tr>
<td>16ii</td>
<td></td>
<td><em>C. pseudodiphtheriticum</em></td>
</tr>
<tr>
<td>B17i</td>
<td>2</td>
<td><em>Staph. schleriferi.</em> ***</td>
</tr>
<tr>
<td>17ii</td>
<td></td>
<td><em>C. pseudotuberculosis</em></td>
</tr>
<tr>
<td>B18</td>
<td>1</td>
<td>*** <em>Staph. lentus</em></td>
</tr>
<tr>
<td>B19</td>
<td>1</td>
<td><em>Micrococcus sedentarius</em></td>
</tr>
<tr>
<td>B20i</td>
<td>4</td>
<td><em>C. pseudodiphtheriticum.</em> **</td>
</tr>
<tr>
<td>20ii</td>
<td></td>
<td><em>Staph. aureus</em></td>
</tr>
<tr>
<td>20iii</td>
<td></td>
<td><em>Bacillus mycoides.</em></td>
</tr>
<tr>
<td>20iv</td>
<td></td>
<td><em>Staph. carnosus</em>**</td>
</tr>
</tbody>
</table>

* could not be identified to the species level.
** Coagulase positive
Table 4: Bacteria Isolated from Vaginal Swabs of Group C She-camels.

<table>
<thead>
<tr>
<th>Vaginal swab No.</th>
<th>Number of isolates</th>
<th>Types of isolates</th>
</tr>
</thead>
<tbody>
<tr>
<td>C1</td>
<td>0</td>
<td>-</td>
</tr>
<tr>
<td>C 2</td>
<td>0</td>
<td>-</td>
</tr>
<tr>
<td>C 3</td>
<td>1</td>
<td>Staphylococcus *</td>
</tr>
<tr>
<td>C 4</td>
<td>1</td>
<td>Vibrio. fluvialis</td>
</tr>
<tr>
<td>C 5</td>
<td>1</td>
<td>Strept. zooepidemics.</td>
</tr>
<tr>
<td>C 6</td>
<td>1</td>
<td>Strept. pyogenes.</td>
</tr>
<tr>
<td>C 7</td>
<td>1</td>
<td>Streptococcus. *</td>
</tr>
<tr>
<td>C 8</td>
<td>0</td>
<td>-</td>
</tr>
<tr>
<td>C 9</td>
<td>0</td>
<td>-</td>
</tr>
<tr>
<td>C 10</td>
<td>1</td>
<td>Staphylococcus. *</td>
</tr>
<tr>
<td>C 11</td>
<td>1</td>
<td>Micrococcus sedentarius.</td>
</tr>
<tr>
<td>C 12</td>
<td>1</td>
<td>Staph. aureus **</td>
</tr>
<tr>
<td>C 13</td>
<td>1</td>
<td>Staph. warneri. ***</td>
</tr>
<tr>
<td>C 14</td>
<td>1</td>
<td>Staph. cohnii ***</td>
</tr>
<tr>
<td>C 15.1</td>
<td>2</td>
<td>Staphylococcus.* Strept.dysgalactiae.</td>
</tr>
<tr>
<td>15.2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>C 16.1</td>
<td></td>
<td>Staph. caprae. *** Staph. schleiferi *** Vibrio fluvialis</td>
</tr>
<tr>
<td>16.2</td>
<td>3</td>
<td></td>
</tr>
<tr>
<td>16.3</td>
<td></td>
<td></td>
</tr>
<tr>
<td>C 17.1</td>
<td>2</td>
<td>Streptococcus. * C. diphtheriae</td>
</tr>
<tr>
<td>17.2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>C 18</td>
<td>1</td>
<td>Micrococcus kristinae.</td>
</tr>
</tbody>
</table>

* could not be identified to the species level.

** Coagulase positive

*** Coagulase negative
Table 5: Prevalence of Different Bacteria in Vaginal Swabs of 55 She-camels.

<table>
<thead>
<tr>
<th>Bacteria</th>
<th>Frequency</th>
<th>(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Staphylococcus</em></td>
<td>7/84</td>
<td>8.3</td>
</tr>
<tr>
<td><em>Staph. aureus</em></td>
<td>5/84</td>
<td>5.1</td>
</tr>
<tr>
<td><em>Staph. schleriferi</em>**</td>
<td>6/84</td>
<td>7.1</td>
</tr>
<tr>
<td><em>Staph. cohnii</em>**</td>
<td>2/84</td>
<td>2.4</td>
</tr>
<tr>
<td><em>Staph. warneri</em>**</td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Staph. caseolyticus</em>**</td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Staph. xylosus</em>**</td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Staph. ariettae</em>**</td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Staph. chromogenes</em>**</td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Staph. gallinarum</em>**</td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Staph. lentus</em>**</td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Staph. carnosus</em>**</td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Staph. caprae</em>**</td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Streptococcus</em></td>
<td>5/84</td>
<td>7.1</td>
</tr>
<tr>
<td>*Strept. dysgalatiae.</td>
<td>4/84</td>
<td>4.8</td>
</tr>
<tr>
<td><em>Strept. pyogenes</em></td>
<td>5/84</td>
<td>5.1</td>
</tr>
<tr>
<td>*Strept. zooepidemics</td>
<td>2/84</td>
<td>2.4</td>
</tr>
<tr>
<td>*Strept. equisimilis</td>
<td>2/84</td>
<td>2.4</td>
</tr>
<tr>
<td><em>Strept. porcinus</em></td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Strept. suis</em></td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Strept. mutans</em></td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td>*C. pseudophtherticium</td>
<td>4/84</td>
<td>4.8</td>
</tr>
<tr>
<td><em>C. diphtheria.</em></td>
<td>3/84</td>
<td>3.6</td>
</tr>
<tr>
<td>*C. pseudotuberculosis</td>
<td>2/84</td>
<td>1.4</td>
</tr>
<tr>
<td><em>C. mutuissimum</em></td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>C. cystitidis</em></td>
<td>2/84</td>
<td>2.4</td>
</tr>
<tr>
<td><em>Corynebacterium</em></td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Vibrio fluvialis</em></td>
<td>3/84</td>
<td>3.5</td>
</tr>
<tr>
<td><em>Stomat. mucilaginosus</em></td>
<td>2/84</td>
<td>2.4</td>
</tr>
<tr>
<td><em>Bacillus badius</em></td>
<td>2/84</td>
<td>2.4</td>
</tr>
<tr>
<td><em>Bacillus. mycoides</em></td>
<td>4/84</td>
<td>4.7</td>
</tr>
<tr>
<td><em>Bacillus cereus</em></td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Micrococcus kristinae.</em></td>
<td>2/84</td>
<td>2.4</td>
</tr>
<tr>
<td><em>Micrococcus sedentarius</em></td>
<td>2/84</td>
<td>2.4</td>
</tr>
<tr>
<td><em>Clostridium chauvoei</em></td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>E. coli</em></td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Morganella morganii.</em></td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Entero. faecalis</em></td>
<td>1/84</td>
<td>1.2</td>
</tr>
<tr>
<td><em>Aero. spp</em></td>
<td>1/84</td>
<td>1.2</td>
</tr>
</tbody>
</table>

*could not be identified to the species level.
** Coagulase positive
*** Coagulase negative. #: No. of isolates / Total no. of positive swabs

Table 6: Fungi Isolated from Vaginal Swabs of Camels in The Three Groups.

<table>
<thead>
<tr>
<th>Fungus isolated</th>
<th>Frequency of isolation from camels in</th>
<th>Group A</th>
<th>Group B</th>
<th>Group C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aspergillus</td>
<td>1/17#</td>
<td>0</td>
<td>1/18#</td>
<td></td>
</tr>
<tr>
<td>Mucor</td>
<td>0</td>
<td>1/20#</td>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

#: No of isolates / total no of samples.
Table. 7: Degree of Reaction for *Brucella* Antibodies in The Sera of She - camels.

<table>
<thead>
<tr>
<th>Degree of reaction</th>
<th>Number of animal (%) in each group*</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>A (52.9%)</td>
<td>B (50%)</td>
</tr>
<tr>
<td>Negative (-)</td>
<td>9</td>
<td>10</td>
</tr>
<tr>
<td>Positive (+)</td>
<td>3 (17.6%)</td>
<td>3 (15%)</td>
</tr>
<tr>
<td>Positive (+++)</td>
<td>3 (17.6%)</td>
<td>6 (30%)</td>
</tr>
<tr>
<td>Positive (+++)</td>
<td>2 (11.8%)</td>
<td>1 (5%)</td>
</tr>
</tbody>
</table>

* Number of animals in groups A, B and C were 17, 20 and 18 respectively
Table 8: Plasma Progesterone (P4) Concentration (ng/ml) in the Three Groups of Camels.

<table>
<thead>
<tr>
<th>Group</th>
<th>Pregnant</th>
<th>Infertile</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>1.16 ± 0.06</td>
<td>0.10 ± 0.01*</td>
</tr>
<tr>
<td>B</td>
<td>1.17 ± 0.08</td>
<td>0.11 ± 0.11*</td>
</tr>
<tr>
<td>C</td>
<td>1.29 ± 0.08</td>
<td>0.16 ± 0.33*</td>
</tr>
</tbody>
</table>

Data are shown as mean ± SD

Group A: Repeat breeder (17 animals)
Group B: Became infertile after abortion of first pregnancy (20 animals)
Group C: Multiparous became infertile (18 animals)

Data are expressed in mean ± SD
* P > 0.05 within rows
Table 9: Concentration of Some Blood Biochemical Parameters in The Plasma of Infertile Repeat Breeder and Pregnant Heifer (group A).

<table>
<thead>
<tr>
<th>Blood</th>
<th>Pregnant</th>
<th>Infertile</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein (g/dl)</td>
<td>7.54 ± 1.52</td>
<td>7.59 ± 1.41</td>
</tr>
<tr>
<td>Calcium (m mol/l)</td>
<td>10.36 ± 1.34</td>
<td>9.36 ± 3.14</td>
</tr>
<tr>
<td>In-organic phosphorus</td>
<td>3.43 ± 0.72</td>
<td>4.75 ± 2.14</td>
</tr>
<tr>
<td>Iron (µ mol/l)</td>
<td>16.85 ± 7.82</td>
<td>16.66 ± 13.83</td>
</tr>
</tbody>
</table>

- No. of pregnant camels (2), infertile (15)
- Data are expressed in mean ± SD
Table 10: Concentration of Some Blood Biochemical Parameters in The Plasma of Animals Became Infertile or Pregnant after Abortion of First Pregnancy (group B).

<table>
<thead>
<tr>
<th>Blood</th>
<th>Pregnant</th>
<th>Infertile</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein (g/dl)</td>
<td>8.06 ± 0.08</td>
<td>7.58 ± 0.99</td>
</tr>
<tr>
<td>Calcium (m mol/l)</td>
<td>6.91 ± 3.47</td>
<td>8.71 ± 2.15</td>
</tr>
<tr>
<td>In-organic phosphorus (m mol/l)</td>
<td>4.47 ± 2.60</td>
<td>5.10 ± 2.00</td>
</tr>
<tr>
<td>Iron (µ mol/l)</td>
<td>42.17 ± 0.00</td>
<td>14.57 ± 9.04**</td>
</tr>
</tbody>
</table>

- No. of pregnant camels (2), infertile (18)
- Data are expressed in mean ± SD
- ** P> 0.0) within rows
Table 11: Concentration of Some Blood Biochemical Parameters in The Plasma of Multiparous Animals Became Infertile or Pregnant at Sampling Time (group C).

<table>
<thead>
<tr>
<th>Blood</th>
<th>Pregnant</th>
<th>Infertile</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein (g/dl)</td>
<td>8.37 ± 0.01</td>
<td>7.36 ± 0.05*</td>
</tr>
<tr>
<td>Calcium (m mol/l)</td>
<td>5.31 ± 0.64</td>
<td>8.73 ± 2.13*</td>
</tr>
<tr>
<td>In-organic phosphorus (m mol/l)</td>
<td>7.09 ± 0.21</td>
<td>5.14 ± 1.93**</td>
</tr>
<tr>
<td>Iron (µ mol/l)</td>
<td>5.93 ± 1.07</td>
<td>14.59 ± 7.90*</td>
</tr>
</tbody>
</table>

- No. of pregnant camels (2), infertile (16)
- Data are expressed in mean ± SD
  
  * P> 0.05 within rows
  ** P> 0.01 within rows


**Discussion**

The role of the camel as domestic animal is undergoing fundamental changes as subsistence nomadism shift towards semi-sedentary cash-demanding system. Problems associated with lack of knowledge, due to insufficient research in the past, are further compounded today by the challenge of change. The camels of poor families in arid and semiarid areas must become more productive and competitive if the communities concerned are to survive.

This study was conducted to determine the prevalence and etiology of infertility in camels. Emphasis was made on the relationship between the presence of certain microorganisms in the reproductive tract of the dromedary females and nutrition and hormonal factors and subsequent infertility problems.

Different workers used different body fluids, and swabs for the diagnosis of different animal diseases at the field level (Bone, 1954; Osborne; Low and Perman 1973; Brown and Wilson, 1984). In the present study, vaginal swabs and sera were used for the laboratory diagnosis of infertility in camels. However, it was not possible to collect some samples from all suspected animals due to reluctance of the owners and lack of facilities in the field.

Different microorganisms were isolated from vaginal swabs as pure or mixed cultures; 49 (89.1%) of the 55 vaginal swabs examined gave positive bacterial cultures, 23 (46.9%) of these cultures were pure and 26 (53.1%) were mixed, whereas six swabs (10.9%) gave no growth.
As shown in tables 5 *Staphylococcus aureus* was the predominant bacterium and was isolated from five vaginal swabs (5.1%), followed by *Streptococcus* spp., *C. pseudotuberculosis*, *Bacillus* spp., *E. Coli*, enterococci, and *Micrococcus* spp. These bacteria were either isolated in pure cultures or in association with other bacteria. Similar microorganisms were isolated from camels with reproductive problems by Hegazy, Yousef and Selim (1971) in Egypt and by Domenech *et al.* (1977) in Ethiopia. Dowson (1969), Barlow *et al.* (1986) and Al Sayed (1994) isolated similar bacteria from the vagina and cervices of cows with reproductive problems. This indicates that bacteria associated with infertility in she-camels, are similar to these implicated in reproductive problems in cattle.

Several species of coagulase-negative *staphylococci* were isolated in this study, these were *Staph. shleriferi* (7.1%) *Staph cohnii* (2.4%), and *Staph. warneri*, *Staph xylosus*, *Staph carnosus*, *Staph. arlettle*, *Staph. caseolytics*, *Staph lentus*, *Staph gallinarum*, *Staph. chromogenes* and *Staph. caprae* which were isolated from 1.2% of samples. These bacteria were associated with reproductive abnormalities and probably play a role in infertility in she-camels in the Sudan.

Many species of the genus *Streptococcus* were isolated in pure cultures or in association with other microorganisms. They included *Strept. dysgalactiae*, *Strept. equismillius*, *Strept. procinus* and *Strept suis*. Two strains of *Strept. zooepidemics* were recovered (2.4%), one was in pure culture and the other was mixed with *Strept. pyogenes*. Wernery and Wernery (1992) isolated several microorganisms from uteri of she-camels, and concluded that the isolates were similar to those obtained from equine and bovine
uteri and included *Strept. zooepidemics*, a common pathogen recovered from equine uteri.

Two strains of *C. pseudotuberculosis* (2.4%) were isolated mixed with staphylococci. Mason (1917) reported a prevalence rate of 3% of *C. pseudotuberculosis* in the reproductive tract of she camels in Cairo abattoir whereas Elmossalimi (1971) reported its prevalence rate as 0.3% in Sudan. Domenech et al. (1977) reported 37% prevalence rate of *C. pseudotuberculosis* in Ethiopian camels.

*C. pseudodiphtheriae* and *C. diphtheriae* were isolated from vaginal swabs in pure cultures or mixed with staphylococci; *C. diphtheriae* is a human pathogen which causes diphtheria (Carter, 1986). The isolation of this organism from camels was unexpected and it is possible that these strains reached the vagina of examined animals from milkers or camel attendants.

Seven strains of *Bacillus* spp. were isolated in this study, *B. mycoides* four times (4.7%), *B. badius* twice (2.4%) and *B. cereus* was isolated once. Counter (1985) reported cases of abortion in cattle due to *Bacillus* spp., in particular *B. licheniformis*, in some parts of the United Kingdom. He observed that placentities due to *B. licheniformis* was similar to that following mycotic infection and was able to recover the organism from the fetus, especially the abomasum, and from the placenta and vaginal swabs. *Bacillus cereus* has been incriminated as a cause of abortion in cows (Mikesell et al., 1983).

One strain of *E.coli* was isolated from vaginal swabs in a mixed culture with *Morganella* spp. *E.coli*, like many other enterobacteria, is associated with certain infections in man and animals. Arthur et al. (1998) reported that inflammation of the vagina and cervix in cattle due to *E.coli* was likely to follow obstetrical trauma or during
relief of difficult dystocia, and in these circumstances as well as with delayed involution of the uterus and retention of after birth, it is usually accompanied with pueral metritis. Rowe and Smithies, (1978) and Moorthy (1985) reported sporadic abortion in cattle due to *E. coli*. They suggested that following stress, the organism reached the fetus and placenta via haematogenous spread or by ascending the genital tract.

Thirty two (58.18%) of serum samples examined by the RBPT were found sero-positive for Brucella antibodies, which indicates that brucellosis was prevalent in the area surveyed. This result was higher than that obtained by Palgov and Zhslobovski (1964) who reported brucella antibodies in 15% of camels in Chad. Mustafa and Awad Elkarim (1971) reported 1.8 and 5.6%, prevalence in Butana and Abu Damir *et al.* (1984) reported 4.9% infection rate in camels in both sexes, in Kassala Richard (1980) reported a prevalence rate of *Br. abortus* in 5.5% of Ethiopian camels. Zaki (1948) showed the presence of *Br. abortus* agglutinins in 14% of males and 26% of females Egyptian camels. Most of these investigators used the RBPT. It must be noted that in this study only suspected infertile she-camels were examined which may explain the high prevalence rate of brucellosis. Tibin (1985) found 7.8% of camels in Darfur were sero-positive for brucellosis and in areas where camels were reared with other animals, the prevalence rate reached 23.3% and in some localities it was high as 89.5%. He concluded that the disease in camels was due to lateral infection through direct or indirect contact with the primary hosts; cattle, goats and sheep.

It is clear that from the present study, brucellosis is quite prevalent in Eastern Sudan, where the largest population of camels is kept. Although brucella antibodies have
been demonstrated in aborting she-camels (Musa, 1990), the economic importance of brucellosis as a cause of abortion in pregnant females remains to be investigated.

The detection of brucella antibodies in the serum of camels gives warning against the indiscriminate slaughter and use of meat and milk of camels for human consumption, bearing in mind that many people believe that camel products and bi-product like meat, livers, milk and urine are remedies for many diseases. The lack of proper examination of camels before milking and slaughter could be regarded as a factor in the failure of control of some of zoonotic diseases including brucellosis.

In this study two *Aspergillus* spp. and one *Mucor* spp were isolated from vaginal swabs. These fungi were isolated by Ainsworth *et al.* (1973), Pepin (1983) and Njoku (1992) from mycotic abortion and reproductive problems in cows. Al Sayed *et al.* (1994) isolated these fungi from infected calver cows in Sudan. Mycotic infection of the reproductive tract of she-camels is relatively uncommon and is rarely clinically diagnosed. It is possible that these fungi may play a role in reproductive tract infections in camels.

Most of the microorganisms isolated in this study which include coagulase + ve and coagulase – ve *staphylococci, streptococci, E. coli, Aspergillus* and *Mucor* spp. have been isolated from she-camels with disease conditions like pyometra and endometritis.

The present study showed significant decreases in blood protein of infertile camels compared to pregnant camels of group C (Table 11). This finding confirms the observation of Rossiter (1970) that inadequate diet delays puberty causes anoestrus and depresses ovarian function in older animals. Jacobson (1978) reported that protein and energy deficiencies usually occur concurrently in underfed livestock. He emphasized that
deficiency of energy in young animals resulted in retarded growth and delayed the onset of puberty. In mature animals there was marked decline in milk production and shortened lactation.

Studies on the effect of nutrition upon reproduction in camels in the Sudan are scarce. The results of the present study concur with that of Gould (1969) who reported that high level of protein in the diet had adverse effect on fertility, because the feeding of excess degradable proteins resulted in increased ammonia and urea production in the rumen, these substances might influence the uterine environment due to elevated blood concentration, and their toxicity might affect spermatozoa, oocytes and embryos. Anderson and Davis (1958) reported that 20% of dietary crude protein in dry mater (CPDM) increased the incidence of retained placenta, dystocia and post-partum meteritis compared with 13% level. It has been suggested that there was also impaired intrauterine leucocyte function in cows fed higher level of proteins.

The present study showed a significant increase in level of progesterone in pregnant animals compared to infertile animals as well as significant decrease in blood protein level in group C. These finding are in agreement with Ferguson and Chalupa (1989) who used logistic regression analysis and found a decrease in the serum progesterone concentration as CPDM was increased. The source of dietary protein can also affect peripheral progesterone concentration (Garveick et al., 1971).

Little work has been done on minerals supplementation in camels (Ahmed, 1976). The present study showed a significant increase in iron in infertile animals of group B and C and a highly significant increase in calcium in infertile animals of group C and a significant decrease in the inorganic phosphorus in infertile animals of group C. Hignett
(1956) reported that functional forms of infertility occurred in cattle on diets with calcium to phosphorus ratio outside the range of 1:2. He observed reduced fertility and other symptoms including hypocalcemic tetany in adult cows. Manston (1964) also reported infertility and other signs as a result of phosphorus deficiency. Morrow (1969) observed retarded growth and reduced fertility as the earliest signs of phosphorus deficiency and concluded that infertility was accompanied by anoestrus and irregular oestrus and delayed sexual maturity. Call (1986) observed clinical symptoms in animals with experimentally produced phosphorus deficiency, expressed as unthriftness, marked body weight loss, reduced feed consumption and finally impaired production.

In the present study significant decrease and increase in blood iron values were recorded. Rudolphi and Pfau (1978) noted a high incidence of still births in litters of sows suffering from iron deficiency anemia. Weinberg (1984) also reported an increase in susceptibility of animals to a variety of infections when there was an excess of iron in their bodies which they were unable to bind properly.

The effect of malnutrition in fertility is likely to be general and infertility is often related to lack of total average intake of nutrients rather than to specific deficiencies. Asdell (1946) reported that, nutritional deficiencies or excesses may cause infertility by acting via hypothalamus, thus influencing the production of gonadotropin which affects the time of onset of puberty.

As shown in RIA results there was a significant increase in the level of progesterone level in pregnant animals compared to infertile animals. This is in agreement with Cristofori and Quaranto (1990) who recorded similar differences between pregnant and non pregnant she-camels; they also recorded a similar significant
difference in oestradiol – 17B level between anoestrus and cyclic subjects and its stages in she-camels.

Hormone assay results showed a high value of progesterone level in 6 (11.1%) pregnant, and low value in 49 (89.09%) of the examined animals. Xu et al. (1985) found that persistent corpora lutea were present only in pregnancy. Daffala et al. (1988) and Musa and Abu Sinenia (1978) observed high ovarian activity in camels of central Sudan during the period from March to August; this was the time of sampling in this study.

Morris (1976) suggested that most functional aberrations occur because of some endocrinological abnormalities resulting from inherited factors, nutritional deficiencies and social influences which may arise from modern husbandry methods. For instance, the grouping of large numbers of animals may interfere with the establishment of stable social hierarchy and the stray of production. As a rule functional forms of infertility tend to affect individual animals within a herd but in the aggregate they constitute an important cause of infertility, as they frequently affect other problems, especially nutrition (Drew, 1978).

In conclusion the fertility of the 55 she-camels examined was found to be unacceptably poor, because of bacterial infections. A high percentage of bacterial isolates 71(83.5%) from vaginal swabs were of significance while 16.5% of the isolates were of no significance in causing genital tract infections. Furthermore, 58.2% of the serum samples were positive for brucellosis and 5.5% of the animals were infected with fungi. There was a high degree of excess or deficiency in some blood biochemical parameters which resulted in infertility.
Recommendations

1. Establishment of efficient laboratory of bacteriology, virology and immunology in areas where a large population of camels is located, in particular Showak area.

2. Further comprehensive studies in the role of different bacteria and mycotic infection in infertility are required.

3. More investigation should be carried on the relationship between diet, husbandry and blood biochemicals deficiencies and infertility in males and females camels.
References


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