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FINE NEEDLE ASPIRATION CYTOLOGY: A QUICK AND COST EFFECTIVE CANCER DIAGNOSTIC AID

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Abstract

In canines, cancer is the primary cause of death and to tackle this disease condition, early diagnosis is the better strategy. FNAC (Fine Needle Aspiration Cytology) is a valuable, quick, minimally invasive and cost-effective technique to detect cancer early. FNAC is used to harvest samples from suspected masses for cytological examination. It is possible to differentiate between benign and malignant tumors by cytological examination based on nuclear characteristics, including their shape, size, number, and chromatin pattern. Lymphoma and Mammary adenocarcinoma are the most common type of cancer found in canines. In the case of mammary tumors, if we spay a dog before its first heat, we can reduce the chance of mammary cancer by eight-folds because it is hormone-dependent. Presently, there is no clarity about the cause of cancer, so overall prevention is difficult. But its early diagnosis can prevent cancer from spreading to the other organs.

Keywords: Canine, cytology, diagnosis, mammary cancer

Cancer is a disease condition where cells in the body starts dividing uncontrollably. If cancer is not diagnosed early, it can spread to other body parts via circulatory or lymphatic systems. In canines, cancer is the most common cause of death. Especially older dogs are more prone to develop cancer (over the age of 10) because their cells have more time to accumulate mutation and genetic abnormalities. Usually, tumors can be visualized as fleshy solid lumps under a dog's skin. But sometimes tumors are too deep within the body, and it isn't easy to recognize them. However, detection at the early stage is the only key to the successful treatment of cancer.

Now-a-days, FNAC (Fine needle aspiration cytology) has become a valuable tool in diagnosing various types of canine cancer like mammary tumors (Mendoza et al., 2011), Lymphoma, and hepatoid gland tumors. FNAC can be described as a process of generating negative pressure in a syringe (10-20 ml), and cellular material is introduced into the fine needle (22 to 25 gauge) due to the pressure difference (Fig. 1). Cytology is concerned with the microscopic evaluation of cells. This technique is generally used for palpable lesions (which can be felt by touch or sometime guided by ultrasound transduce in case of tumor of internal organs. A procedure of collecting FNA using ultrasound

transduce has been depicted (Fig. 2). The primary goal of FNAC is that by looking at the morphology of the cells, we can differentiate benign lesions from the malignant ones. The three major areas in which FNAC performs a significant role are the following: (a) Benign tumor diagnosis in symptomatic palpable nodules, (b) staging of the tumor, and (c) prognosis of metastatic disease at distant sites following cancer treatment. Accurate tumor evaluation and tissue analysis constitute the first step in most cancer therapy. Currently, precise tumor diagnosis relies upon a triple evaluation technique that includes clinical, imaging, and pathologic examinations. The accuracy of FNAC is determined by three primary factors: (a) an adequate sample of the lesion (without blood contamination); (b) appropriate processing and staining without artifact and (c) correct cytological material interpretation with a clear report. In the diagnosis of cancer, FNAC has grown in popularity due to its quick and easy approach. It is inexpensive and can be done with a few minor complications.

For cytological study, the collected FNA is released on a charged glass slide and a thin smear is made. Smears are air-dried and stained with May–Grunwald–Giemsa or another suitable stain to visualize the morphology of cells, microscopically. While for the molecular analysis, FNA is collected in appropriate reagents to protect nucleic acids (DNA/RNA) and stored in deep freezer.

Cytological Classification

The FNAC technique is used mainly for obtaining samples for cytological examination in several organs like the mammary gland, lymph nodes, salivary gland, and thyroid. The cytological classification is entirely based on the nuclear characteristics, including shape and size of nucleus, nucleolar number, and chromatin pattern as given below:



Fig. 1. Collection of FNA from canine mammary tissue using 22-G needle.

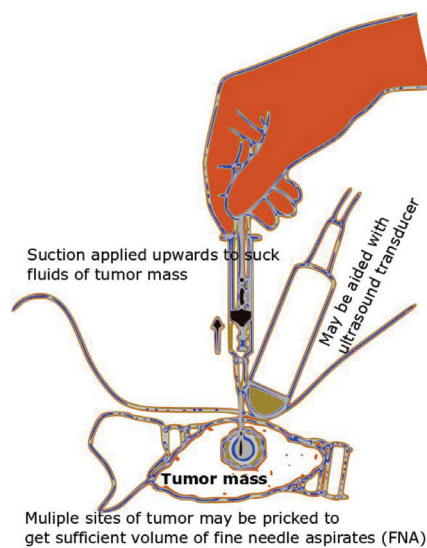


Fig. 2. Collection of FNA using ultrasound guided transducer.

S. No.	Nuclear Characteristics	Normal cell criteria	Malignant cell criteria
1.	Nuclear number and size	Single	Multinucleated and Anisokaryosis
2.	Nuclear shape	Spheroid shape	Pleomorphic
3.	Position of nucleus	Usually constant at a central position	Eccentric position
4.	N/C Ratio	Variable during cell division but not high	High due to increasing nuclear Size
5.	Nucleolar number	Ten possible nucleoli per cell.	Multiple and unequal in Size
6.	Chromatin pattern	Fine and evenly distribution	Dense and unequal distribution
7.	Mitosis (Mitosis in malignant tumors is seen as relatively rare compared to histology samples and may appear regular or irregular)	Few and appear regular	Frequent mitosis

Common Type of cancer in canine and their diagnosis through FNAC:

According to these reports of cytological classification, it can be decided whether patients have to go for major surgery or not.

- Canine lymphoma or lymphosarcoma:** Canine Lymphoma is a broad category of cancer that develops from the lymphocytes and is commonly diagnosed in dogs. Lymphoma can affect any organ in the body, but it most often begins in the lymph nodes before spreading to other organs. FNAC generally steps one in assessing an enlarged lymph node to diagnose canine lymphoma. Cytological characteristics of Lymphoma include lymphoblast predominance, irregular mitosis, degenerated nucleus due to blast, and anisokaryosis. A dog with more than one peripheral lymph node enlargement is more prone to have Lymphoma than a dog with only a single enlarged lymph node. Canine lymphoma is cytologically similar to the NHL (Non- Hodgkin lymphoma), which is cancer in humans. FNAC has a >90% sensitivity and specificity in differentiating a malignant process from reactive hyperplasia in human lymphoma (Oh et al., 2014) and thus has been used first-line diagnostics (Seelig, et al., 2016).

- **Mammary gland tumor:** Mammary tumor is primarily an adenocarcinoma, i.e., cancer of glands. In female dogs, mammary tumor accounts for 50% of all neoplasms. Mostly found in older pets, the onset age of canine mammary tumors (CMT) is between 10 and 11. Chances of the tumor increase after each estrous cycle as mammary tumors are hormone-dependent in bitches. Due to high incidence of mammary cancer in female dogs, many cases of CMT are being operated routinely in the GADVASU clinics (Figure 3), FNAC is being used in differential diagnosis of neoplasms of cats and dogs (Pavel et al., 2016). Cytological diagnosis of mammary tumor based on acinar arrangement (pyramid shape) of the hyperplastic epithelial glandular cells, multinucleated condition, irregular pattern of chromatin, anisocytosis. FNAC coupled with Robinson's method of cytological grading is recommended to evaluate malignant CMT (Dolka et al., 2018). Tumor genomic profiling is now a standard of care for many types of malignancies. Transcriptome-based expression signatures have become the standard of care in the management of early-stage cancer especially detection of estrogen-receptor negative breast cancer. These assays provide prognostic significance in the setting of adjuvant endocrine therapy.
- **Osteosarcoma:** This type of tumor usually originates from the metaphyseal region of bones of the limbs. Cytological examination of osteosarcoma shows the malignant osteoblasts, osteoclasts, and all other nuclear characteristics of malignant cells. Histopathological evaluation is also considered along with cytological evaluation for better diagnosis because sometimes there is difficulty obtaining an adequate FNAC sample from a bone lesion due to the challenge of penetrating the bone cortex. FNAC has been described as a very valuable procedure for osteosarcoma patients (Mohit et al., 2018).



Fig. 3: Inguinal mammary gland tumor in female dog (*Photo: Multi Speciality Veterinary Hospital, GADVASU*)

Prevention and Treatment

Overall, prevention of cancer is difficult because till date the exact cause of most cancers is unknown. But in the case of a mammary tumor, if we spay a dog before its first heat, we can reduce the chance of mammary cancer by eight-fold just because of the hormonal influence. There are other detection techniques like CT scan (computerized tomography), MRI (magnetic resonance imaging), Ultrasound, and X-Ray. But these

techniques detect cancer when there is a visible change in tissue. Thousands of tumor cells could have proliferated and even metastasized by that time. So, FNAC is a valuable tool in the early diagnosis of cancer. It is also less expensive compared to other techniques.

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STEP BY STEP PROCEDURE OF ULTRASOUND GUIDED BIOPSY FOR THE ABDOMINAL ORGANS IN DOGS

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Abstract

Ultrasound guided biopsy is a minimally invasive confirmatory diagnostic tool that complements the routine diagnostic modalities of clinical signs, laboratory tests and radiography. Both Free hand biopsy and guide assisted biopsy techniques yields adequate tissue sample for histopathological analysis.

Keywords: *Free hand biopsy, guide assisted biopsy, abdominal affections, ultrasound*

Application of ultrasound guided biopsy (USGB) is a helpful diagnostic approach towards various abdominal affections. Ultrasound guided biopsies are generally performed using 5-12 MHz transducer. The procedure allows direct visualisation of the biopsy needle penetrating through percutaneous tissue and then entering into the target organ thus ensuring accuracy and safety of the modality (Giordano et al., 2005). As ultrasonographic patterns are non-specific to a particular disease. Thus ultrasonography integrated with USGB is routinely indicated to confirm a diagnosis of the disease condition (Warren-Smith et al., 2012). This paper is focussed on technique of USGB using tru-cut biopsy gun / fine needle aspiration.

Two types of biopsy samples can be obtained namely fine needle aspiration biopsy using hypodermic needle ranging from 18 to 21 gauge and tissue core biopsy using larger bore tru-cut biopsy needle ranging from 16 to 18 gauges. The above type of biopsy procedures can be done either free hand ultrasound guided (Fig. 1) or through a guide attached to ultrasound probe generally called guide assisted biopsy (Fig. 2).

In free hand technique the needle or tru-cut biopsy gun is inserted through skin in an angle oblique to long axis of transducer (Fig. 1). This technique allows better flexibility as the direction of needle can be slightly adjusted to compensate the patient movement (Vijayaraghavan et al., 2011).

In Guide assisted USGB, trucut biopsy needle or a fine needle is inserted through a biopsy guide that is attached to the transducer allowing ultrasonographic visualisation of needle within the scanning plane (Fig. 2). This technique holds good for focal lesions where the trajectory angle can be traced out on the scanning plane.



Fig 1. Free hand ultrasound guided biopsy technique



Fig 2. Guide assisted ultrasound guided biopsy technique



Fig 3. Unloaded Automated tru cut biopsy gun

Tru-cut biopsy guns provide spring firing. The spring fired biopsy gun can be single spring device that fires the outer biopsy cutting canula which is preferred in some cases because inner stylet can be manually adjusted. The double spring fired biopsy devices are fully automated biopsy gun which first launch inner stylet first and then releases cutting cannula rapidly (Rothuizen & Twedt, 2009).

Step by step procedure for USGB procedure (Fig. 4)

Step 1: Perform a B-mode ultrasound scan to evaluate the organ and determine the biopsy region/site.

Step 2: Perform complete blood count and ensure platelet adequacy (Minimum platelet count of 1,50,000/ μ L) as a prerequisite to any organ biopsy to avoid possible bleeding risks associated with the procedure. Other Coagulation profile tests include Prothrombin time (PT) and activated partial thromboplastin time (aPTT). Normal PT value in dogs is less than 10 seconds and normal activated PTT values range between 15 seconds to 20

seconds. Values greater than above mentioned suggest a potential risk to biopsy procedure due to bleeding especially in case of liver biopsy.

Step 3: Patient preparation and local anaesthesia: Clip the sampling area and infiltrate 2% lignocaine at the biopsy site followed by aseptic preparation of the site as well as transducer probe contact surface using a surgical scrub or 70% isopropyl alcohol.

Step 4: Locate the biopsy site: Care must be exercised to avoid large blood vessels via Color Doppler visualisation.

Step 5: Proper needle placement: The needle should always be directed along the long axis of the transducer and always keep in mind the transducer orientation so that it gives an idea from which direction of the monitor the needle is entering (Kerwin, 1995). Generally, it is advised to insert needle from the same side of transducer pointer/notch. In guide assisted technique the angle can be pre-planned for the different location of lesions, deeper lesions require steeper angle whereas superficial lesions require shallow angles.

To facilitate needle placement make a small skin incision with a No. 11 blade.

Step 6: Needle placement: Preload the biopsy gun or biopsy needle attached to syringe. By constant visualisation of biopsy needle tip, advance the needle to the area of interest and fire the biopsy gun by single press on the button provided at the side of biopsy gun.

Step 7: Sample retrieval: After sampling the lesion, retract the cutting cannula edge by releasing halfway of the button. The sample is preserved in 10% Neutral Buffered formalin for histopathologic diagnosis.

Step 8: Inspect for haemorrhage: Every time a biopsy is made; the site should be inspected for the evidence of haemorrhage. Color Doppler can help visualise active bleeding.

Complications

- Post biopsy bleeding
- Hematoma at biopsy site
- Local or generalised peritonitis
- Haematuria

Contraindications

There are some contraindications for biopsy that involves any hemorrhagic tendencies, critically ill patients with compromised respiration, large volume ascites, vascular tumour, microhepatia and coagulopathies. It is advised to avoid tissue core biopsy from spleen as there is fair chance of bleeding. Likewise avoid biopsy of gall bladder as it leads to peritonitis

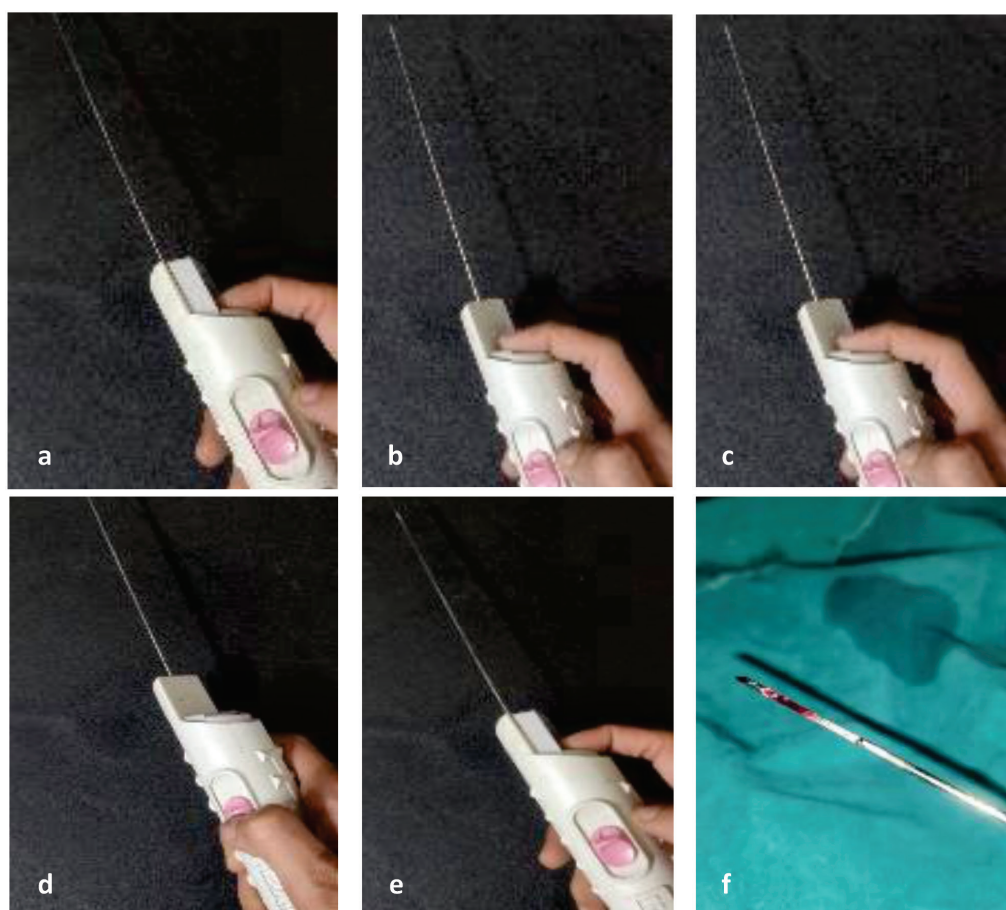


Fig 4. Stepwise procedures to be followed while using Tru cut biopsy gun for an organ biopsy. a). Press down the first trigger button; b). Press down the second trigger button; c). Biopsy gun ready to penetrate through the skin. d). shoot the biopsy gun by pressing the side button; e). press down the first button to check if sample is present in the stylet ridge of biopsy needle; f). biopsy sample in the stylet ridge.

Possible complications can be avoided by investigating coagulation profile of the patient prior to biopsy and aseptic measures taken while performing biopsy would help to avoid peritonitis. It is also important to restrict physical activity of the patient for at least thirty minutes post biopsy.

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SERUM ENZYMES OF DIAGNOSTIC SIGNIFICANCE IN FARM ANIMALS

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Abstract

A comprehensive blood profile is vital for disease diagnosis in farm animals. Most of the vital organs in the animals harbour specific enzymes which vary between the species and their levels are reported to rise in the blood with tissue damage. The literature present here discusses diagnostic enzymes of major as well as minor significance, specific to large animals, which would find immense application for veterinarians to diagnose organ functions and diseases in farm animals.

Keywords: *Diagnosis, enzymes, farm animals, organ dysfunction*

The analysis of various blood parameters is a routine part of clinical evaluation of animals. Blood or serum tests reveal the health status of the animal and provide a true reflection of the functional or metabolic status of the tissues underneath. Tissue damage causes the enzymes to leak out of their tissue cells into the blood, often resulting in a remarkable spurt in their plasma levels which can be used for diagnostic purposes. Their rate of appearance and disappearance from plasma can, thus, help in anticipating the future course of disease and can be used to dictate timely therapeutic interventions. In the light of such significance, routine assessment of these tissue specific enzymes becomes crucial and indispensable to a well-rounded and effective clinical approach.

Species differences have been reported in enzyme activity and distribution in different tissues. Genetic variation has been reported in cattle, sheep, human beings and other species which is influenced by ecological conditions. Given below are the serum enzymes which constitute an important diagnostic palette for routine clinical assessment.

A. Aminotransferases

Aspartate Amino Transaminase (AST) or Serum Glutamate Oxaloacetate Transaminase (SGOT) and Alanine Amino Transaminase (ALT) or Serum Glutamate Pyruvate Transaminase (SGPT) are the most requested enzymes for disease diagnosis. The presence of these enzymes in diverse tissues such as heart, liver, skeletal muscle,

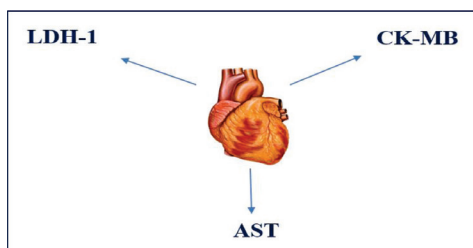
kidney and erythrocytes renders the discretion of clinician in differential disease diagnosis extremely important (Kaneko *et al.*, 2008)

a) SGOT or AST

Clinical significance in Large Animals

Hepatocytes of mature horses, cattle, sheep and pigs contain significant levels of AST (GOT), in the cytosol as well as mitochondria. Discussed below are the causes of increased AST:

1. In horses: Increasing age, excessive training, paralytic myoglobinuria
2. In sheep: myodegeneration, ingestion of toxic plants and carbon tetrachloride poisoning, white muscle disease in lambs
3. In poultry: Hereditary muscular dystrophy in chickens
4. Other reasons include *in-vitro* haemolysis, severe haemolytic anaemia, after surgery, circulatory failure with shock and hypoxia, acute viral or toxic hepatitis, cirrhosis (Hoffmann & Philip, 2008)



Note: In large animals, elevation of AST is more specific than that of ALT in evaluating hepatic disorders.

Analyte	Unit	Horse	Cow	Sheep	Goat	Pig
(AST, GOT)	U/L	226–366	78–132	60–280	167–13	32–84

b) SGPT or ALT

The presence of high concentrations of ALT in the cytoplasm of hepatocytes of dogs, primates and some other small animal species, imparts it specificity in those species for liver disorders. Minimal ALT activity is noted in large domestic animals and hence, it is not used for the evaluation of hepatic diseases in horses, cows, sheep, goats and pigs

Causes of increased plasma concentrations include:

1. Hepatocellular damage/necrosis, hepatocyte proliferation, hepatocellular degeneration
2. Corticosteroid treatment
3. Muscle damage or degeneration, alongside increased CK

Analyte	Unit	Horse	Cow	Sheep	Goat	Pig
ALT (SGPT)	U/L	3–23	11–40	10–30	6–19	31–58

B. Serum Alkaline Phosphatase (ALKP)

Clinical significance in Animals

The various isoenzymes of alkaline phosphatase are present in high concentrations in liver, bone (osteoblast), placenta and intestinal epithelium in dogs, cats, horses and ruminants (Divers & Barton, 2018). However, consistent values are obtained for dogs and cats and thus, it is used as a marker of liver and bone disorders in these species. The normal serum alkaline phosphatase activity varies considerably in ruminants and its utility as a diagnostic enzyme is limited. Increase in ALKP levels can be observed under physiological as well as pathological conditions.

Physiological increase seen during: Bone growth, pregnancy, lactation, aging, after fatty meal

Pathological increase associated with:

1. **Hepatic:** Acute hepatitis (viral and toxic; 2-5 fold increase), cirrhosis, cholestatic lesion (stones and tumors), granulomatous inflammation, abscesses. (Kataria *et al.*, 2011)
2. **Osteoblastic:** Bone metastasis, rickets, bone fracture, acromegaly, osteomalacia, Paget's disease of bone)
3. **Other causes include:** Renal origin, hyper-parathyroidism, malnutrition, induced by various drugs



Analyte	Unit	Horse	Cow	Sheep	Goat	Pig
ALKP	U/L	143–395	0–488	68–387	93–387	118–395

C. γ -Glutamyl Transferase or GGT

The predominant levels of the serum enzyme, GGT, are derived from the biliary tract (bile duct epithelium). Low levels of this enzyme are also associated with kidney (renal brush border) and pancreas. Thus, mainly it is useful in the diagnosis of hepatobiliary disorders, especially which are of cholestatic nature. Ruminant serum generally has higher levels as compared to dogs and cats.

High plasma activity is due to:

1. Cholestatic liver disorders (sensitive and specific marker of cholestasis and bile duct proliferation in horse, cattle, sheep and pigs)
2. Induction by anticonvulsant drugs (phenytoin), glucocorticoids (prednisolone)

Note: Generally, liver diseases without cholestasis will not have marked elevation in GGT levels, although ALT and AST may be elevated.

Analyte	Unit	Horse	Cow	Sheep	Goat	Pig
GGT	U/L	4.3–13	6.1–17.4	20–52	20–56	10–60

D. Glutamate Dehydrogenase (GDH)

GDH, a mitochondrial enzyme, is used as a marker of liver diseases in cattle. Although, the level of transaminases increase significantly during liver injury as compared to GDH, it could be useful in differential diagnosis as former could be elevated in other disorders.

Analyte	Unit	Horse	Cow	Sheep	Pig
(GDH)	U/L	0–11	31	20	0

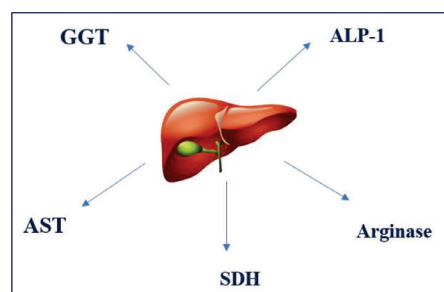
E. Sorbitol Dehydrogenase (SDH)

Sorbitol dehydrogenase, also called as L-iditoldehydrogenase, is a reliable indicator of acute liver cell lesions in ruminants while its serum levels in dogs and horses are lower than in other domestic animals. Its levels rise significantly within 6 h of the acute hepatic intoxication. The half-life is higher in sheep cattle and goats as compared to horses.

Analyte	Unit	Horse	Cow	Sheep	Goat	Pig
SDH	U/L	1.9–5.8	4.3–5.3	5.8–27.9	14.0–23.6	1.0–5.8

F. Arginase

The activity of arginase in liver is higher than in other organs and elevated serum arginase levels are observed in acute liver injury for horses, cattle, sheep and goats. There is a rapid rate of increase and decrease in plasma following hepatocellular injury; prolonged rise is indicative of a grave prognosis. (DeNotta & Divers, 2020)



Analyte	Unit	Horse	Cow	Sheep	Pig
Arginase (ARG)	U/L	0–14	1–30	0–14	0–14

G. Creatine Phosphokinase or Creatine Kinase (CPK or CK)

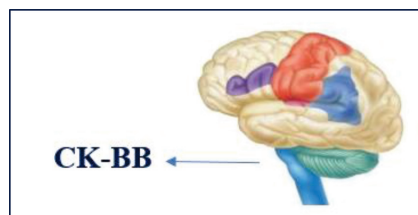
Clinical significance

The three isoenzymes of CK are composed of 2 different subunits: M, muscle and B, brain. They are localised in different tissues (CK-MM in skeletal muscles, CK-MB in myocardial muscle and CK-BB in brain). Plasma levels are mostly associated with CK-

MM which is of muscular origin.

Elevated CK is seen in:

1. Myocardial infarction (CK-MB): 30 fold rise within 24 hours of chest pain
2. In myopathy, nutritional myopathy (5-10 fold rise along with increased LDH and aldolase levels), muscle anoxia, prolonged recumbency, myositis
3. Excessive exercise
4. Muscle necrosis after intramuscular injections
5. CNS damage (elevated CK-BB)



The half-life of CK is very short and levels decrease rapidly.

Elevated CK values indicate active or recent muscle damage.

Analyte	Unit	Horse	Cow	Sheep	Goat	Pig
(CK)	U/L	2.4–23.4	4.8–2.1	8.1–12.9	0.8–8.9	2.4–2.5

H. Lactate Dehydrogenase (LDH)

The five different isoenzymes of LDH, each with four polypeptide chains (H, heart and M, muscle) (HHHH (LD1), HHHM (LD2), HHMM (LD3), HMMM (LD4), and MMMM (LD5) occur in a variety of tissues, hence, it is not a tissue specific enzyme and assessment of its levels in isolation can be misleading.

Clinical significance

Elevation in LDH is noted in the following conditions:

1. After myocardial infarction (LD1 and LD2)
2. Acute leukemia (LD2 and LD3)
3. Liver or skeletal muscle damage (LD5)
4. Chronic glomerulonephritis
5. Systemic lupus erythematosus
6. Diabetic nephrosclerosis
7. Bladder and kidney malignancies
8. Haemolysis (increased levels RBC's)

Analyte	Unit	Horse	Cow	Sheep	Goat	Pig
LDH	U/L	162–412	692–1445	238–400	123–92	380–634

I. Aldolase:

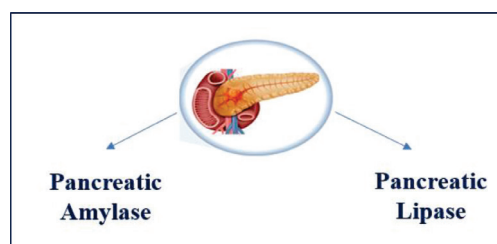
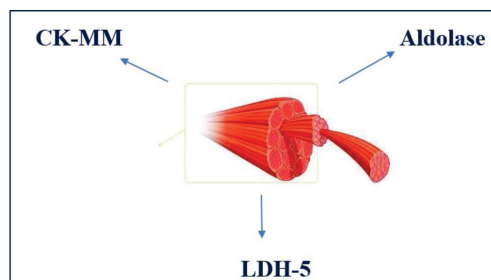
Aldolase is present most significantly in skeletal and heart muscle. Increased levels are observed in skeletal muscle damage (progressive muscular dystrophy).

J. α -Amylase

α -Amylase is present in high concentrations in pancreatic juice and in saliva in the digestive systems of humans and many other mammals. It catalyses the breakdown of starch and glycogen to maltose. Its high levels are associated with acute pancreatitis.

K. Lipase

Lipase is found in multiple tissues like gastric, pulmonary and intestinal mucosa but the highest concentration is seen in pancreas. Lipase levels tend to increase in pancreatitis and are preferred over amylase for the diagnosis of pancreatic dysfunction as it is absent in the saliva.



Analyte	Unit	Horse	Cow
Amylase	U/L	3-8	14-50
Lipase	U/L	7-16	5-13

Biochemical parameters are underutilized for evaluation of diseases in large animal practice. Appropriate usage of these enzymes is imperative for determining organ function tests and rational diagnosis. These enzymes could be employed in preclinical diagnosis, assessing progression of diseases, prognosis of metabolic and infectious diseases and monitoring response to therapy.

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AN OVERVIEW ON THE DIAGNOSIS AND TREATMENT OF BOVINE BABESIOSIS

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Abstract

Bovine babesiosis is an economical tick borne haemoprotozoan infection in the tropical and subtropical countries primarily caused by Babesia bigemina and Babesia bovis, transmitted by one host tick, Rhipicephalus (Boophilus) microplus. Although being allied closely and transmitted by the same ticks, B. bovis and B. bigemina cause oddly diverse form of the disease; haemolysis and circulatory disturbance. Most of the clinical signs of haemoprotozoan infections are overlapping like fever, anaemia, and emaciation common, hence the diagnosis is of paramount significance. The diagnostic techniques employed for the direct detection of the parasite are conventional microscopic examination of stained blood smears, nucleic acid based assays and in vitro cultivation of the parasites. For epidemiological surveys, immunodiagnostic tests like ELISA, IFAT are commended. Common chemotherapeutic agents used are diminazene diaceturate and imidocarb globally. The integrated control measures including chemotherapy, immunization and vector management is the crucial aspect of babesiosis.

Keywords: *Babesia, buffalo, diagnosis, haemolysis, haemoprotozoan parasite*

Bovine babesiosis, a tick borne disease caused by haemoprotozoan parasite of the genus *Babesia*, belongs phylum Apicomplexa, suborder Piroplasmida, class Sporozoea, family Babesiidae. The main species of *Babesia* affecting the bovines are *B. bigemina*, *B. bovis*, *Babesia major* and *Babesia divergens*. Among these *B. bigemina* and *B. bovis* are the most common species that affect cattle and buffaloes of the tropics and subtropics including Asia (Bock et al., 2004). *Babesia bigemina* is mainly infecting the bovines of the Indian subcontinent (Chauvin et al., 2009). The disease is also known as Texas fever (because disease was of immense importance in USA at one time), Cattle tick fever, Red water fever or piroplasmosis.

Both the species of *Babesia* is transmitted by *Rhipicephalus (Boophilus) microplus* as main tick vector. The national annual economic loss in livestock is about 57.2 million US dollars (McLeod & Kristjanson, 1999) and globally 1.2 billion cattle population at risk of the disease. These fiscal impacts are a consequence of direct losses through mortality, reduction in meat and milk yield and indirect losses through the expenditure incurred on the control measures.

Life Cycle, Pathogenesis and Clinical Signs

After the bite by tick multiplication of *Babesia* organisms in the vertebrate host occurs in the erythrocytes by a binary fission or budding process (schizogony) to form two, four or more trophozoites. These are liberated from the erythrocyte called merozoites and invade the other cells until a large percentage of erythrocytes are parasitized. Preferential invasion of young erythrocytes by merozoites occurs in acute *B. bigemina*. Ticks acquire the infection after ingesting the immature gametocytes from the infected cattle and buffaloes, gametogony and sporogony process takes place in ticks. Transovarian transmission occurs in one host *R. microplus* in ticks, *Babesia* organism through the ova transferred to the next generation.

Babesia bigemina and *B. bovis* are the large and small forms of piroplasms, respectively transmitted by the common *Rhipicephalus (Boophilus) microplus*, cause disease remarkably by two principle mechanism; hemolysis and circulatory disturbance. In *B. bovis* infections, the disease pathology can be both due to over production of pro-inflammatory cytokines and the direct effect of red blood cell destruction by the parasite. The pro-inflammatory cytokines from the macrophages results into vasodilation, hypotension, oedema, increased capillary permeability, coagulation disorders, circulatory stasis, endothelial damage to brain and lungs leads to cerebral babesiosis and a respiratory distress syndrome. Progressive haemolytic anaemia (macrocytic and hypochromic) and haemoglobinuria in acute cases is often present during the course of *B. bovis* infections. In subacute infections, clinical signs are less pronounced and sometimes difficult to detect.

In *B. bigemina* infections, pathogenesis is almost entirely similar except coagulation disorders, cytoadherence and hypotensive state seen in *B. bovis* (Bock et al., 2004). The pathology relates more directly to the destruction of erythrocytes. Haemoglobinuria is present earlier and is more consistent than *B. bovis* infection. There is no cerebral involvement and recovery in non fatal cases is usually rapid and complete. However, in some cases the disease can develop very rapidly with sudden and severe anaemia, jaundice and death that may occur with little warning (Bock et al., 2004). Passively acquired immunity from colostrum lasts for two months followed by innate immunity from three to nine months of age. Thus calves exposed to babesiosis early in life rarely show clinical signs and recovered cases remain symptomless carriers for years with the duration of infection being breed dependent.

Prevalence Status in Punjab: Based on the microscopic examination of the Romanowsky Stained blood smears, several case reports and studies on prevalence of *B. bigemina* infection from different districts of the state are in range 0.6-46.66 percent reported (Kaur et al., 2021; Bal et al., 2016). Molecular prevalence of *B. bigemina* cattle and buffaloes in

the Punjab varied 3.96-30.39% by PCR and nested PCR, respectively (Kaur et al., 2021; Bhat et al., 2014). Nested PCR has the sensitivity to detect the latent infections of *B. bigemina* with one infected erythrocyte in 10^8 cells (Figueroa et al., 1992). Seroprevalence of babesiosis showed presence of antibodies in 21.0% of buffaloes by indirect fluorescent antibody test (Singh et al., 2009) and 30.0 % by indirect ELISA (Kaur et al., 2016)

Diagnostic techniques

Microscopy: The thin blood smears stained with Giemsa stain is routinely used for the detection of the clinical cases of babesiosis. For the best results, blood smears should be prepared from peripheral circulatory system in *B. bovis*, however in *B. bigemina* infections, parasitized cells evenly distributed throughout the blood circulation. Thick blood smears are ten times more sensitive than thin blood smears and are therefore, very useful for the detection subclinical or latent cases with low level of parasitaemia. *Babesia bigemina* is larger form, 2 to 5 μm in length pear shaped piroplasms arranged in erythrocytes in acute angle. *Babesia bovis*, small form having size less than 2 μm and piroplasm present in obtuse angle. In case of mortality, smears made from brain, kidney or liver within 2-3 hours provide the confirmation to know the cause of death. The presence of *Babesia* in tick vector can also be detected by presence of kinete stages from haemolymph smear preparation after cutting a leg of the tick and subsequently stained as a regular blood smear.

Serological assays: Among various immunological techniques, the most sensitive and specific recommended test for the certification to animals from the endemic area of *Babesia* species is indirect florescent antibody test (IFAT). The limitation of IFAT is laborious with low sample output. This limitation overcomes by ELISA that added accuracy, higher sensitivity and ease of large scale handling of samples. The immunochromatographic test (ICT) detects antibodies against a specific antigen in a small amount of serum by means of specific antibody and a recombinant antigen both impregnated on a nitrocellulose membrane. ICT a strip based pen side rapid diagnostic test for farmers and veterinarians (Tayebwa et al., 2020), easy to perform and interpret, does not require a trained technician or any special equipment and is very cost effective.

Molecular assays: Molecular methods aimed to detect the direct presence of the *Babesia* parasite through nucleic acids, have gained the potential over the recent years inherits the property of high sensitivity and specificity and are very useful when conventional and immunological test fails. A number of different approaches including conventional PCR, nested PCR, multiplex PCR have been developed to detect latent cases of *Babesia* sp. in their hosts and vectors. PCR based techniques are reported to be as much as 1000 times more sensitive than microscopy for detection of *Babesia* sp., with detection at parasitaemia levels ranging from 0.001% to 0.0000001% (1 parasite in 10^9 erythrocytes) (Criado-Fornelio, 2007). PCR assays generally not well suitable for large large-scale

testing, due to the cost and time consumption as it requires the post PCR analysis in form of gel electrophoresis and chances of cross contamination. Contrary are useful as confirmatory tests for species identification and for regulatory testing in some cases. The post PCR analysis and contamination limitation overcome by Real-time PCR (qPCR) that retains the ability to quantify and detect the parasitaemia in real reaction.

Differential diagnosis

- In babesiosis pear shaped piroplasm present in the RBCs in Giemsa stained blood smear, affects the adults than young calves. Postmortem (PM) findings show urine bladder filled with coffee colour urine.
- Leptospirosis: Contrary to babesiosis it is more common in young animals than in adults. High mortality, haemoglobinurea and abortions are reported. It can be treated with streptopenicillin 5 gram daily for 5 days.
- Anaplasmosis: In anaplasmosis urine dark brown in colour, no haemoglobinurea, and in case of death, postmortem findings show gall bladder full of bile pigments and jaundice
- Plant poisoning: hematuria no haemoglobinurea.
- Post parturient haemoglobinurea: normal or subnormal temperature occurs 2-4 weeks after parturition or calving, no ticks on the body, response to phosphorus therapy.
- Eperythrozoonosis: clinical signs of eperythrozoonosis are not only rare but are liable to be escape unnoticed due to milder nature.

Chemotherapy

Successful treatment depends on early diagnosis and prompt administration of effective drugs. The most commonly used chemotherapy for *Babesia* infections in cattle is diminazene aceturate (Berenil®, Hoechst Ltd.) @ 3-5 mg/kg intramuscularly (IM). It has fast action for *B. bigemina* and can protect cattle for 2 to 4 wks. Imidocarb @ 1-3 mg/kg subcutaneously (SC) gives protection from *B. bovis* for 4 weeks and *B. bigemina* for two months. The indiscriminate use of anti-*Babesia* prophylactic agents including the administration of the drug at sub lethal blood levels to animals can produce the development of drug resistant parasites. Recently, several new pharmacological compounds; atovaquon (1 mg/kg; sc), amicarbalide (5-10 mg/kg; SC) developed and evaluated that are under research trials, offering new options to control the disease. Quinuronium and acridine derivatives are also found effective. The prevention and control of bovine babesiosis with aim by integrated approaches including immunization, chemoprophylaxis and vector control and exploitation of breed resistance and the development and maintenance of enzootic stability.

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METHODS OF PREGNANCY DIAGNOSIS IN CANINE: AN OVERVIEW

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Abstract

Accurate evaluation of canine pregnancy includes confirmation of pregnancy, litter size and assessment of fetal viability. The abdominal palpation and radiography have been extensively used traditionally to diagnose pregnancy and assess litter size in dogs. Besides, in current time, hormonal assays and blood tests such as hematological evaluation are also used for pregnancy diagnosis in canines as well as felines. In this manuscript, recent and realistic updates pertaining to the canine pregnancy diagnosis in canines has been discussed.

Keywords: Dog, palpation, pregnancy, ultrasound

Early pregnancy diagnosis is commonly requested by breeders to enable them to schedule their holidays to coincide with the arrival of the litter of puppies, to decide whether they should mate additional female dogs in the kennel or simply out of curiosity. When a method of pregnancy diagnosis is being chosen, the most important criteria are the accuracy of the method, the practicality of the method, and the window of pregnancy during which the method is accurate. Pregnancy diagnosis in the dogs can be based on behavioral, physical, or hormonal changes or on precise clinical examination involving palpation or imaging of uterine contents. Therefore, addressing these, different methods for diagnosis of pregnancy in canines are discussed in this manuscript.

1. Visual signs

Some early signs of pregnancy may be recognized easily by the owner, such as a persistent swelling of the vulva after estrus and a slight enlargement of the nipples, which also become pinker in color from the third week of pregnancy. These signs are easiest to recognize in the primiparous female dog. Enlargement of the mammary glands is usually observable from the fifth week (but this may also be a sign of pseudo-pregnancy). Finally, signs of malaise may occur during the third week coinciding with implantation of the embryos and/or during the fifth week as a result of pressure from the distended uterine horns on the stomach and liver. The malaise normally only lasts for a day or two and it is important to inform breeders that a longer duration may be an indication that there is a problem and the female dog should be examined. At the time of implantation, a slight

mucoid or hemorrhagic discharge may occasionally be observed from the vulva, but this should be considered normal if it is a small volume and persists for just a day or two. Distension of the abdomen is often obvious from the fifth week, and fetal movements can be seen and palpated from week 7.

Table 1: Chronological events of the development of the embryo and fetus in the dog relative to the LH peak

Event	Day with respect to LH surge
Oocyte present in the uterine tube	3 to 5
Fertilization	3 days
Migration of early embryo into the uterus	4 to 8
Non-fixed, mobile uterine stages of the blastocyst	9-13 to 20-21
Implantation	18 to 20
Formation of embryonic vesicles	18 to 23
Individual gestational sacs	20 to 30-35
Confluent gestational sacs	30-35 to 45
End of embryogenesis	30-32
Beginning of ossification	40-42
Beginning of mammary development	30-42
Detection of fetal movement with transabdominal palpation	55

2. Manual palpation

Diagnosis of pregnancy by palpation of the fetal swellings of the uterine horns can be performed between the third and fifth weeks of pregnancy. At 3rd weeks the fetal swellings are approximately 15 mm in diameter. They are round in shape, firm and well separated from each other, like a string of hazelnuts along the uterine horns. At this early stage they are carried high in the abdomen. After 4 weeks they are approximately 25 mm in diameter and somewhat more oval in shape. They are still well separated from each other in the uterine horns, which by now as a result of the increase in weight have attained a more central position in the abdomen. At 5 weeks the uterine swellings are 30–35 mm in size, oval in shape and becoming soft to the touch owing to the increase in fetal fluids. They can no longer be palpated as separate structures, and are more ventral in position. After 5 weeks of pregnancy the increase in fetal fluids and size of the uterine horns makes it difficult to determine pregnancy by palpation. Abdominal palpation is best performed during days 22-30 of pregnancy. Mineralized pups may be palpable late in gestation. Abdominal palpation is difficult or even impossible in obese animals and in animals that are tense when their abdomen is palpated. This technique has poor accuracy for

determination of litter size, particularly during the last trimester of pregnancy. Abdominal palpation has been reported to be 88% accurate for pregnancy diagnosis in the female dog. The most common mistakes are false-positive results from palpation of a stool-filled colon and segmental uterine enlargement resulting from pyometra.

3. Ultrasonography

The earliest stage possible for the diagnosis of pregnancy depends on the quality of the equipment, the frequency of the transducer used, patient characteristics (e.g., obesity, nervousness, breed, age), and most important, the expertise of the operator. Using ultrasound, the fetal and the surrounding structures can be identified from approximately day 17 of pregnancy. Using a simple Doppler ultrasound instrument the fetal heartbeats (fetal viability) can be detected from approximately day 24–28 of pregnancy. However, because of the variation in the timing of mating during estrus it is recommended to repeat the examination. Ultrasonographic imaging of the uterus is done trans-abdominally with a sector or linear transducer with a frequency of 5.0 or 7.5 MHz depending upon the patient size (Balaji et al., 2018). Ultrasonographic examination usually is performed with the female dog in dorsal recumbency. Standing examination also may be performed and has the advantage of reducing the distance between the fetus and the probe. With either positioning, the hair of the abdominal wall needs to be clipped from the pelvis to the umbilical scar, and coupling gel should be applied to improve image quality. Next, the probe is placed on the linea-alba just cranial to the pelvic brim. The urinary bladder is visualized as the primary landmark. The uterus usually is detected dorsal to the bladder. The non-pregnant uterus is difficult to visualize, whereas the pregnant uterus is relatively easy to image. Each uterine horn can be examined along its entire length. Diagnosis of pregnancy with ultrasound is possible as soon as the embryonic vesicles can be identified as discrete anechoic spherical structures. As with manual palpation, ultrasonography cannot be used to assess the exact number of fetuses' present, although early examinations are generally more accurate than later examinations. Gestational sacs can be identified reliably after about day 25 of pregnancy by using abdominal ultrasonography (Table 1). Beating fetal hearts are first visible between days 22-29, and fetal movement can be identified after about day 28 of pregnancy (Kustritz, 2005). The embryonic limb buds and choroidal plexus of the brain are sufficiently differentiated and identifiable by day 32 of pregnancy. The identification of the head, trunk, and abdomen is possible by day 35. The hyperechoic fetal skeleton is evident from day 33 onward. The heart valves and aorta become easily identifiable at this time as well. The diameter of the trunk exceeds that of the head after day 40 of pregnancy and at this stage of pregnancy, other organs become recognizable; the lungs, diaphragm, liver, and stomach are the most easily detected (Aissi & Slimani, 2008). During the last 3 weeks of pregnancy, the kidneys, fetal vasculature (including umbilical vessels), and intestines are detected. Real-time (B-mode)

ultrasonography is the best technique for assessing the fetal viability. Also using ultrasonography fetal age can be calculated based on measurement of the bi-parietal or trunk diameter.

However, if the estimation is limited to whether the female dog is carrying five pups or more versus four pups or fewer, the predictive value of ultrasonography increases to 100% and 83.2 %, respectively. The best stage for determination of litter size is between days 25-35 after implantation. One of the problems encountered in early pregnancy detection and fetal numbering is the incidence of fetal loss. Fetal resorption is known to occur throughout pregnancy without effect on the adjacent conception. Embryonic resorption is recognized by the low volume and increased echogenicity of embryonic fluid, loss of embryonic mass and heartbeat, collapse of the conceptus with thickening of the uterine wall, and reduced size in comparison with adjacent conception.

4. Radiography

Radiography is an accurate method of pregnancy diagnosis, but only during the later stages of pregnancy. Radiography allows accurate determination of litter size, fetal abnormalities, and fetal size and their positions. It is therefore the method of choice for a pre-partum examination. Although the outline of the pregnant uterus may be observed as early as day 21, pregnancy cannot be differentiated at this time from other causes of uterine enlargement.

A positive diagnosis requires detection of fetal calcification, which is generally not observed until after day 42 (Fig.2). In practice, it is better to wait until days 45-50 after the LH peak to increase the accuracy of diagnosis and determination of fetal number. Determination of fetal number is based on enumeration of skulls and associated vertebral columns. Fetal viability should be questioned if radiographs show intrauterine gas pockets or misshapen fetal skeletons. Frequent use of radiology is not recommended during the same pregnancy. Exposure of the fetus to radiation may cause abnormalities, particularly during the organogenesis phase of fetal development



Fig 1: Ultrasonography of a 35-day pregnant female dog. Y denotes yolk sac part of placenta

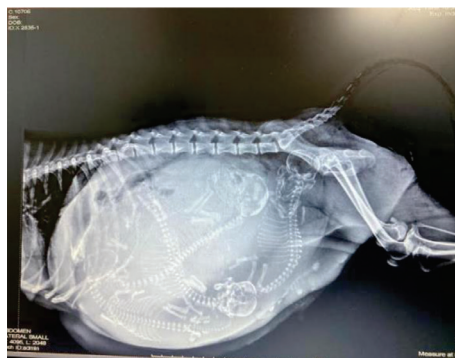


Fig 2: Radiograph of near-term pregnancy illustrating fetal skulls and vertebral columns in a female dog.

(30-35 days). Prediction of dystocia resulting from feto-maternal disproportion has been suggested based on measurement of fetal skulls in relation to the maternal pelvis, but the usefulness of this is debatable.

5. Laboratory methods

5.1 Hematologic changes

Normal packed cell volume (PCV) in the non-pregnant female dog during diestrus is 40%-55%. A decrease in PCV is observed in pregnant female dogs starting at approximately day 20 after the LH peak. Around day 35 and near term, PCV may be below 40% and 35%, respectively. However, this parameter has limited diagnostic utility because of the large variability between the animals. Pregnant female dogs usually have a physiologic normocytic, normochromic anemia. Mean corpuscular volume and hemoglobin concentrations remain unchanged. This anemia probably results from hemodilution because of increased plasma volume.

5.2 Measurement of serum proteins

A significant increase in the serum C-reactive protein (CRP) is observed in pregnant female dogs between days 30-50 of pregnancy. However, currently homologous canine CRP assay is unavailable commercially. Fibrinogen, though, increases significantly in pregnant female dogs after implantation and peaks at approximately mid-gestation. Fibrinogen concentrations of greater than 280-300 mg/dl are consistent with pregnancy. Gentry & Liptrap (1981) observed a threefold rise in serum fibrinogen concentrations during pregnancy, with peak values occurring 4 to 5 weeks after mating. Because this phenomenon did not occur at the corresponding stage of the luteal phase in non-pregnant female dogs, it can be used as a method of detecting pregnancy. Eckersall et al. (1993) reported an acute phase response in pregnant female dogs as demonstrated by the rise in serum CRP in mid-gestation in response to the implantation of the embryo causing tissue damage. However, precise mating dates are required for these tests. There is possibility of false positive results if there is infection and inflammation elsewhere.

5.3 Measurement of hormones in blood

Relaxin, produced by canine placenta, is the only gonadotrophic hormone known to be specific for pregnancy. In the female dog, relaxin is detected from the third week of pregnancy. Plasma concentration peaks at 4-5 ng/ml at days 40-50 of pregnancy. Plasma relaxin concentrations decrease slowly after parturition but remain at detectable levels for the first 4-9 weeks postpartum. In-house enzyme-linked immunosorbent assay (ELISA) kits can be used from around 25 days after ovulation to measure plasma relaxin concentration. These kits are generally very accurate and are useful under field conditions. In the case of an early negative result, the female dog should be retested 5-6 days later. The persistently elevated progesterone concentrations in the peripheral blood are used to

detect pregnancy in the polyestrous species, but this is of no value in the dogs because of the prolonged luteal phase (pseudo-pregnancy) in non-pregnant animals.

Pregnancy in dogs could be diagnosed by observing behavioral and physical changes, manual palpation, radiography and ultrasonography. Behavioral and physical changes may be misleading owing to similar changes in pseudo-pregnancy. Manual palpation is the cheapest with optimum accuracy during 22 to 30 days after fertilization. Radiography is useful after calcification i.e. day 42-45 after fertilization in determination of fetal number and positions. In all techniques, ultrasonography is the gold standard test for diagnosis of pregnancy as well as fetal viability.

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DIAGNOSIS AND MANAGEMENT OF CORNEAL ULCERS IN DOGS

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Abstract

Corneal ulcer is usually a non-healing corneal pathology which ultimately participates in vision impairment. If untreated, can leads to Pthesis bulbi and demands eye enucleation. This article deals with fundamentals of corneal anatomy, healing of damaged cornea and corneal ulcers (types, diagnosis and management).

Key words: *Cornea, healing of corneal injury, treatment of corneal ulcers*

Avascularity and transparency is very characteristic feature of cornea and such feature should be well maintained otherwise vision impairment occurs. Healthy cornea is dehydrated, multilayered and devoid of blood vessels. As compared to other body tissues, cornea responds very differently against the pathological stimulus (Neto et al., 2018). Because of avascularity, cardinal signs of ocular inflammation are manifested by different ways. Generally hyperaemia take place immediate after injury in majority of body tissues but especially in eyes hyperaemia does not occurs. The aim or function of hyperaemia in eyes after injury will be done through ocular fluids (aqueous humour and tears) that are produced by ciliary body and conjunctiva; therefore a clinician must be aware about anterior uveitis, conjunctivitis and epiphora. Inflammation within the eye (endophthalmitis) is very serious problem as compared to other body tissues because inflammation leads to adhesions in between the ocular structures and ultimately damages the eye (Slatter, 2003). Transparency is not significant for body tissues excluding cornea because presence of scar is not a worrying matter for other tissues. In case of corneal repair, if even pin point scar (nebula) is left then dog will definitely suffers.

Direct corneal insult (like cat scratch), systemic diseases (adenovirus causes bilateral corneal insult), changes in intraocular pressure (damage to endothelium), nutritional deficiencies (Hypovitaminosis A) and anatomical considerations of skull and orbit (corneal melanosis in brachycephalic breeds) are the leading factors for development of endophthalmitis (or may be intraocular adhesions, if not treated) and ultimately vision loss occurs (Mahant and Mahajan, 2021).

Treatment should be done on the basis of drug penetration within the eye. Cornea and blood ocular barrier permits specific drugs to reach within the eye and therefore topical or systemic agent should be selected accordingly. Topical eye medications reach to anterior chamber through corneal penetration and for that try to choose agent which crosses cornea very well and much concentration of drug will be available in aqueous humour. However, bilateral ocular insults also demands systemic administration of drugs to relieve systemic problem also.

Healing of Diseased Cornea

Before approaching to corneal ulcers, understanding of corneal wound repair mechanism is necessary for beginners in the field of veterinary ophthalmology. Cornea is made-up of four different distinct layers in dogs and therefore all these layers will be repaired by different healing process. Uppermost layer i.e. corneal epithelium has excellent regenerative capacity. Divisions of epithelial basal cells from deep to superficial and transfer of fresh epithelial cells from limbus to centre of the cornea are the two different but simultaneous way of epithelial regeneration. The innermost single cell layer of cornea (endothelium) regenerates very rarely and if it gets damaged corneal transparency will be lost. Descemet's membrane is produced by endothelium. Understanding of stromal repair is very important and it has two different mechanisms. Compared to other components of cornea, stromal repair is very slow and responsible for loss of corneal transparency. Mechanisms for stroma healing includes avascular and vascular healing patterns. During avascular healing pattern the quiescent *keratocytes* (resting within stroma) will be activated and converted into fibroblasts. These fibroblasts are able to secrete stromal collagen. Whereas vascular healing pattern is characterised by extensive deposition of inflammatory cells as well as invasion of blood vessels which is called fibrovascular granulation in later stage. Ghost vessels are visible on slit-lamp examination in chronic cases.

Corneal Ulcers in Dogs

Corneal ulcer means exposure of stroma to external environment by acquired aetiology. Acquired aetiology may be infectious or non-infectious. Continuous irritation to cornea and tear film problems is considered as non-infectious cause (Pandey et al., 2018). Systemic diseases can also cause corneal damage. Simple ulcers will heal within a week and usually such ulcers are not deep. If simple ulcers are left untreated then may convert in deep ulcers followed by descemetocoele. Descemetocoele is left untreated then it results in iris prolapse which will further follow anterior synechia. Simple corneal ulcers can be treated with topical medications with or without parenteral drugs. If ulcer is complicated then surgical interventions must be followed as per indications (Slatter, 2003).

Type of corneal ulcers reported routinely

1. *Superficial ulcers* (Fig. 1) (*very thin layer of stroma is lost*)
2. *Indolent ulcers* (*superficial and usually non-healing*)
3. *Melting corneal defects* (Fig. 2) (*Enzymatic corneal degradation*)
4. *Deep corneal ulcers* (Fig. 3) (*partial stromal loss*)
5. *Descemetocoeles* (Fig. 4) (*complete stromal loss*)
6. *Perforated corneal defects* (*Traumatic ulcers*)

DIAGNOSIS OF CONREAL ULCERS

Fluorescein staining (FDT) is usually performed for confirmatory as well as differential diagnosis of corneal ulcers. Corneal stroma is stained by the *fluorescein dye* (Fig. 1b) and therefore helps in its diagnosis. Remember that descemet membrane will not take any stain and hence looks black or brown after FDT. When any deep punch (looks like deep ulcer) is negative after FDT and looks black or brownish with fluorescein stained periphery then usually case should be diagnosed as descemetocoele (Mezzadri et al., 2021). Descemetocoeles occurs after complete stromal loss (Fig. 4) and sometimes superficial staining on FDT shows liping of corneal epithelial tissue. Periphery of corneal ulcer is not sharp and shows “halo”. This liping of cornea is referred as indolent ulcer and such defects takes time to heal.

Superficial Ulcers are simple corneal ulcers (Fig.1 a & b) which may heal if treated properly with topical medications. Sufficient tear film is required to prevent corneal epithelium damage. Pathology of tear film formation such as due to Keratoconjunctivitis sicca will cause superficial ulcers.

Indolent ulcers are also superficial ulcers but such defects are non-healing ulcers and required surgical treatment as first choice. However infection is not present in theses defects (Singh et al., 2014). Generally it is advised that if any superficial ulcer is not giving response to topical medications then surgical interventions i.e. keratotomy should be done for healing. Sometimes microorganisms invade the corneal stroma and results in collagen lysis. These lytic corneal defects are known as *corneal malacia or Keratomalacia*. In theses defects cornea looks like melting cornea and therefore this defect is further referred as *melting corneal ulcers* (Fig. 2). Melting corneal ulcers are usually treated with EDTA, topical tetracycline, acetylcystein and autologous serum because these agents ultimately inhibit the lysis of collagen.

If stroma is lost upto 50% then term ulcer will come in category of *deep corneal ulcers* (Fig. 3). Note that there is still corneal stroma in deep ulcers and hence such defects will be stained after FDT. If complete stromal loss occurs with deep punch and will not stained after FDT then such corneal defects are referred as *descemetocoeles*

(Fig. 4). All the descemetocoeles are deep ulcers but all the deep ulcers cannot be the descemetocoeles. Both deep ulcer and descemetocoele requires surgical interventions for treatment (Mezzadri et al., 2021).

MANAGEMENT OF CORNEAL ULCERS

- a. *Medicinal trials*
- b. *Surgical management*

Medicinal trials

Usually superficial and simple corneal ulcers are managed by topical ocular drugs. Frequent administration of topical antibiotics directly over diseased cornea especially in initial days can recover the uncomplicated corneal ulcers. However complicated ulcers are basically subjected to surgical management. If ulcer is deep and complicated (associated with hair trichiasis and entropion) then use of indicated surgical technique along with topical drugs must be tried.

Table: 1 routinely used therapeutic protocol for uncomplicated (simple) corneal ulcers (Slatter, 2003)

Pharmacological Class of agent	Name of therapeutic agent for ocular use as eye drops	Recommended dose
Antibiotics	<i>Gatifloxacin (0.3% W/V)</i>	Frequent administration is required. For single eye, usually 2 drops every two hours up to 2 weeks is recommended.
NSAID's	<i>Flurbiprofen (0.03% W/V)</i>	One drop, once a day for maximum 4 days is usually recommended.
Lacrymostimulents	<i>Cyclosporine (0.1% W/V)</i>	One drop, twice a day till the healing of ulcer.
Lacrymomimetics	<i>Carboxymethylcellulose (0.5% W/V)</i>	One drop, every 4 hours till the healing of ulcer. If KCS is diagnosed along with ulcer then prolong use is recommended.

Surgical treatment of corneal ulcers

Temporary tarsorrhaphy, third eyelid flap, keratotomy and conjunctival flap technique are routinely used feasible surgical procedures for treatment of unresponsive corneal ulcers (Singh et al., 2014 and Singh et al., 2016). If superficial corneal defects are grossly visible and ulcer is confirmed with FDT then try to correct the diseases with

topical medications at least for a week with or without Temporary tarsorrhaphy. From surgical point of view regarding delayed or complicated case, keratotomy and conjunctival flap technique are associated with good results. However defects like indolent ulcers, deep ulcers and descemetoceles generally required surgical treatment and it should be done as soon as possible to prevent further eye damage

Grid keratotomy is first choice of surgical intervention for superficial non-healing ulcers (Fig. 5). It should be performed under general anaesthesia. Take a 24G needle and fix it at right angle within the artery forceps. Make straight lines (called as grid lines) covering the ulcers along with 1-2 mm healthy cornea (Singh et al., 2014). Use topical antibiotics upto a week and NSAID's for three days after surgery. Scar is commonly seen on cornea after grid keratotomy. Use topical steroids after FDT at least for 14 days to minimize the cornea scarring.

Conjunctival pedical flap technique is indicated for treatment of deep corneal ulcers (6 & 7), descemetoceles (complete stromal loss) and corneal perforated wounds. A flap is formed by incising the bulbar conjunctiva near the limbus and this flap is then suture with corneal defects. First holding suture should be done then followed by subsequent sutures. Both continuous and interrupted suture can be used. Temporary tarsorrhaphy must be done for two weeks. Use antibiotic (every two hours for 14 days) and NSAID (every 12 hours for three days) along with artificial tears (Table.1). Conjunctival flap having intact blood supply and therefore systemic administration of antibiotic is also beneficial in conjunctival flap technique because of direct active blood supply to corneal defect (Qureshi, 2020).

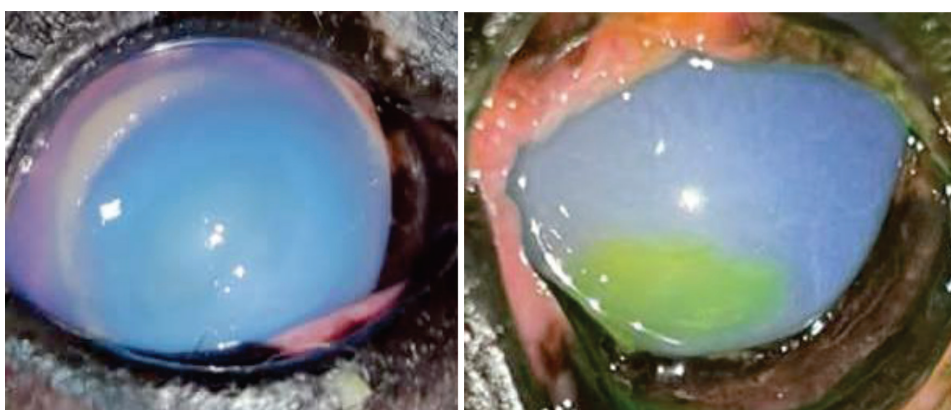


Fig. 1. Superficial corneal ulcer with diffuse corneal oedema (a), after FDT showing superficial corneal ulcer (b)

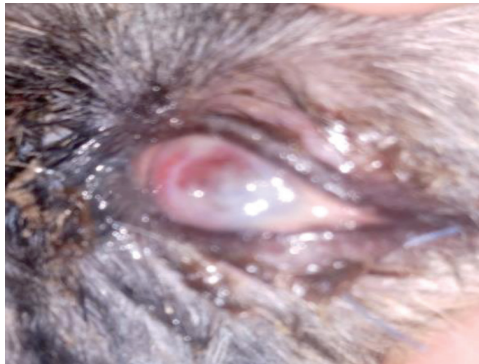


Fig. 2. Melting Ulcer

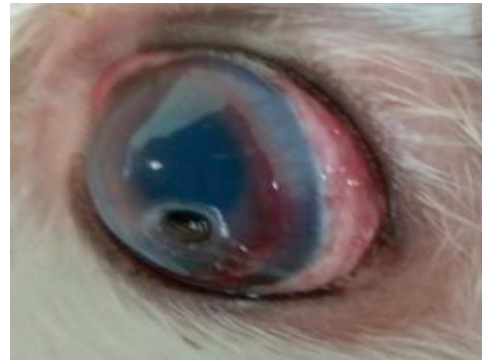


Fig. 3. Deep corneal ulcer with hedge pattern



Fig. 4. Descemetocel



Fig. 5. Grid keratotomy for non-healing superficial ulcer under general anesthesia



Fig. 6. Immediate postoperative photo showing use conjunctival flap for treatment of deep corneal ulcer



Fig. 7. Corneal repair after 1 month of Conjunctival flap application (treated for Deep corneal ulcer, another eye)

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MODIFIED ROBERT JONES BANDAGING FOR THE FORELIMB IN DOGS

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Abstract

The article explains the technique of modified Robert Jones bandaging for the forelimbs in dogs. The bandaging in dogs is required for wounds, first aid of fractures, as an adjunct to internal stabilisation of fractures and postoperative protection of the incision site. The bandage must provide optimum stability and a contamination free environment. The bandaging of proximal aspect of forelimbs is tricky and may be associated with complications like limb swelling, joint stiffness or joint laxity if not applied properly.

Keywords: Bandaging, casts, fractures, splints, wounds

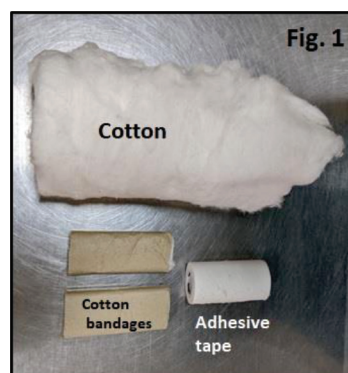
Modified Robert Jones Bandaging is used over conventional Robert Jones Bandaging (RJB) as a stabilizing bandage for fractures in dogs. Modified RJB uses only 1.5-2.0 cm thick pad of cotton, contrary to RJB which uses 4-8 cm thick cotton pad making it a lot less bulky and hence, preventing loosening or slipping of the bandage (Oakley, 1999). Cotton padding is followed by application of cotton bandages and adhesive tape. It immobilizes the limb, prevents further soft tissue damage due to sharp bony fragment and also reduces oedema. It is more suitable for fractures distal to elbow and stifle joints. But, these can also be applied on femur and humerus in an over the body fashion. Splints (PVC or aluminium) are usually incorporated.

Indications

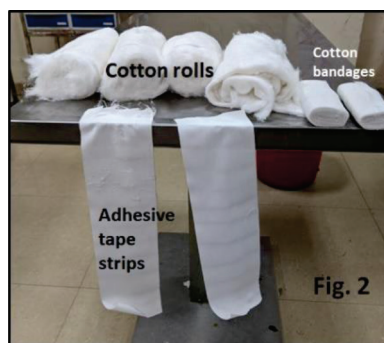
1. Humerus fracture
2. Elbow hygroma resection/drainage, tumour removal
3. Scapula fracture
4. Lacerations or growth removal in this region
5. Musculo-skeletal affection of Radius and ulna region in dogs with heavy muscles in distal humerus region and if bandage of radius and ulna is slipping down.

Materials required (Fig. 1)

1. Cotton (100gms for ≤ 8 kgs dogs, 200gms for ≤ 20 kgs dogs)
2. Cotton Bandages (2" for toy breeds, 4" for ≤ 10 Kgs dog and 6" for > 10 kgs dogs)
3. Adhesive Cloth tape (4 or 6 inches)
4. PVC (poly vinyl chloride) splint of length from just below elbow to the metacarpal region for the cases showing radial nerve deficit due to humerus fracture. The splint should be little wider than the limb in radius and ulna region and should be strong enough to not bend.

**Preparations (Fig. 2)**

1. Make 6/4/2 inch cotton rolls (5-6) as per the size of the dog and keep aside. The thickness of the roll is half to one inch depending on the size of the dog.
2. Take out appropriate sized bandages out of the wrapper (usually 2 required) as mentioned above. If 2" bandages are not available, the 4" can be cut into half with a scalpel blade.
3. Take out strips of adhesive tape (5-6 in numbers) with the open end of tape adhered to some slab or table. The length of the strip varies from 8" to 20" depending on the size of the dog. The length should be such that at least one roll is completed with one strip. For toy breeds, make longitudinal halves of the 4" strip if 2" tape is not available.

**Restraint of Dog for bandaging (Fig. 3)**

1. The dog is restrained in the lateral recumbency with the affected limb kept upwards and while standing, the affected side is oriented towards the doctor. The owner is allowed to stand on the other side.
2. The dog is muzzled and the owner is allowed to hold the head of the dog.
3. The doctor holds the unaffected fore and hind limbs of the dog which are on his/her far side and pulls them simultaneously on its side to make the dog lie down on the table.



4. The owner will keep the head pressed with one hand and can also hold the lower unaffected forelimb with the other hand (white star). The elbow of the owner is allowed to press on the dog scapula region at this point.
5. The affected forelimb, which faces upwards is kept free for bandaging at this point.
6. One person is required to hold both the hind limbs of the dog.
7. If required, the chest of the dog can be lightly pressed.
8. The third person will hold the paw of the affected limb with one hand and will keep it pulled but at the level of the body and in correct orientation to prevent mal-alignment of fracture fragments (Black arrow).

Technique of Bandaging



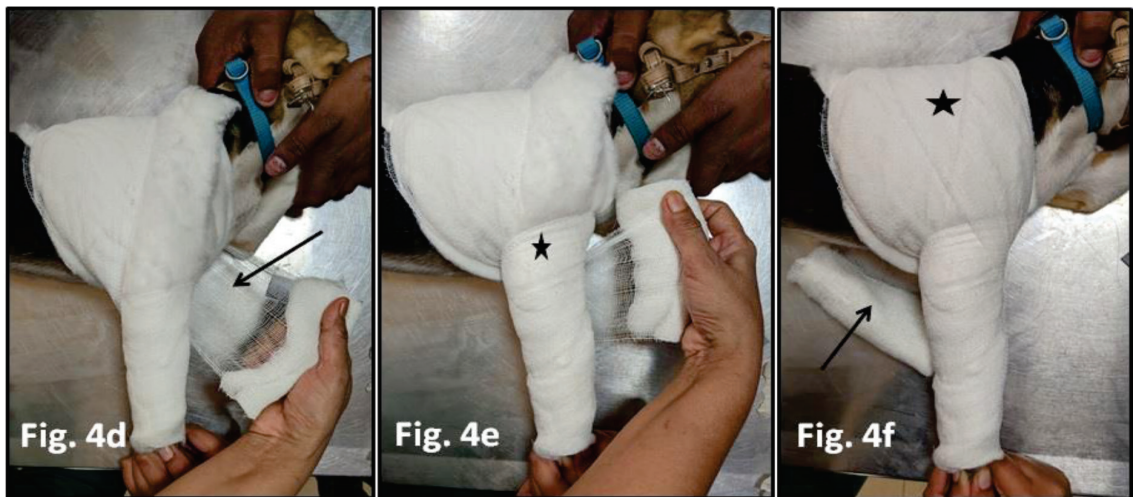
Fig. 4a. The pre-made cotton rolls are unrolled on the scapula, humerus, elbow and radius ulna region as per the requirement. The cotton rolls are applied in an oblique manner from medial to lateral side (black star).



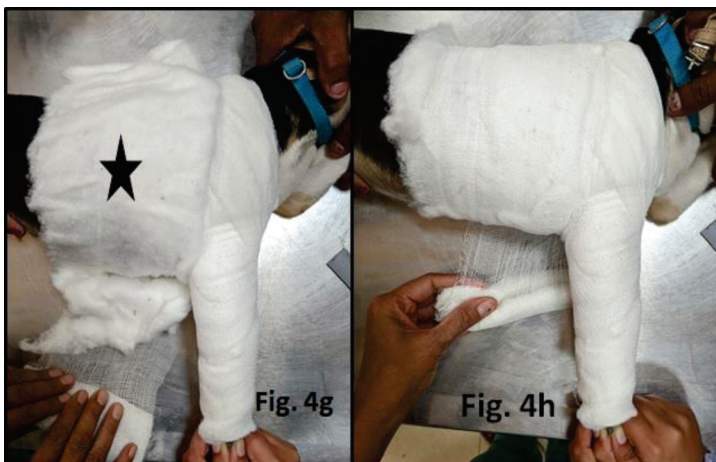
Fig. 4b: The unrolling of the cotton bandages is started from the lower most region (Black star), which is then advanced upwards.

Fig. 4c: While applying the bandage from below, at the elbow region, the bandage is taken over the shoulder (Black arrow) and to the other side of the body by slightly lifting the body of the dog.

The first layer of bandage can be applied lightly and is just to keep the cotton in place.



When the bandage comes on the front side (Black arrow, Fig. 4d), it is to be brought cranial to the elbow region and is rolled in elbow region or below for one or two times (black star, Fig. 4e) and is now again taken upwards over the shoulders from the cranial aspect (black star, Fig. 4f).



The bandage can also be wrapped on chest after cotton padding just caudal to the humerus and scapula region for a better support (black star, Fig. 4g). This process is repeated to achieve a clean and uniform look (Fig. 4h).

While wrapping the bandage, always apply pressure on the proximal aspect of the bandage and the distal should be left free, this will help in preventing the tourniquet effect due to bandaging.



Fig. 4i

If a PVC splint is to be applied for radius ulna (like in fracture of radius ulna as well or in humerus fracture with nerve deficit), it can be applied now on the caudal aspect of the radius and ulna (black arrow, Fig. 4i). The length of the splint is from the proximal metacarpals to the proximal radius region.



Fig. 4j

Fig. 4k

The last part of each bandage may be embedded below the cotton on the cranial or caudal aspect of the scapula region (Fig. 4j). Once the bandaging is complete, make the edges of cotton clean by rolling then underneath (Fig.4k).



Fig. 4l

The first tape strip is applied on the cranial and dorsal most aspect of the bandage involving half of the haired skin and half of the bandage part (Fig. 4l) and is brought on the caudal part of the limb distally.

**Fig. 4m****Fig. 4n****Fig. 4o**

Fig. 4m: The second strip is applied on the caudal and dorsal part of the bandage and may be rolled over the chest or underneath the limb and brought cranially.

Fig. 4n: The third strip is now applied in oblique manner as we did for the cotton rolls from dorsal to medial and again to dorsal on the other side.

Similarly all the strips are applied, till entire bandage is covered. The last strip on the distal most part must include partial skin and is to applied loose as there is no cotton in this part (Fig. 4o)

IMPORTANT INSTRUCTIONS FOR PET OWNERS

1. Keep bandage clean and dry.
2. The limbs can be covered with plastic coverings while going out for walks to prevent it from getting soiled. These coverings should be removed within half an hour to prevent excessive accumulation of moisture.
3. The bandaged limb should be observed for any swelling. In case there is swelling on the toes, owner is advised to press the toes frequently to reduce the swelling. Alternatively, an inch of bandage can be cut from most distal site to slightly loosen the bandage, which will eventually reduce the swelling. If swelling is not yet resolved, bandage has to be reapplied.
4. Evaluate the bandage for slippage. Observe patient for any discomfort or pain, indicated by excessive licking or chewing the bandage.
5. Thin skinned dogs like Greyhound are more prone to ischaemic necrosis of the limb due to tight bandaging and may be seen as change in colouration of the limb and other gangrenous changes (Tivers, 2010).

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CAMEL: JOURNEY FROM “SHIP OF DESERT” TO “MATTER OF RESEARCH”

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Abstract

The dromedary camel (Camelus dromedarius) has helped humankind for centuries in survival under adverse climatic conditions in different arid and semi-arid regions across the world. Besides, use in drought purposes and its milk and meat as food items, camel products and by-products are also being used as medicines in various human ailments by nomads rearing camels. Recent, research reports have not only validated some of their therapeutic potential, but has also opened new avenues for their potential use in biomedical research and development. The present article aims to provide a glimpse of unique properties and usage of camel products and by-products.

Keywords: Ailments, anticancerous, camel, milk, nano-bodies

Camel, named as “ship of desert” due to their adaptability in harsh climatic conditions, is a unique animal of family Camelidae, Kingdom Animalia. It first came into existence around 35 million years ago in North America. Human survival, specially residing in semi-arid or arid zones largely depends on them due to their wide acceptability as a mean of transportation, source of food and for protective purposes. Genetic basis of their unique adaptability in harsh climatic conditions have not been widely explored till date.

There are two major types in camelidae family:

1. Camelini: Include large camelids i.e. *Camelus bactrianus*, *Camelus dromedarius* and *Camelus ferus*
2. Lamini: Include small camelids i.e. Llama and Alpaca

Camelus dromedaries: Single humped camel present in hot and semi-arid regions like East Asia (Rajasthan, Gujarat in India) and in Northern part of Africa.

Camelus bactrianus: Double humped camel present in Mongolia, China and Cold desert of Asia i.e. Siberia etc.

***Camelus ferus*:** Wild Bactrian camel, critically endangered species found in parts of north-western China and in south-western Mongolia (Gobi desert areas).

According to Food and Agriculture Organization (FAO), estimated population of camel in the world was 27 million in year 2014, with annual increase @ 2.1 % (Ali et al., 2019). Recent census estimates world camel population close to 35 million (FAO, 2019), most of which are in Somalia, Sudan, Niger, Kenya etc. The total camel population in India is 2.5 lakhs as per 2019 livestock census which is 37.5% lower than that recorded in previous census. Thus, camel population in India is declining rapidly which an alarming situation for the sustained survival of this valuable germplasm of our country.

What makes camel unique?

Probably the physiological adaptation and enormous inimitable characteristics suited for survival in arid conditions makes this species a unique one. Few of them are listed below:

- Body temperature of camel may fluctuate from 34 to 41 degree Celsius within a day.
- The camel can digest dry matter and crude fibre better than other ruminants. Higher dry matter and fibre digestibility might be attributed to the longer retention time of large particle in its fore-stomach.
- Camel can survive long without food, because of persistent nitrogen availability through Urea Nitrogen recycling in gut and over-expression of H^+ -ATPase in their brain to provide a minimum required usable supply of energy (Warda et al., 2014).
- It can also survive for longer period without water as it can regulate loss of water through kidney by producing high concentrated urine. The reabsorbing capacity of water from intestine is also huge and thus produces dry faeces (Ouajd & Kamel, 2009).
- Camel has unusual elliptical shape RBC, which facilitate their flow in circulatory system especially in small blood capillaries. In addition, camel RBC can swell up to 240% of their original volume without bursting, due to altered distribution of membrane phospholipids in the Red Blood Corpuscles.
- Over-expression of alfa-actin in the heart results into adaptation for fluctuations in blood concentration, associated with alternative drought rehydration period (Ali et al., 2019).
- Presence of multiple copies of CYP2J genes provide camels, the ability to take large quantity of salt without developing hypertension (Wang et al., 2012).
- Presence of adipocyte “Vimentin” in camel hump is probably one of the reasons

behind their tolerance to high blood glucose. “Vimentin” might be an inducer of cellular trap for glucose and plays a significant role in their adaptation for survival in arid condition (Bazzi et al., 2013).

Camel genome and immunogenetics

The first draft of size of Bactrian camel genome was reported to be size of 2.38 Gb, containing 20,281 genes (Jirimutu et al., 2012) indicating camel has a smaller genome size than other mammals (Human: 3.2 Gb, Horse: 2.7 Gb, Cattle: 2.9 Gb).

Uniqueness about camelid antibody

The immune system of camel is unique. Besides conventional antibodies, camels possess unique antibodies that are smaller in size and known as heavy chain Antibodies (HC Abs), and are called nano-bodies. These are smaller in size (15 KDa) and have high stability, binding abilities and enhanced potency (Tillib et al., 2014).

Future insights about camelid Nano-bodies

- The small size makes camelid nano-bodies amazingly effective in the field of oncology, neurodegenerative disease, type-II Diabetes mellitus, prostate cancer, atherosclerosis and colon cancer. They can also be used as antiviral agent, snake/insect antiserum, anti-tumor agent etc.
- Camelid nano-bodies are highly suitable for oral immunotherapy due to their resistance to extreme of pH and their ability to bind to the target at higher concentration of chaotropic agent.

Camel milk

- Camel milk is known for human health benefits since last 5,000 years. Nomadic people use camel milk for the treatment of many ailments, but in the last 20 years, many studies further validated therapeutic use of camel milk in human ailments like diabetes (Type 1), autism, tuberculosis, food-allergy, GIT disorders, cancer etc.
- Basically, the composition of camel milk is very similar to the cow milk (in terms of protein and lactose component), but a special feature lies in its fat, that does not form a layer, rather evenly distributed throughout the milk as small micelles, making it easily digestible.
- Camel milk contains 86-91% Water, 1.9-2.2% Fat, 2.8-3.6% Protein, 2.8-4.2% Lactose, 11.4 Meq/L Sodium, 80 mg% Calcium. (Yagil et al., 1994).
- It also contains high concentration of long chain fatty acids, possessing distinguished property to inhibit the growth of microorganisms. The protective proteins and enzymes e.g. lactoferrin, lactoperoxidase and peptidoglycan proteins contribute special anti-bacterial and anti-viral properties in its milk. Generous

amount of insulin is also available, thus beneficial as anti-diabetic agent.

- Camel lactoferrin has special role in treating Hepatitis C virus. Camel lactoferrins have more antiviral activity in early stage of infections like Herpes simplex virus 1 and 2 (HSV1 and HSV2) and also against HCV (El-Fakharany et al., 2013).

Camel urine

Camel urine is being used for medicinal purposes in early folklore since centuries. It is believed that Prophet Mohamed advised its use in the treatment of a wide range of diseases (Alhaider et al., 2011). It has several unique activities; some of them are enlisted below:

- **Anti-cancerous activity:** Camel urine has anticancer properties via the considerable repression of the expression of the gene encoding carcinogen activating enzyme CYP1a1 at the m-RNA level in neoplasm of liver cells (Alhaider et al., 2011). The possible anti-malignant action of camel urine is attributed to the presence of lactoferrin and canavarine in it (Ahamad et al., 2017)
- **Antimicrobial activity:** Antimicrobial activity of camel urine is due to its high salt concentration and alkalinity along with natural bio-active compounds, probably found due to xerophytes consumed by them.
- **Cardiovascular activity:** - Camel urine shows anti-platelet aggregatory activity like aspirin and clopidogrel by repression of prostaglandin synthesis pathway and ADP mediated pathway. Benzo-propanoic acid, a fatty acid derivative may be responsible ingredient for its anti-platelet aggregatory properties (Ahamad et al., 2017)
- **Antioxidant:** Camel urine possess anti-oxidative property (potent scavenger of peroxy radicals, hydroxyl radicals and singlet oxygen) (Ames et al., 1981) and it can chelate metal iron by converting them to the least reactive form.

Camel Meat

- Camel meat is a valuable source of food, rich in many essential amino acids, minerals (e.g. iron, zinc and selenium) and vitamins (Vitamin B, E). Bio-active compounds (e.g. Q10, carnosine, anserine, glutathione etc) and some essential fatty acids like omega-3 are also found in sufficient quantity in its meat.
- Kadim et al., (2008) indicated that camel meat has traditionally been used for treatment of fever, sciatica, shoulder paid, corneal opacity, hemorrhoidal pains, asthma, gastrointestinal parasites etc.

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MICROALGAE PROTEIN AS AN ALTERNATE FEED INGREDIENT IN DOG FOOD

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Abstract

Microalgae, small-sized algae has an excellent nutritional composition proteins, carbohydrates, lipids, vitamins, minerals and enriched bioactive compounds like carotenoids and others. Comparison to other vegetarian protein it has more desirable amino and linoleic acid - polyunsaturated fatty acids (LA-PUFA) profile. Use of microalgae protein as an alternate vegetarian protein can be beneficial to boost health status, immunity, disease resistance especially in diseased and sensitive, and elderly companion dogs. Numerous studies have been undertaken on micro algae, omega 3 PUFA (DHA or EPA) enrichment in poultry, pig, sheep, goats and ruminants diet and to improve milk, meat or egg quality. Micro algae protein as an alternate protein can assure quality protein supply with maintenance and development of sustainable agriculture and livestock farming, in an environmental friendly way.

Keyword: *Feed, micro algae, pet foods, protein*

Chicken, beef, lamb, salmon and even turkey and legumes, pulses and soybeans are non-vegetarian and vegetarian source of conventional protein in pet foods. These are food of human choice too thus impart food-feed competition. Furthermore, over demand of conventional protein source will increase the feeding cost, may affect expansion of livestock business. These facts extend need to identify alternative protein sources (e.g. insect meals, food waste, agro-industrial byproducts) and other newer proteins. Referring to alternated protein; yeasts, fungi, bacteria and micro-algae, the name Single Cell Protein (SCP) were subjected through fermentation technologies for the protein biomass production at commercial level. There are distinct groups of microalgae, being the eukaryotic diatoms (*Bacillariophyceae*), green algae (*Chlorophyceae*) and golden algae (*Chrysophyceae*) the most abundant in nature. More than 100,000 species have been estimated with a large share of diatoms. Typically, autotrophic microalgae are used for CO₂ fixation and to produce organic compounds (biomass) with the sunlight energy and augments oil in a very productive way (Christi, 2007). While, heterotrophic microalgae utilizes organic compound as a source of energy instead of sunlight. Some of them have been successfully cultivated for their specialized nutraceutical values, rich in vitamin A, C and E and Beta-carotene (Pirwitz et al., 2016), astaxanthin (Gouveia et al.,

2010). Long-chain n-3 PUFA, docosahexaenoic (DHA, 22:6, n-3) content in *Isochrysis galbana* is about twice to fish oil. Studies have suggested that autotrophic microalgae can modulate milk meat or egg nutritional profile on supplementation in animal diet. Similarly, microalgae are increasingly being used in pet food to improve palatability and meet the requirements for natural ingredients in pet food formulations. A microalga also has a longer shelf life and is available in powder and liquid forms, making it simple to compounding. The nutritional properties of *spirulina* microalgae are propelling the global market for pet food. According to a report (<https://www.futuremarketinsights.com>) only *Spirulina* was expected to account for more than 64% of the market in 2021.

Chemical composition of micro-algae

Chemical composition of different algae have studied in many previous studies and general overview on the major constituents, selected data of various micro-algal species are presented in Table 1 (Yaakob et al., 2014). Microalgae composed of proteins, carbohydrates, lipids, vitamins, minerals, and bioactive substances like carotenoids and other macro and micro nutrients. Microalgae biomass contains all essential nutrients to replace artificial supplement with natural sources. Specifically, one of the primary criteria favoring their use in feed production is the high protein content of several microalgae species (Kovac et al., 2013). The majority of microalgae protein fractions have an average quality that is comparable to or even superior to that of traditional plant protein fractions (Becker, 2004).

In terms of quality, dry *chlorella* is comparable to yeast, soy flour, and milk protein, with a protein concentration of between 50 to 60 % (Kovac et al., 2013) and *Arthrospira* microalgae about 60 to 70 % on dry weight with all the essential amino acids (Gutiérrez-Salmeán et al., 2015) but low in methionine and cysteine content. The amino acid profile of selected microalgae have been compared under studies with recommended balanced protein source (FAO/WHO, 1973), it has been found that the amino acid profile of algal protein was below to egg white protein but better than the any vegetarian protein, including soybean protein (Morales de León et al., 2005).

Carbohydrates are also important nutrients in microalgae. Indeed, microalgae cell wall typically comprise of hemicellulose (Gutiérrez-Salmeán et al., 2015), which may be beneficial to animals' gut. In the case of *Arthrospira*, there is an efficient digestion of its carbohydrate fraction by ruminants when used in levels up to 20% of total feed intake, compared to other algal feed types, like *Chlorella* or *Scenedesmus obliquus* (Gouveia et al., 2010). Microalgae are a rich in minerals and vitamins (Christaki et al., 2011), specially, Green algae (such as *Chlorella*) are good source of cobalamin (vitamin B12). *Aurantiochytrium* are used as an excellent source of carotenoids to astaxanthin (Aki et al., 2003).

Table 1 Chemical composition of common Micro algae (% DM) used in animal feed

Algae species	Protein	Carbohydrates	Lipid
<i>Anabaena cylindrical</i>	43-56	25-30	4 to 7
<i>Aphanizomenonflos-aquae</i>	62	23	3
<i>Chlamydomonasrheinhardii</i>	48	17	21
<i>Chlorella pyrenoidosa</i>	57	26	2
<i>Chlorella vulgaris</i>	51-58	12 to 17	14-22
<i>Dunaliellasalina</i>	57	32	6
<i>Euglena gracilis</i>	39-61	14-18	14-20
<i>Porphyridiumcruentum</i>	28-39	40-57	9-14
<i>Scenedesmusobliquus</i>	50-56	10-17	12-14
<i>Spirogyra sp.</i>	06-20	33-64	11-21
<i>Arthrospira maxima</i>	60-71	13-16	06-07
<i>Spirulinaplatensis</i>	46-63	08-14	04-09
<i>Synechococcus sp.</i>	63	15	11

Applications of microalgae in feed

Animal feed industry consumes more than 30% of global microalgae production (Becker, 2004) particularly, *Schizochytrium* sp., *Chlorella* sp., *Arthrospira* sp., *Isochrysis* sp. and *Porphyridium* sp. *Arthrospira*. Studies have shown that microalgae inclusion in animal diets can improve growth and meat quality in ruminants, pigs, poultry, and rabbits. However, results are highly dependent on the composition of microalgae and their amount in the diet. *Schizochytrium* sp. improves fatty acid composition in pork and poultry meat, owing primarily to its high docosahexaenoic acid content (DHA).

Vegetable-protein sources which contain rich α -linolenic acid (C18:3n-3) as a precursor for the n-3 LC-PUFA, DHA and EPA source are limited in numbers, like linseed and sunflower oil seed. Microalgae such as the genus *Schizochytrium* have been used as potential source of omega 3-PUFA and high DHA concentration (approximately 20%) sources used in dog food. Other species of *Phaeodactylum tricornutum* and *Nannochloropsis* sp. (autotrophic) rich in EPA 39% of total FA (Adarme-Veja et al., 2012) used in animals feeds. Additionally, they also carries anti-inflammatory properties, alter skeletal muscle function, improve reproductive status of animals, skin and appendage health. Souza (2018) reported that DHA-rich algae at 0.4% inclusion level in dry pet food resulted in higher protein digestibility and palatability in dogs. Further, the *Spirulina platensis* as whole dried powder is beneficial when used at 10.7% of pet food on DM basis (Zhang et al., 2001).

Menezes Souza et al. (2019) reported palatability, growth performance, health status, oxidative stability of dog food improved on micro algae supplementation. DHA- PUFA enriched composition of microalgae can improve growth performance, supports the immune functions, inflammatory modulators eicosanoids (prostaglandins, leukotrienes, thromboxanes, and prostacyclins) with less inflammatory action, resolvins, and protectins, which reduce inflammation (Calder, 2012) admires disease resistance, antiviral and antibacterial action, gut function and probiotic colonization stimulation. DHA directly regulates inflammatory function through gut associated immune organs. Higher concentration of beta-carotene, vitamin E, sterols, phenolics and flavonoids imparts oxidative stability, increase nutrient digestibility and higher metabolisable energy intake. However, studies have reported variable response of microalgae cell wall composition on growth performance in monogastric and ruminants, microalgae cell walls contain cellulosic material can be differentially fed to ruminants, while it is necessary to process algal as biomass before appropriate in monogastrics animals diet (Becker, 2004). Furthermore, the use of carbohydrate-active enzymes with micro algal protein will significantly improve the efficiency of nutrient utilization in monogastric animals.

In conclusions, microalgae can be used as alternate feed ingredients staple food crops such as corn and soybean, mitigating current competition with the human food chain and contributing to agricultural sustainability. Inclusion of microalgae in pet food can be an option for the pet owner seeking palatable vegetarian diet enriched n-3 LCPUFA alternative to fish oil of best oxidative stability. Nutritional composition of microalgae can improve growth performance, health status, immune function and feed quality supplements of will in pet food will support.

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SIGNIFICANCE OF DIETARY DCAD MANAGEMENT IN TRANSITION COWS

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Abstract

The transition period is the most vulnerable period that is associated with incidences of metabolic diseases like milk fever, ketosis, and acidosis which may result in a detrimental effect on cow's performance. DCAD is an important tool that can be utilised to prevent these metabolic diseases and improve health. Negative DCAD (–100 to –150 mEq/kg) during prepartum induces acidic intestinal pH for calcium absorption and facilitates recruitment of bone reserves, increases the excretion of calcium in the urine and consequently activates calcium reabsorption and increases production and/or receptor receptiveness to $1,25(\text{OH})_2\text{D}_3$ and parathyroid hormone (PTH). Post-partum positive DCAD (+200 to +400 mEq/kg of diet) improves DMI and milk production by eliminating the physiologic acidosis by raising buffering capacity of the blood, reflected by increased blood pH and blood bicarbonate (HCO_3^-) concentration, hence DCAD has a significant positive effect on the productive performance via changing the rumen environment and improving acid-base homeostasis in the dairy cows.

Keywords: Acid-Base homeostasis, energy balance, metabolic diseases.

Over the last three decades, genetic improvement of dairy cows has markedly increased milk yield that also increased the incidences of metabolic disorders like milk fever and acidosis. The transition period (extending from three weeks before and after calving) is particularly vulnerable due to the cow's metabolism shifts from pregnant non-lactating to non-pregnant lactating animals, which results in increased demand for energy protein and other minerals like Ca, P, Zn. etc. Dairy cow experience about a one-third decrease in feed intake during the last three weeks before calving, with a significant reduction observed in the final week before parturition (Hayirli et al., 2002). This is mainly due to an increase in the concentration of circulating estrogens and less capacity for the rumen to expand because of increased foetus size. The resultant decreased dry matter intake (DMI) and the inability of the cow to cope with the increasing energy demands of lactation during the first phase of lactation lead to negative energy balance (NEB).

During early lactation, the drive to produce milk is given priority over nearly all other physiological processes leading to the drain of nutrients via milk, especially

calcium resulting in milk fever or parturient paresis. Which can further complicate the already complex periparturient metabolism and reduce milk yield and as a severe consequences may produce downer syndrome and death, if not treated (Constable et al., 2016). Negative energy balance combined with reduced calcium available for cellular activity can predispose cattle to other metabolic and infectious disorders. A causal relationship has been drawn between subclinical hypocalcaemia and ketosis, abomasal displacement, dystocia, uterine prolapse, mastitis, udder oedema and decreased immune function, leading to infections such as mastitis and metritis (Goff and Horst, 1997), in addition to impaired neuroendocrine and metabolic systems, impaired immune cell function, immunosuppression, the decline in plasma zinc concentration (an essential cofactor for many metabolic, signal transduction and transcription factors) and even neonatal hyperthyroidism and hypo-insulinemia (Carafoli, 1987).

Parturient paresis (Milk Fever) and Its Etiology

The prevalence of hypocalcaemia is as high as 70% for multiparous cows, although only 8% exhibited clinical hypocalcaemia which lowers the 16% yearly milk yield. Milk fever is characterized by and the result of severe hypocalcaemia. In these cows the calcium homeostatic mechanisms, which normally maintain blood calcium concentration between 9 and 10 mg/dl, fail and the lactational drain of calcium causes blood calcium concentration to fall below 5 mg/dl. This hypocalcaemia impairs muscle and nerve function to such a degree that the cow is unable to rise.

The adaptation to the onset of lactation during the critical first days of lactation is accomplished by release of parathyroid hormone (PTH), which reduces urinary calcium losses, stimulates bone calcium resorption, and increases 1,25-dihydroxyvitamin D synthesis to enhance active intestinal transport of calcium. All three must be operational if hypocalcaemia is to be minimized.

Cows fed diets that are relatively high in potassium or sodium are in a relative state of metabolic alkalosis, which increases the likelihood that they will not successfully adapt to the calcium demands of lactation and will develop milk fever. Evidence suggests that metabolic alkalosis induces conformational changes in the PTH receptor, which prevents tight binding of PTH to its receptor. The parathyroid glands recognize the onset of hypocalcaemia and secrete adequate PTH. However, the tissues respond poorly to the PTH, leading to inadequate osteoclastic bone resorption and renal 1,25-dihydroxyvitamin D production (Goff et al., 1991). Full recovery from milk fever occurs only after the cow has responded to the PTH by producing 1,25-dihydroxyvitamin D.

DCAD and its role in preventing Parturient paresis:

DCAD stands for Dietary Cation Anion Difference which represents the acid-base status of the feed given to the animal. Sanchez & Beede (1991) coined the term dietary

cation-anion difference (DCAD) which is a way to balance the electrical charge of the cations and anions in the diet. These cations and anions drive the pH of the medium. Adjustment of such ions in the ration in such a way that the dietary cation–anion difference (DCAD) favours the amounts of anions (Cl and S) can activate calcium metabolism pathways. With higher levels of strong anions, DCAD will tend to be more negative, whereas higher levels of strong cations will deliver more positive DCAD values.

A negative DCAD has been shown to affect calcium metabolism. It can induce a more acidic intestinal pH for calcium absorption and facilitate recruitment of bone reserves, increase the excretion of calcium in the urine and consequently activate calcium reabsorption, increase production and/or receptor receptiveness to $1,25(\text{OH})_2\text{D}_3$ and parathyroid hormone (PTH) (Goff et al., 1991). Another proposed mechanism is increased release of calcium from endoplasmic reticulum reserves in response to cellular acidosis.

How the DCAD is calculated:

DCAD is calculated in Milliequivalents (mEq) and Milliequivalents are calculated by multiplying the content of each element in the diet by a conversion factor. The common factors for Sodium, Potassium, Chloride and Sulphur are 435, 256, 282, 624, respectively.

For example, a diet containing 0.15% sodium, 1.1% potassium, 0.2% chloride and 0.2% sulphur, the milliequivalents would be:

Element	% In diet(A)	Conversion factor(B)	mEq/kg(A*B)
Sodium	0.15	435	65.25
Potassium	1.10	256	281.6
Chlorine	0.20	282	56.4
Sulphur	0.20	624	124.8

DCAD equation:

The equation for calculating the DCAD for a diet (or ingredient) is:

$(\text{sodium} + \text{potassium}) - (\text{chloride} + \text{sulphur}) = \text{DCAD in mEq/kg}$

From the above example, the result is:

$(65.25 + 281.6) - (56.4 + 124.8) = \text{mEq/kg}$

$(346.85) - (181.2) = +165.65 \text{ mEq/kg}$

The DCAD equation and conversion to milliequivalents can be combined as follows into one step:

$[(\text{sodium} \times 435) + (\text{potassium} \times 256)] - [(\text{chloride} \times 282) + (\text{sulphur} \times 624)] = \text{mEq/kg}$

Impact of DCAD and its potential role in improving health, production and reproduction:

i) DCAD for prepartum cows

The primary goal in late pregnancy is to provide a ration with a negative DCAD to reduce the risk of hypocalcaemia and clinical milk fever around calving. Simply using feedstuffs with lower concentrations of K and Na will lower DCAD enough to improve transition cow performance. This may also reduce udder oedema. When it is not possible to reduce dietary K and Na enough, supplementation with anions (chloride and sulphate) will reduce DCAD.

A target DCAD of recommended negative values -100 to -150 mEq/kg may improve transition cow health and performance. Anion sources to reduce DCAD include the so-called anionic salts such as ammonium chloride, calcium chloride, magnesium chloride, ammonium sulphate, calcium sulphate, and magnesium sulphate.

ii) DCAD for lactating cows

The potential effect of positive DCAD on lactating dairy cows has been explored, and results indicate that DCAD and production are related possibly through acid-base regulation (Hu & Murphy, 2004). As to meet the nutritional demand for high milk production, large proportion of concentrates are given to the animal that predisposes that animal to acidosis or sub-acute ruminal acidosis (SARA). A high DCAD diet neutralises the extra acid produced during rumen fermentation of the concentrates leading to an optimal rumen environment resulting in better fermentation and utilisation of nutrients which results in increased DMI and milk production (Iwaniuk & Erdman, 2015). The DCAD may be increased by reducing anions or by supplementing with sodium bicarbonate or potassium carbonate.

Currently, the NRC (2001) recommends dietary concentrations of 0.22% Na, 1.06% K, and 0.28% Cl for Holstein cows averaging 90 days-in-milk (DIM) producing 45kg of milk that contains 3 % milk fat and 3.0% protein. Based on these recommended requirements of Na, K, and Cl in the diet, the suggested minimal DCAD concentration ($\text{Na} + \text{K} - \text{Cl}$) for dairy cows would be 295 mEq/kg of diet dry matter (DM) (NRC, 2001). However, a more recent meta-analysis of published research (Hu & Murphy, 2004) suggested that a higher DCAD concentration (400 to 450 mEq/kg) might result in improved milk yield and milk fat production.

So generally, in lactating cow's rations, DCAD of value ranging between +200 to +400 mEq/kg of diet are being fed.

iii) Role of Calcium, DCAD on health and reproduction

One of the functions of calcium is to allow muscle contraction. Clearly, a reduction

in muscle contractility will lead to a decrease in dry matter intake (DMI) as rumen function decreases, leading to severe Negative energy balance (NEB). As consequence, there is an increase in the fat mobilization that may result in fatty liver syndrome and ketosis. An excess of ketone bodies can further suppress appetite (Boland et al., 2001), it has been shown that plasma calcium concentration of 5mg/ml reduces abomasal motility by 70% and the strength of the contraction by 50%. Low calcium concentrations also prevent insulin production, further exacerbating this situation. Ultimately, milk yield will be reduced and fertility will suffer. Muscle tone in the uterus will also be adversely affected with cows experiencing prolonged calving and retained placenta. Uterine involution may also be impaired giving rise to fertility problems. As DCAD maintains the calcium levels in the blood thus helping in maintaining the health and reproduction status of the cow.

Monitoring DCAD

Monitor cow urine when using anionic products. Urine pH is a reasonable indicator of metabolic pH status and reflects the effectiveness of anionic products. The urine pH of dairy cows fed typical rations without anion supplementation is between 7.8 and 8.2, a normal value for ruminants. But with anionic supplementation (–100 to –150 mEq/kg) urine pH should be 6.0 to 6.5. Another way is to calculate the dietary DCAD value.

DCAD products available in the market

NutriCAB™ Dry, DCAD Minus Anionic Mineral Supplement

Feeding recommendations

- Cow & buffalo: 100-200 gm/ animal daily for 3 weeks before parturition.
- Anionic mixture consisting of calcium chloride 33.4%, magnesium chloride 33.3%, sodium chloride 18.3%, magnesium sulphate 8.3%, and calcium hydrogen phosphate 6.7% can be given to the cow @90g/d for three weeks prepartum (ICAR, 2013).

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DOES CLIMATE CHANGE AFFECT REPRODUCTION IN BOVINES?

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Abstract

Different environmental variables like temperature, humidity, wind speed, radiations directly influence the reproductive and productive performance of livestock. Among these, heat stress is the most important factor affecting livestock. Climate change is not only responsible for decrease fertility but also decrease milk production in dairy animals. Farmers should be made aware about climate change triggered poor reproductive performance of dairy animals which can be maintained by adopting measures to reduce environmental stress, prevent fertility losses and other health consequences in animals.

Keywords: Buffalo, environment, fertility, health, stress

During last decade, various changes have been observed in the climatic conditions in the country due to which the reproduction and production efficiency of dairy animals has been adversely affected. These effects are expected to further increase in future due to increase in global warming. Globally, the temperature is rising day by day. Climate change has led to extreme weather incidents like high rainfall and/or extreme heat wave along with continuous increase in the reoccurrence of these incidents. The negative influence of heat stress on cattle and buffaloes can be quantified through formulating temperature humidity index (THI). In this index, environmental temperature and humidity are taken into consideration. If THI is below 72, the animals feel comfortable between 72 and 79, the animals experience mild stress, between 80-89, moderate stress and above 90, the animals experience severe heat stress (Ouellet et al., 2021). An increase in temperature humidity index (THI) worsens the environmental conditions which are stressful for the dairy animals, causing decreased production ability of dairy animals and subsequently reduced profitability from dairy farming. The current manuscript is an overview of how climate change affects reproduction in dairy animals.

What is heat stress?

Restlessness and discomfort which are due to increase in temperature are indicative of heat stress. Normally, animals with the help of their biological processes keep their

body temperature within normal range. But, gradually temperature starts rising along-with humidity during summer season followed by rains. During this time it becomes difficult for the animals to control the thermoregulation of their bodies and they start feeling stressed due to heat. Such symptoms are also associated with the decrease in the feed intake. Exotic and crossbred cattle are capable of controlling body temperature at 24-26°C and 33°C, respectively. However, buffaloes can manage their body temperature very well upto 36°C (Upadhyay & Singh, 2009). But, when environmental temperature rises beyond its higher sensitive temperature also at that time body temperature of an animal also starts rising and this is the point when dairy animals start feeling heat stress.

Susceptible animals

Indigenous cattle are capable to tolerate heat stress as compare to exotic and cross-bred cows. They can survive well at high temperatures as compare to exotic/crossbred cattle. It indicates that exotic/cross-bred cattle get badly affected by rise in the temperature. Therefore, special management is required to protect them from heat stress.

Buffaloes compared to cattle feel more stressed during summer season. There are basically two reasons; Firstly due to their body colour since black colour absorbs the maximum heat and secondly that buffaloes have only 1/6th of the sweat glands as compared to cattle. Therefore, due to fewer sweat glands, buffaloes sweat less and have smaller range to control their body temperature and are exposed to higher threat of heat stress.

Cardinal signs of heat stress in dairy animals

- During summer stress pulse rate and respiration rate of animals increases.
- Laboured breathing and dehydration is observed.
- Increase in heart rate too is observed in dairy animals.
- Increased salivation is also observed in stressed animals.
- Due to heat stress some weak animals become unconscious.
- Skin of the stressed animals become wrinkled and rough.
- Body temperature of animals' increases to 106-108°F.

Tips to avoid heat stress

Following points can be adopted to keep dairy animals safe from the heat stress:

- Try to keep animals as much as possible in the shades during summer season. Animals should be kept in sheds or under trees during day time.
- Roof of sheds should be minimum at the height of 9 feet.

- If possible, roof of shed should be covered with rice husk. Moreover, concrete roofs should be painted white from outside to prevent the hazardous effects of heat.
- Hang curtains on the windows, so that animals can be protected from heat waves. Further, sprinkle water on curtains so that while passing through them heat waves turn into cool breeze.
- Sheds should be equipped with ceiling and exhaust fan. These fans help in keeping the temperature of sheds lower and keep the air circulating inside the shed.
- It is recommended that in bigger sheds showers should be fitted to regulate the temperature of sheds. But make sure that sheds would have ceiling and exhaust fans.
- The showers should run at the rate of 5 minutes per 20-30 minutes time interval.
- Buffaloes should be bathed in ponds during day time to lessen the effect of heat stress.
- Animals should always be provided with *ad lib* clean and fresh water during summer season.
- Animals should be fed during early morning or late evenings when temperature is low.
- Animals should not be grazed during day time in direct sun light.
- Animals should be fed with less fibrous and easily digestible fodder.
- Animals should also be fed with mineral mixture.

Effect of climate change on estrus detection

The decrease in estrus detection efficiency and accuracy is the major reason behind the delay in getting the animal inseminated in time. About 60% buffalo display estrus between 6 pm and 6 am. Others reasons behind poor estrus detection are limited exhibition of signs of estrus, deficiency of male animal rearing and non-display of estrus signs by 80% buffalo during heat period (Madan & Prakash, 2007). Therefore, there is a need to get acquaintance of dairy farmers for estrus synchronization to reduce the problems of estrus detection.

Reasons for unsuccessful heat detection

- Giving less importance to identification of estrus.
- Suckling of dam by calves for longer duration.
- Giving less time for observation of animals.
- Environmental stress decreases the efficiency of animals in displaying estrus.

- Lack of knowledge about secondary signs of estrus.
- Increased milk production by animal decreases its ability to exhibit estrus.
- Some animals have genetically short time for estrus display.
- Ignorance in observation of animals during night and early morning when they show most heat signs.

Tips for induction of estrus in animals

- Induction of estrus in heifers depends on weight rather than age. A heifer should have 250-300 kg bodyweight at the time of first estrus.
- Efforts can be made to protect the animals from seasonal effects viz. making animals bathe in a pond, use of sprinklers and tying under tree in summer. During monsoon, when humidity is high, fans can be used to provide cooling effect to animals. In winters, keeping animals in shed covered with sack help protection against cold.
- Animals should be dewormed and vaccinated regularly. Optimum nutrition plays a pivotal role in enhancing reproductive efficiency in farm animals. Therefore, specific feeding strategies need to be formulated to exploit round the year potential. Regular supplementation of green fodder along with 2-4 kg easily digestible mineral mixture should be provided to animals before parturition.
- The presence of a bull accelerates physiological processes in primiparous / pleuriparous animals that initiate resumption of estrous cycles and shortens postpartum anestrus interval. Keeping sterile teaser bulls with cows during early postpartum period could be a useful tool in managing postpartum anestrus in dairy animals.
- Animals which do not show behavioral signs of estrus should be inseminated artificially because breeding activity in such animals is normal.
- Proper and adequate record of each animal of herd should be kept viz. date of parturition, date of first estrus, symptoms of estrus etc. At any time, if the proportion of animals that fail to come in estrus within 90 days following parturition is more than 10 percent, in such conditions thorough examination of the herd should be get done by a qualified veterinary doctor.
- If still the animals do not come in estrus, they should be subjected to ultrasonography and advised for breeding protocol accordingly.

Effect of climate change on conception rates and economic losses

The primary reason is delay in conception of heifers as well as an increase in inter-calving interval. More the number of days an animal remains non-pregnant beyond

prescribed waiting period, lesser will be the production of milk owing to which the owners will not be able to compensate the profits. During summer stress, the conception rates following AI in dairy animals reduced significantly (Amundson et al., 2006). The average conception rate following AI is around 40%. This remains 27% during summer and 54% during winter. Thus, rising temperature has deleterious effect on conception following AI (Wolfenson et al., 2000). The delay in getting the animal pregnant causes approximate loss of Rs 150-250 per day per animal.

To reduce the economic losses due to an increase in inter-calving interval by impending climate change, the use of estrus synchronization is an option for these animals. Estrus synchronization means the use of hormones on some fixed days to bring animals into estrus artificially and then inseminate the animals at the predetermined fixed time. With the use of this technique, there is no need to identify an individual animal in estrus. There are other benefits also of estrus synchronization like reduced labor cost, improved breeding efficiency and improved conception rate by fixed time insemination. This technique can be used in dairy animals about 60 days after calving; hence the service period is reduced to 60-90 days. The planning of time of estrus synchronization can be done in a way that the animals calve when there is less milk production and there is more demand for milk production. The calf crop can be managed together for their better health through better management practices.

Effect of adverse climatic conditions on the incidence of repeat breeding rates in dairy animals

According to various studies, it had been reported that the ideal temperature for milk yielding cows is between 5-25°C. A sudden change in the temperature (either increase or decrease) adversely affects animal's physical activity, growth, production and reproduction. The well-known fact is higher the reproduction rate of an animal more will be the milk production at farm. The animals with low reproductive ability cannot conceive and they become repeat breeders. Repeat breeder animals have regular estrous cycle but fail to conceive either by artificial insemination (AI) or natural service with fertile semen for more than three times. There can be huge economic losses to dairy owner or farmers under these conditions of animals for not being able to conceive because:

- Calving to conception interval is increased.
- Intercalving period is increased.
- Total number of calving is reduced.

Dairy animals are homoeothermic animals with rectal temperature ranged between 101.3-102.8°C. So they exhibit optimum performance in their neutral environment which is known as thermo neutral zone (TNZ). In exotic dairy animals like HF and crossbred, this TNZ ranges between 1 and 25°C, and are called lower critical temperature (LCT)

and upper critical temperature (UCT). The width of the TNZ depends on age, species and breed, level of nutrition, previous state of temperature acclimatization and level of productivity. At LCT, the animal needs to increase metabolic heat production to maintain body temperature. At UCT, the animal increases heat production as a consequence of a rise in body temperature resulting for inadequate evaporative heat loss.

Climate change and nutrition

Research revealed that only due to decrease in dry matter intake, milk yield lower by 40-50%. Rest dip in milk yield due to various change in body due to heat stress. Udder cell need glucose to make lactose, the amount of later in the udder determines the milk quantity produced by animal. However, in summer, skeletal muscle start using more glucose, so less glucose goes to udder cell, as a result less lactose synthesis and milk quantity decrease (Wheelock et al., 2010). Increase demand of milk in summer lead to increase in milk price. On the other side due to summer stress, milk yield of dairy animals decreases leading to economic losses to the farmers. Enhancing dry matter intake or nutrient intake through high density feed along with proper summer management can increase the milk yield from our dairy animals. Feed the quality fibre in the form of green fodder/silage so that animal must receive appropriate amount of nutrients. Fibre has comparatively high heat increment as compared to other nutrients. Feeding ripened fodder or fodder with high fibre content lead to more heat production in animal body. As a consequence animal experience more heat stress. To avoid this internal heat, animal lower its intake. Therefore, feed the animal on quality fodder/silage.

Water is a very good source and nutrient to lower the stress due to soaring temperature. Milk contains 87% water and 4-5 litre water is required for 1 litre of milk production. Water help in removing heat from the body through perspiration and enhancing blood supply to peripheral organs. So, cold and clean water should be available all the time, near the animals and in shade. Animal prefer to remain in shade if water source is away.

Care of dairy animals under environmental stress

Dairy animals must be protected from environmental stress as follicle production in females goes down in summer season. Excessive heat or excessive chill causes hormonal imbalance. Environmental stress causes increase in prolactin hormone in blood which causes decline in reproduction. To protect animals from summer stress, shady trees must be planted around sheds. Many progressive farmers install water sprinklers or foggers at their farms and also bathe their animals 2-3 times in a day. Animals under heat stress should quickly be transferred to the cooler places, so that their risen body temperature can be controlled. Animals should be given shower with cool water and should be kept under the fans. During summer stress, digestion is also poor. So, animal

should be fed with good quality green fodder during cool hours of day. Animal should be groomed with brush thereby removing ectoparasites and increasing the blood flow. During winter, animals should be covered with blankets or gunny bags so that they may be protected from chilling wind. Therefore, farmers are advised to protect their animals from environmental stress so that reproductive efficiency may be maintained. Immediately, a veterinarian should be contacted to prevent any major loss/mortality due to environmental stress.

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GASTROENTERITIS IN DOGS: THE PREVALENT CLINICAL CONDITION AND ITS VIRAL CAUSES

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Abstract

Gastroenteritis is a common clinical illness in dogs, with a viral etiology detected in 40–60% of samples of diarrheal faeces. The viruses which can cause gastroenteritis in dogs are Parvovirus, Distemper and Corona virus. Rotavirus, Astrovirus, and Adenovirus are some of the other viruses that can cause canine enteritis. PCR, immunomigration assays, immunochromatography assays, and ELISA are some of the diagnostic methods used to detect DNA and RNA viruses. Supportive care is used to treat viral gastroenteritis. Each pet must be protected through effective immunisation in order to reduce the number of susceptible animals in a region and promote “herd immunity.”

Keywords: Dogs, gastroenteritis, immunisation, parvovirus, viruses

In dogs, gastroenteritis is a prevalent clinical condition, with viral etiology found in 40–60% of diarrhoeic faecal samples (Baumann et al., 2014). The viruses which can cause gastroenteritis in dogs are Parvovirus, Distemper and Corona virus. Rotavirus and Adenovirus are some of the other viruses that can cause canine enteritis and are studied extensively nowadays along with the major viral etiologies of gastroenteritis in canine population.

Canine parvovirus (CPV) is non enveloped DNA virus. It is the most well-known cause of transmissible viral diarrhoea in dogs. Oral contact with infected faeces or contaminated surfaces distributes canine parvovirus (e.g., soil, shoes, dog toys etc.). Puppies between the ages of 6 months and weaning are currently regarded the most vulnerable to CPV infection. Depression, anorexia, vomiting, a high fever, and severe diarrhoea are common early symptoms. The temperature rises somewhat in the early stages of the sickness, then drops to a subnormal level as vomiting and diarrhoea progress. The faeces has no constant appearance; it may be watery, yellow in coloration, or stained with blood in extreme cases. Rapid dehydration is a risk, and dogs may vomit and have diarrhoea till they die, which generally happens three days after symptoms appear. Clinical indications often appear 3–5 days after infection and last 5–7 days, depending on the virus's infectious load. The second type of CPV is cardiac syndrome, often known as myocarditis, which affects puppies less than three months. The most severe symptom of CPV myocarditis is sudden mortality in young puppies, generally around the age of four

weeks. Cold extremities, pale mucosae, and gasping respiration or fatal convulsions may be observed in the collapsed dying puppy. Puppies between the ages of 4 and 8 weeks develop acute cardiac failure with respiratory depression.

Domestic dogs are susceptible to canine distemper virus (CDV), which causes a serious systemic infection in unvaccinated or inadequately vaccinated animals. Canine Distemper is enveloped, RNA virus. The disease “distemper” is very contagious. A distinct sign of the illness is a diphasic fever, which appears 7 or 8 days after infection, drops quickly, then rises again by day 11 or 12. The disease is characterised by mucopurulent oculonasal discharges, conjunctivitis, respiratory distress, anorexia, vomiting, diarrhoea, and dehydration, as well as a cutaneous rash, though clinical signs of distemper are frequently absent or at first mild during this time (Kapil & Yeary, 2011).

Canine adenovirus (CAV), a non-enveloped, DNA virus. Direct contact with contaminated saliva, urine, or faeces allows the virus to enter the host. In addition to a high fever—often over 40°C—apathy, anorexia, abdominal pain, blood in the faeces, interstitial nephritis, acute/chronic hepatitis, tenderness, vomiting, and diarrhoea, CAV also causes these symptoms. As a result of anterior uveitis and oedema, dogs may experience bronchopneumonia, conjunctivitis, photophobia, and a transient corneal opacity, or “blue eye,” which may occur after clinical recovery. Canine Coronavirus (CCoV) is enveloped, RNA virus. It is transferred by the fecal-oral route. CCoV is a mild but highly contagious enteritis that affects young pups, usually under the age of 12 weeks. CCoV infection can be lethal in some situations, especially in puppies that are also infected with other diseases like canine parvovirus. Canine Rotavirus is non enveloped, double-stranded RNA virus so named because, when viewed under an electron microscope, its viral capsid resembles a wheel. It causes lethargy, anorexia, fever, diarrhoea, and vomiting in puppies fewer than two weeks old. (Fig. 1) Patients usually recover in two weeks, however severe enteritis in puppies under two weeks old has been reported to be deadly. Because rotavirus infection has no pathognomonic clinical indications, most dogs can be asymptomatic, and signs can often be confused with parvovirus, laboratory tests for differential diagnoses are required. Astroviruses are non-enveloped, RNA viruses with a distinctive star-shaped shape. A self-limiting gastroenteritis is the most common symptom of astrovirus infection in animals. Many infections are most likely undetected. In most cases, the incubation period lasts 1 to 4 days, followed by watery diarrhoea that lasts 1 to 4 days or more. Vomiting occurs frequently.

Diagnosis

Leukopenia is discovered by haematology, along with lymphopenia and an elevated PCV as a result of dehydration. Hypokalemia, decreased bilirubin, and the liver enzymes alanine transaminase and alkaline phosphatase are the most common electrolyte

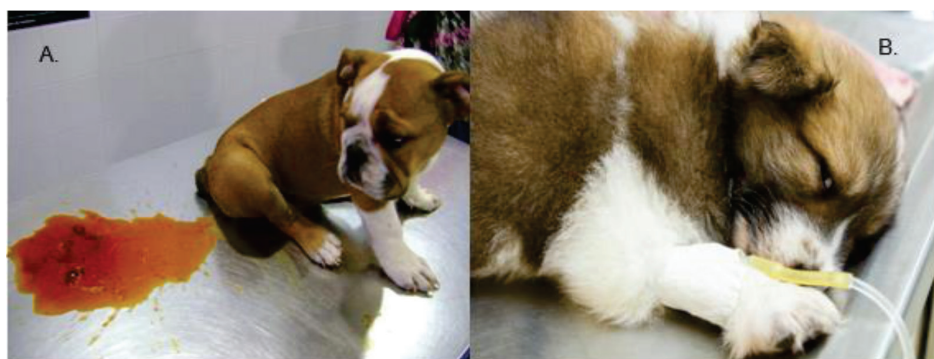


Fig. 1. Photographs showing hemorrhagic diarrhea (A), dull depressed (B) dog suffering from gastroenteritis

imbalances. Intestinal fluid and gas-filled loops, hypomotile intestines, and possibly thinned mucosal layers are among the largely nonspecific changes that abdominal radiography or ultrasonography can detect (Stander *et al.*, 2010). Rapid point-of-care tests like ELISA, immunomigration assays, and immunochromatography assays applied to faecal or rectal swab samples are the most economical assays for virus detection. A variety of PCR assays (such as real-time or conventional nested PCR, RT-PCR) are available from several veterinary diagnostic laboratories for the detection of DNA and RNA viruses.

Treatment

Viral gastroenteritis is treated with symptomatic support. Nothing should be given orally until the vomiting and diarrhoea have stopped for 24 hours. Fluid therapy is a crucial component of care. Those who appear dehydrated or those who are unable to tolerate oral fluids may be given intravenous fluids. Since there is significant vomiting and diarrhoea, it is preferable to administer intravenous fluid, particularly Ringer's solution @ 100-150 ml/kg/day. Following the correction of fluid deficits, maintenance IV fluid therapy is given at a rate of 70–120 mL/kg/day using an isotonic fluid that may also contain 20 mEq/L of potassium chloride. fluids at a continuous rate infusion of 2.5–5% (Hansen & Vigani., 2017) .It appears that the main method of transmission is dog faeces. In dogs with severe viral enteritis, parenteral administration of broad-spectrum bactericidal antibiotics is advised due to the significant risk of septicemia brought on by the disruption of the mucosal barrier and the ensuing profound neutropenia. Ampicillin, gentamicin, amoxicillin, and amoxicillin-cloxacillin are reasonable empirical options to provide defence against bacteria. In dogs with severe vomiting, metoclopramide, a dopaminergic antagonist, may be administered as a bolus or as a constant-rate infusion. The effectiveness of antiemetic treatment for dogs has significantly increased since the

recent introduction of maropitant, a neurokinin1 receptor antagonist (Prittie, 2004). H_2 receptor blockers, like ranitidine or famotidine, can control the production of gastric acid. In cases of severe diarrhoea, oral antidiarrheals such as bismuth subsalicylate or loperamide can be administered. Hemorrhages can be controlled by administering coagulants like adrenochrome monosemicarbazone. Immune system boosters include vitamin C.

Effective immunisation is crucial for the protection of each pet as well as for reducing the number of susceptible animals in a region and encouraging “herd immunity” (Day *et al.*, 2016). Modified live vaccines (MLVs) are currently used throughout the world to provide prolonged immunity (7 years or longer), which would provide protection from both disease and infection (Schultz *et al.*, 2010). Typically, the first round of a puppy’s vaccinations begins at 6 to 8 weeks of age, and continues every 2 to 4 weeks until the puppy is 16 weeks old or older” (Day *et al.*, 2016). Combination of MLV for CPV and CD can be used (Fig. 2).



Fig. 2 Commercially Available Vaccines

Prevention

Transmission mainly spreads through dog faeces. Thus for disinfection, sodium hypochlorite solutions (like Clorox -5.25% sodium hypochlorite solution containing 5% available chlorine by weight by Clorox chemical company) are advised (Cavalli *et al.*, 2018). Isolating puppies who are at risk for infection is the only way to stop it. Since well-vaccinated adults with normal faeces can still shed viruses like CPV and be potential sources of exposure, it is crucial that clients are educated to prevent exposing at-risk puppies to other dogs before the puppy has completed its full series of vaccinations. Employees in animal shelters and veterinary clinics are expected to wash their hands thoroughly after each patient and put on fresh gloves. Thermometers, stethoscopes, fluid pumps, tables, cages, and bedding, among other items, should all be thoroughly cleaned and disinfected on a regular basis using a detergent and virucidal solutions. Barrier methods with disposable gloves, a cap, a gown, and booties should be worn when

handling any diarrheal patient, even one who has had a negative faecal ELISA test, to prevent cross-contamination and infection spread.

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LUMPY SKIN DISEASE

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Abstract

One of the main health issues affecting the livestock economy now-a-days is lumpy skin disease, which is caused on by the lumpy skin disease virus and is a transboundary disease. The majority of causes of infection are skin nodules. The virus is mostly mechanically transmitted to susceptible hosts by hematophagous arthropods, biting flies, mosquitoes and ticks. Under experimental conditions, the typical lumpy skin disease lesions may appear 7 to 14 days after infection. Firm, confined, few (mild forms) to numerous (severe forms) skin nodules that occasionally invade the mucous membranes of the respiratory system, urogenital system and other internal organs are the hallmarks of lumpy skin disease. The most efficient method of reducing the spread and financial burden of lumpy skin disease is mass immunization along vector control and suitable control measures.

Keywords: *Capripoxvirus, cutaneous nodules, lumpy skin disease, transmission*

Lumpy skin disease (LSD) also known as, Neethling virus disease, pseudo-urticaria, exanthema nodularis bovi is a poxvirus disease in cattle. It causes nodules on the skin, mucosal membranes and internal organs as well as emaciation, swollen lymph nodes, skin oedema and occasionally mortality. It is significant economically as it can result in a temporary decrease in milk yield, temporary or permanent infertility in bulls, damage to hides and occasionally death. LSD can lead to acute or subacute illness in water buffalo and cattle. Cattle of all ages and breeds are infected. Due to its potential for rapid spread and considerable economic losses, the World Organization for Animal Health (OIE) has listed this transboundary disease as a notifiable disease. Lumpy Skin Disease is caused by Lumpy skin disease virus which belongs to Poxviridae family and genus Capripoxvirus, antigenically, it is very similar to the goat and sheep pox virus. Capripoxvirus is double-stranded DNA viruses of roughly 150 kilobase pairs (230–260 nm) size (Onyejekwe et al., 2019). They have capsids and are brick- or oval-shaped. While LSDV can experimentally infect sheep and goats, there have been no reports of natural LSDV infections in sheep and goats.

Transmission

At normal temperatures, LSDV can survive for a very long time in the environment, especially in dried scabs. The virus reportedly lingers in desiccated crusts

for up to 35 days, in necrotic skin nodules for up to 33 days, and in air-dried hides for at least 18 days. The virus can be rendered inactive at temperatures of 65°C for 30 minutes and 55°C for 2 hours (Mulatu and Feyisa, 2018). Skin lesions or nodules are thought to be the most significant source of infection for healthy animals since the virus may survive there for an extended period of time and has a high affinity for dermal tissue (Babiuk et al., 2008). Arthropods, particularly blood-sucking insects are the main mode of transmission. The mosquito genera *Aedes* and *Culex* have LSDV isolates (Kahana-Sutin et al., 2017). Other mechanical vectors may include flies like *Stomoxys calcitrans* and *Biomyia fasciata*, as well as other insects like ticks (Ixodid, *Amblyomma hebraeum* and *Rhipicephalus appendiculatus*). Other mode of transmission includes transmission through contaminated food and water and direct transmission during the later stages of the illness through saliva, nasal secretions and semen are all ways that the LSDV is spread. According to some research, there is little direct virus transmission, at least in the early stages of the disease, compared to the greater significance of indirect transmission. These studies also found no significant link between cattle density and infection rates (Mulatu and Feyisa, 2018). Since LSD outbreaks tend to occur during the summer, when arthropod activity is at its peak, this could mean that different types of vectors, particularly those that feed on blood, are involved in the propagation of the virus.

Incubation period

The incubation period ranges from 2 to 5 weeks. Lesions first occur at the inoculation site in four to twenty days after the disease has been incubating in the field for two to five weeks. The incubation period is 28 days according to the 2016 Terrestrial Animal Health Code published by the World Organization for Animal Health (OIE).

Morbidity and mortality

Mortality is typically low (between 1 and 3 percent) and morbidity seldom exceeds 20 percent but can range from 3 to 85 percent. The morbidity and mortality of LSD can vary greatly depending on the cattle breed, the population's immune condition, the insect vectors transmitting the disease (Al-Salihi, 2014).

Clinical Signs

Depending on the number of lumps, LSD can be divided into moderate and severe forms (nodules). The age of the host, the strain of the Capripoxvirus, the host's immunological state and breed are just a few of the variables that affect the severity of the clinical signs of LSD. The clinical signs of slightly infected cattle include the formation of one or two lumps or nodules within two days after the commencement of the fever (1 to 5 cm), Emaciation, agalactia, excessive salivation, anorexia, nasal and ocular discharge and depression. Furthermore, nodular lesions may be seen on an animal's body (Figure 1) and are painful and hyperemic, particularly in the skin of the scrotum, legs,



Fig. 1. Photograph showing ulceration of the nodular lesions



Fig. 2. Photograph showing typical LSD nodular lesions

back, nares lower ear, nasal and oral mucosa, perineum, eyelids, tail and lower ear (Salib & Osman, 2011). Ulceration of nodules occurs after 1 to 2 months of infection (Figure 2). High pyrexia ($40\text{--}41.5^{\circ}\text{C}$) occurs in extreme instances and may last for 7–12 days. Anorexia and severe depression and on entire body of the infected animal, several (>100) nodules that are often quite consistent in size in animal are observed. Necrosis of nodules increases the likelihood of secondary infection. The percentage of animals who acquire skin lesions is only 40–50%.

Due to necrotic plaques and characteristic LSD lesions in the oral cavity, conjunctiva, and nasal cavity, respectively, affected animals also display excessive salivation, lacrimation, nasal discharge, and emaciation. Lymphadenopathy and enlargement of superficial lymph nodes are additional LSD characteristics. Additionally, milk production may decrease noticeably and pregnant cows may abort. Bulls with the infection also develop orchitis and testicular swelling and may have temporary or permanent sterility (Constable et al., 2017). Clinical symptoms in young calves and lactating cows are typically more severe and occur due to intrauterine infection. Overall, even within the same herd, lesions might differ significantly from one animal to another. Recovery is slow, and wounds are frequently left on an animal's skin.

Histopathological findings

In the keratinocytes, macrophages, endothelial cells, and pericytes of skin nodules, histological analysis may reveal pathognomonic eosinophilic intracytoplasmic inclusion bodies that are linked to the escalating degeneration of spinosum cells. Inflammatory cells such as macrophages, lymphocytes and eosinophils are found infiltrating the superficial dermal tissue of affected locations.

Diagnosis

The characteristic clinical indications of LSD, such as generalised nodular skin lesions and enlarged superficial lymph nodes in infected animals, can be used to make the diagnosis, together with test confirmation of the virus or antigen's presence. The most effective technique for identifying capripox viral antibody and antigen is viral neutralisation test and electron microscopy, respectively (Tuppuraine et al., 2011). These methods are considered as gold standard for LSD diagnosis. Using conventional or real-time PCR techniques, the clinical diagnosis of LSD can be confirmed (Tuppuraine et al., 2005). ELISAs Indirect fluorescent antibody test (IFAT) can also be used to detect antibodies against LSD.

Treatment, Prevention and Control

The virus that causes lumpy skin disease has no recognised cure. To treat secondary bacterial infections, fever, inflammation, or to increase the animal's appetite, antibiotics, anti-inflammatory medications, or a vitamin injection are sometimes utilised. Therefore, the only effective way to control the disease in endemic areas is by vaccination. In the past, four live attenuated strains of the capripoxvirus vaccine have been used to manage LSDV outbreaks *viz*; the LSDV strain from South Africa, the Yugoslavian RM 65 sheep pox strain, the Romanian sheep pox strain and the Kenyan sheep and goat pox virus strain (Abutarbush, 2017). Recently a new live attenuated vaccine, Lumpi-ProVac Ind has been developed by The Indian Veterinary Research Institute (IVRI) at Izatnagar, Uttar Pradesh and National Research Centre on Equines (NRCE), India.

Because all strains of Capripoxvirus share a key neutralising site, cattle are immune from vaccines generated from sheep or goats. There are no diagnostic tools or vaccines to distinguish between infected and vaccinated animals (DIVA). Because biting flies and specific kinds of ticks are most likely the disease's primary means of transmission, quarantine and movement restrictions are not very helpful in controlling the condition known as Lumpy Skin Disease. Nevertheless, pest control was ineffective. However, the use of pesticides and repellents simultaneously can be effective means of halting the spread of LSD. Quarantines, movement control, bug control (insecticides/repellents) and stamping-out procedures, followed by proper washing and disinfection of the animal farm, proper disposal of the carcass are crucial in preventing the spread of LSD because pests, animals and animal products can carry disease. Another means of control is vaccination.

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HOUSING OF CALF- AN INDISPENSABLE ASPECT OF DAIRY FARMING

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Abstract

For a good profitable dairy farming scientific rearing of calf is very important as it represents the future replacement stock. Housing of calves is an important aspect in this regard. If the housing conditions are not adequate it may lead to stress to the calves, may cause infections in the calves increasing their morbidity and mortality leading to deprived growth and further reproduction and production losses in the near future. There are various factors which contribute to the successful housing management such as: individual housing/group housing, floor space requirements, feeding and water space requirements, sufficient air space and adequate ventilation system etc.

Keywords: Calf, dairy, housing, replacement stock, scientific rearing

Calves are the future sire or dam. Scientific rearing of calf will help to produce a healthy animal that will produce high yields in the subsequent times. Good care of calf is pivotal for future stability of the dairy sector and is also necessarily required for preserving & maintaining the good germplasm (Tiwari et al., 2007). An important aspect in healthy rearing of calves is good housing. A well-managed housing system helps to provide an ample space for feeding, and their movement, prevents calves from humidity, heat and cold stress, and also ensures proper ventilation in the calf sheds. However, most of the mortality occurs in calves due to bad housing management which further contributes to loss of a future cow/buffalo. Generally, calves are reared in ordinary stall barns or in the same house along with the other adult cattle.

There are lot of queries in the mind of the farmers in context to whether calves should be housed in individual pens or in group with other calves? Farmer should prefer to house the calves in individual pens at least up to one month of age and if possible up to three months of age so as to avoid the chances of infections through naval suckling and through facial or other contact. After one month of age farmer can house the calves in group pens. Some farmers prefer railed pen division in calf housing but the contact is not completely prevented as compared to individual calf housing. After three months, three to five calves can be kept in a single pen. Calf pens should be well ventilated, well

lighted, clean, dry, adequately bedded using soft material.

Individual housing of calves (Fig. 1)

Newborn calves are individually housed during the first few weeks. Since, a calf is quiet susceptible to all kinds of infections, the death rate is high in initial first few days. Calves up to three months should be housed individually as they have a suckling reflex and in absence of such stimulus they have a tendency of cross sucking one another which may lead to transmission of various diseases among them. Thus, individual pens facilitate an ease in keeping an eye on the individual calf and monitor their growth and health. Single pen may have open or partly open sides. Feeding box is generally attached to the gate which facilitates feeding of calf starter to them. Space requirements for individual calf pens are mentioned in table 1.



Fig. 1. Photograph showing individual housing of calf

Table 1. Body weight wise space requirement for individual calf pens (Costa et al., 2016)

Calf weight	Pen size per calf
<60 Kg	1 m × 1.5 m
60-80 Kg	1 m × 1.8 m

Group housing of calves (Fig. 2)

After rearing of calves in individual calf pens for nearly three months they can be housed in groups of three or five calves per pen. The farmer should try that he should not accommodate calves with the adult animals. Generally, for housing the calves should classify the calves into three age groups:

- Calves below the age of three months
- Three to six months old calves
- Calves over six months of age

Space requirements for group housing calves are mentioned in table 2.

Body weight wise space requirement for group housing of calves (Costa et al., 2016)

Calf weight	Space requirements per calf
50- 84 Kg	1.5 m ²
58-140 Kg	1.8 m ²



Fig. 2. Photograph showing group housing system of calves

Curtis et al. (1988) compared behaviour, health and production of calves housed under different management conditions and reported that the housing calves individually reduced mortality and morbidity in comparison to group housing.

Floors and drainage

Various types of floor for calf housing are: Pucca floor (cemented), Kuccha floor (earthen), Mixed type (both kuccha and pucca) and Brick paved. Generally, farmers have cemented floors in their calf sheds. However, the floor type if is kuccha it should be provided with suitable dry bedding in order to prevent various foot problems and other infections. If the floor is cemented it should be smooth so that there is an ease in proper drainage and cleaning.

Type of bedding material used in calf sheds

It is commonly seen that farmers generally use feed waste or wheat or rice straw as a source of bedding for the calves. But, in present scenario various farmers also prefer mats as floor bedding for their calf sheds.

However, dampness in a calf shed may have a negative impact on the growth performance of the calves as it increases the chances of infection and possibility of contamination. A calf should always have a dry bed. The incidence of diseases increased by 90% where calf sheds was not properly bedded (Mustafa et al., 2010). The floor slope in calf shed (straw bedded system) should be in the ratio of 1:20. If the farmer finds that drainage is not competent, then he can cut the connection from the concrete and can form the 75-100 mm gutters away from target area. Floor space requirements of calf shed are mentioned in table 3.

Table 3. Floor space requirement of calf shed (Costa et al., 2016)

Age group	Space (square feet)	Covered area (m ²)	Open area (m ²)
Below one month of age	20-25	1.0	2.0
Three to six months of age	30	1.5	3.0

Types of roof for calf sheds

Various types of roofs for calf sheds are: Asbestos sheet roof, Galvanized iron sheet roof, Thatched roof and Concrete roof. However, it is generally recommended that thatched roofs should be used in the calf sheds as it keeps sheds cool during the summers. However, galvanized sheet roofs and asbestos sheet roofs generally keeps the shed very warm in summers, which may lead to heat stress in the calves leading to lower calf growth performance. Concrete roofs however reduce heat stress inside the shed but they are generally more costly than other type of sheets used as a roof material.

Feed and water

Calves should have a free access to fresh and clean water even when on milk. Drinking water increases the calf starter intake and enhances speed of rumen development. Water troughs, drinkers or feeding troughs should be located above or near drainage with clear access for stock and easy access for the farmer to provide proper cleaning. Feeding and watering space and dimensions of feed mangers requirements for calves are mentioned in table 4 and 5.

Table 4. Feeding and watering space requirement for calves (Kertz et al., 2017)

Feeding space/ manger space (cm)	50/ calf
Water trough space (cm)	10-15/calf

Dimensions of feed mangers (Kertz et al., 2017)

Width (cm)	Depth (cm)	Height of inner wall (cm)
40	15	20

Frequently asked questions by the dairy farmers under field conditions in respect to calf housing?

Q1. How much space does a calf need?

Ans. Allow at least 1.5 m² /calf for group housing or 2.0 m² / calf for individual pens. Larger calves may need a greater area (2.5 m² / calf) to carry out their normal behaviour.

Q2. How much straw can be used to ensure dry conditions on concrete floor?

Ans. Calves require about 15-20 kg/head/week of straw bedding in order to maintain dry conditions on the concrete floor. This quantity can however be reduced by using thin slats (a narrow piece of metal/wood) under the straw to cut down the expenses of straw.

Q3. How can farmers ensure whether the calf bedding material is wet/ dry?

Ans. For this purpose the farmer has to kneel down with all his weight on the bedded floor. If his knees of trouser are wet, this indicates the bedding needs to be changed.

Q4. How to choose appropriate bedding material for calves and what else can be used as a source of bedding material for the calves?

Ans. While selecting bedding materials it is important to check the price and also the degree to which the material compacts over the time. Avoid using dusty material like saw dust, sand etc. as it may aggravate respiratory problems. Farmers can use straw, hay, bark chips, wood shavings and rubber mats along with other bedding material.

Q5. How much air space is required in a calf shed?

Ans. Allow minimum air space of 6-8 m³ per calf. Air space in a calf shed can be calculated by the following formula = Shed length x width x height (in metres)/ number of calves

Q6. How much minimum trough space/calf is required?

Ans. It is 35 cm per calf.

Q7. How much relative humidity and temperature is ideal for a calf in a shed?

Ans. To avoid stress, ideal relative humidity and temperature in a calf shed should be about 65% and 10-20° C, respectively

Tips for better housing management for the dairy farmers:

- Maintain proper hygiene in the calf sheds by regular cleaning routines.
- Calf houses should be dry, free from dampness, moisture, dust and contamination so as to enhance performance of calves.
- Control of moisture through proper ventilation and adequate drainage facilities should be done on regular basis.
- Ensure there is a separate cleanable area so that any moisture from the washing area does not enter in the calf shed.
- Sick animals should not be housed with the healthy animals. They should be isolated away in sick pens or in other calf shed.

- To avoid the chances of respiratory diseases ensure constant fresh air in the shed through ventilation. Also, check for the airspeed which should not be more than 0.5 metres per second.
- During hot weather, maintain the growth performance of calves by providing shade and cold water. Even sprinklers and centrifugal fans can be used in the calf sheds so as to avoid the hot weather stress.
- During cold weather, maintain growth rates by ensuring there is plenty of clean and dry bedding, adding extra energy diet or increasing the feeding. Use of calf jackets or heaters can be also done in case of extreme cold conditions.
- Continuous access to fresh and clean water should be made to the calves. It also enhances the rumen development.
- Calf sheds should not be in hot, windy or wet locations and care should be taken to ensure they provide adequate ventilation, drainage and space.
- Avoid overcrowding in the calf sheds and do not keep adult stock with the calves (Fig. 3).

Hence, the aim of calf housing should be to provide a comfortable and stress-free environment to the calves so as to ensure their welfare which will produce a good, healthy calf subsequently becoming a productive cow/buffalo in near future.



Fig. 3. Avoid keeping adult stock with calves

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PREPARATION OF PLASTINATED SPECIMENS BY LOW COST INNOVATIVE METHOD

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Abstract

Plastination is carried out in different institutions in India and abroad because the plastinated specimens are more durable, permanent, and easy to handle. These specimens can be stored at room temperature and the high teaching value. The preparation of plastinated specimens is required as it reduces formalin exposure to the students, teachers and laboratory staff in the gross anatomy laboratory. There are different steps used in a sequence during plastination i.e. fixation, dehydration, impregnation and curing. A low cost innovative technique was used to prepare the plastinated specimens in which modified plastination solution was used for impregnation and time required for different organs have been standardized as 7 days for kidneys, 12 days for lungs/ liver and 15 days for spleen/heart. As per our experience, it was observed that the plastinated specimens are preferred by the students to learn anatomical details of the organs.

Keywords: *Curing, Dehydration, Fixation, Impregnation, Plastination*

The formaldehyde has been used over a century as a disinfectant and preservative agent to study the gross anatomy specimens. It is a substance with which most of the first year Veterinary students has a first-hand experience to study the gross anatomy. Even after completion of B.V.Sc & A. H., no veterinarian can forget his/her first day in anatomy lab when he/she was exposed to formalin. The formalin fixed specimens are wet, slippery and difficult to handle. Formaldehyde exposure showed an irritant effect on the eye and upper respiratory tract of teachers, technicians and students working in the anatomy and pathology departments. The experimental studies showed that the lab animals exposed to different concentrations of formaldehyde showed carcinogenic effect in the respiratory organs (Bansal et. al., 2011). It has also been reported that the persons exposed to formaldehyde showed irritant symptoms and cytogenic changes in epithelial cells of the mouth and in blood lymphocytes (Kriebel et. al., 1993; Suruda et. al., 1993).

As the health hazards of formaldehyde exposure is of a great concern, so an alternative technique is used to preserve the anatomical specimens is plastination technique. Dr. Gunther Von Hagens developed plastination in 1978, a unique technique of tissue preservation. In this process, water and lipids in biological tissues are replaced

by curable polymers (silicone, epoxy resins, polyester) which are subsequently hardened, resulting in dry, odourless, non-toxic and durable specimens. Plastination is carried out in many institutions worldwide and obtained great acceptance particularly because of the durability and the high teaching value of plastinated specimens (Sittel et. al., 1997). For teaching and demonstration purposes, plastinated specimens are preferred over models and organs preserved in formaldehyde (Sethi et. al., 2008).

Keeping in view, the advantages of plastinated specimens, an attempt was carried out in the Department of Veterinary Anatomy, GADVASU, Ludhiana to establish the technique of plastination for preparation of specimens for teaching and demonstration purposes. By adopting this technique, the use and exposure to the formaldehyde can be minimized. In our department, we followed the low cost innovative technique which is very effective and easy to perform.

The plastination procedure includes fixation, dehydration, impregnation and curing.

1. **Fixation:** The specimens were collected, cleaned and washed properly. After cleaning, the specimens were fixed in 5-20% formalin solution or 5% formal saline solution. The inclusion of saline enhances the quality as well as colour of the preserved specimens. In present study, we have used already fixed specimens and freshly fixed formalin specimens to compare the texture.
2. **Dehydration:** Dehydration is removal of water content from the specimen. Dehydration is important because the resin does not dissolve in the water. The resin penetrates into the dehydrated specimen very smoothly. Dehydration was done through three consecutive changes in acetone for 10-15 days each (depending upon size and consistency of specimen) at room temperature. The amount of dehydrating solution should be ten times the weight of specimen. In the western world, this process is enhanced by using vacuum pump under freezing temperature at -4 to -20°C, which is very costly.
3. **Impregnation:** It is the process in which intermediary solvent is replaced with a polymer at room temperature. Normally, epoxy resin is used for the impregnation and is again carried out in a vacuum chamber under cold condition which is a costly method. This process can also be achieved by a mixture of melamine and chloroform, which is again a costly procedure. In our lab, the dehydrated specimens were immersed for 7-15 days in modified plastination solution (150 gm thermocol and 500 gm petroleum Jelly in 10 litre of chloroform) as described by Ramakrishana and Nanjappa (2018). We have standardized 7 days for kidneys, 12 days for lungs/ liver and 15 days for spleen/heart.

4. **Curing:** In present technique, no chemicals were used in curing. The specimens were taken out from the plastination solution, cleaned and allowed to air dry. After that a single coating with touch wood solution was done to make the specimens presentable for demonstration. However in earlier methods, S10 or S6 or melamine plus hardener were used as a gas curing.

Thus, the present technique is very cheap as compared to the other methods. The plastinated organs prepared are dry to touch, clean, non-toxic, odorless and most of them maintain their original shape and natural look. The specimens of spleen (Fig. 1) and kidney (Fig. 2) of buffalo, lungs of goat (Fig. 3) and external and internal structure of buffalo heart (Fig. 4) were prepared, which are permanent, durable and easy to handle and can be stored at room temperature forever. Our experience showed that the plastinated

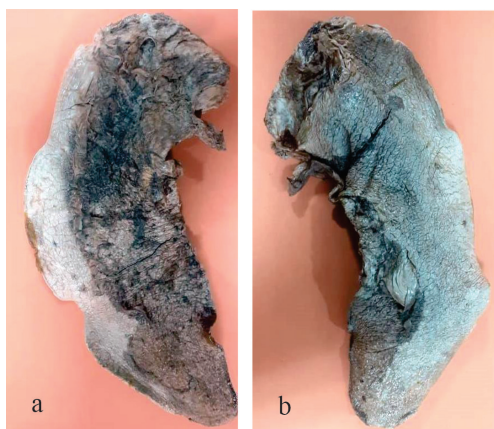


Fig. 1 Visceral (a) and Parietal surface (b) of spleen in buffalo



Fig. 2 Right Kidney of buffalo



Fig. 3 Lungs of goat



Fig. 4. External (a) and Internal (b) structure of buffalo heart

specimens are preferred by the students to learn anatomical details of the organs. They also serve as an excellent museum specimens and used for teaching purpose. However, disadvantage of plastination are shrinkage and inability to study some of the deep structures by removing the superficial parts.

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ANCESTRY, TEMPERAMENT, FACTS, AND GROOMING OF INDIGENOUS 'GADDI' DOG BREED

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Abstract

A dog is a domesticated variant of the Gray Wolf that belongs to the 'Canis lupus familiaris' subspecies. The Gaddi dog is a massive mastiff-type breed from northern India. This breed is popularly known as the 'guardian dogs' due to their aggression and agility which best suits them for the herding of livestock by the local tribes. They are very protective of themselves and their families and are quite easy to train. They are mostly fluffy in appearance and have hairy and long curly tails. Their double coat's long hair, cushioned paws, and long-haired ears make them appropriate for living in such frigid, stress-tolerant, and thermo-tolerant areas.

Keywords: *Gaddi, Himalyan sheepdog, indigenous, shepherd*

Dogs were domesticated for the first time by humans 10,000 to 15,000 years ago. India is bestowed with several breeds of dogs many of which are yet to be characterized at the molecular level. These are primarily used for guarding farms and farmhouses, as well as shepherding livestock species during grazing, migration, and hunting. In human history, the Domestic Dog has been one of the most extensively kept working and companion animals. Some of the well-known indigenous dog breeds are Caravan Hound, Chippiparai, Rajapalayam, Mudhol Hound, Indian Pariah Dog, Indian Spitz, Rampur Hound, Indian Mastiff, Kombai, Gaddi Kutta and Kanni. The National Bureau of Animal Genetic Resources (NBAGR) is the Government body responsible for identifying and declaring new breeds of livestock and dogs. NBAGR identified Rajapalayam, Chippiparai, and Mudhol Hound (<https://nbagr.icar.gov.in/en/new-breeds-lines/>). First, Rajapalayam is an ancient canine that has lived for hundreds of years. With a stretched nose and a pointy forehead, the dog's head is attractive. The hanging ears have dark brown eyes and scissors-bite jaws. The tail is curled and long. Second, Chippiparai grows in the town of Chippiparai in Madurai, Tamil Nadu, India, where its name was coined and became popular. They are produced by royal families in Virudhunagar's Chippiparai area, 53 kilometers south of Madurai in Tamil Nadu, and they continue to be a symbol of grandeur and dignity for the kings of Tirunelveli and Madurai. Chippiparai was a fawn-colored medium-sized dog with a tucked-up belly. Third Mudhol Hounds are beautiful sight hounds native to Maharashtra and Karnataka. The dog has a tall, narrow head with

a pointed muzzle, a long, muscular neck that fits nicely into the shoulders, and a low, tapering tail. Different breeds of dogs are characterized by their unique morphology, behavior, and disease susceptibility.

There are some other Gaddi, like Black Gaddi, Ban Gaddi, as well *Gaddi Kutta* (frequently called by local people of Northern India) or Gaddi dogs are frequently confused with the Tibetan Mastiff, but they are entirely different.

Origin of Gaddi Dogs

It's not clear where this breed originated. They are mastiffs without a doubt, but their pedigree has been lost over time. According to local tradition, they are a mixed between Himalayan wild dogs and Tibetan Mastiffs. The popular Gaddi breed is available mainly in the state of Himachal Pradesh, Northern region of Punjab. Uttarakhand, Jammu and other states in India. So, these dogs are also known as Himalayan Sheepdogs, look to be a strong and healthy breed. Weak canines couldn't live in the Himalayas, therefore only parasite- and virus-resistant dogs survived. *Gaddi Kutta*, however, is thought to have a long history in the North Indian regions. According to mythology, they were created when Mahidant of Merut and According to Indian kennel associations, the Gaddi Kutta is not a "pure" breed.

Gaddi Dogs Characteristics, Looks and Grooming

In the harsh and treacherous conditions of the Himalayas, Himalayan sheepdogs were highly intelligent and capable shepherders and defenders. They move with the dexterity of a champion fighter. The body weight is approximately 35-45 kg (female adult), and 40-45 kg (male adult). The Typical height is between 26 and 34 inches (66 and 86 cm) as shown in Figure 1. The body shape is square with a broader chest that merges into their abdomen. These dogs are cat-friendly and moderately child-friendly. These dogs are known for being strong enough to protect cattle from larger predators like snow leopards, and they can herd lost sheep or goats back to their pens, just like other clever, caring, and affectionate dogs towards their family and if they are well socialised. With such a large frame, they require a lot of activity and suffer greatly if they do not get it. Repetitive games bore them rapidly.

They stand over 34 inches tall and weight nearly 13 Kg, making them big and intimidating. Their overcoat is brownish-black, with tan or brown spots on their face and underbelly to compliment it as shown in Figure 1. Indian dogs and mastiffs are known for having tails that curl back over their bodies and are extremely fluffy. They shed heavily in the fall and spring and moderately all year. Brushing the dog regularly and bathing him every two weeks is sufficient for an Indian Gaddi dog. The Indian gaddi dog may be left alone for long periods of time. Therefore, Gaddi Kuttas come in a variety of colors, including white, black, and light brown, although the breed standard is black and tan.

Temperament and Behaviour

They're enormous, aggressive, and highly powerful. The *Gaddi Kutta* was originally bred as a hunter, but they were later used as shepherd dogs due to their ability to take down a snow leopard, earning them the nickname Indian Panther Hound. These dogs are considered to be fairly amiable for their size. These dogs will be wonderful among people they know and will be used strictly for herding, with obedience and socialising being the only training required.

Health and Reproducibility

Gaddi dog is comparatively a healthy breed, but there are certain health issues like arthritis, bloat, and hip dysplasia. They have a higher energy level than other dog breeds. They tolerate adverse weather conditions and need a lot of exercise. The reproductive cycle of Gaddi female dogs begins with the first called Proestrus period which lasts roughly 9 days and the other period is known as the Anestrus. It is the time frame between heat periods normally lasts about six months. Litter size is generally around 4-8 puppies, although some local Gaddi-raisers claim that this breed can whelp up to 10 puppies.



Fig. 1: General appearance of indigenous Gaddi dogs

SURVEY ON THE JOB SATISFACTION STATUS AMONG THE ANIMAL HUSBANDRY AND VETERINARY RELATED QUALIFIED-PROFILES IN INDIA

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Abstract

The aim of the Google form based WhatsApp-survey was to evaluate the job satisfaction of the animal husbandry and veterinary related professionals employed in different cadres in India. The survey form was circulated to about 2000 heterogeneous veterinarians, and responses were received from 471. The participants were requested to disclose their personal details including educational qualification and to submit their views regarding various parameters of job satisfaction viz. salary, attitude of senior officers, infrastructure at their work-place and whether they would like their siblings or kin to take up their profile in the present organization. Results revealed that the qualification and experience had a significant association with their responses regarding whether they would recommend the profession to near and dear ones; however, there was no association with respect to job profile or organizational sector. Further, their feedback on the available opportunities was significantly associated with qualification, job profile and organization sector. Motivation was observed to be influenced with regards to the differences in the qualification. Besides, the concern of availability of infrastructure and subordinate staff, use of modern technology and satisfaction from remuneration was also significantly associated with qualification, job profile, experience and the type of organization.

Keywords: Motivation, professional satisfaction, survey study, veterinarian

Most often, the veterinary-related profiles remain hidden from the public gaze especially in urban life, and these persons are a sort of back-end employees. Public continues to get regular supply of milk, eggs, meat, wool etc. because of the essential and uninterrupted services provided by the professionals of this field, but rarely come face to face directly with the professionals of this field. So it is important to conduct regular surveys to understand the working environment of the animal husbandry and veterinary professionals with respect to the availability of the infrastructure and subordinate staff, amenities, promotional avenues, adoption to latest technology etc. Therefore, the objective of this survey was to estimate the prevalence of job satisfaction among animal husbandry and veterinary related qualified-profiles working in India.

Population and Nature of the Study: The study included professionally qualified (Diploma, Graduates and above) persons employed in the animal husbandry and

veterinary related profiles dealing with the treatment of domestic animals and pets as well as management practices such as housing, nutrition and breeding. Some of the postgraduates (MVSc and PhD) were also employed as teaching faculty and as Researchers in the Veterinary Colleges.

Due to Covid-19 pandemic, it was not possible to contact the participants personally. Therefore, modern communication technology i.e. Google-form was created and circulated among various WhatsApp groups of the employed-veterinarians spanning over many states of India. In the questionnaire, all the participants were first requested to disclose their educational qualification, profile and the length of their service, followed by to submit their views regarding different parameters of job satisfaction viz. salary, attitude of senior officers, infrastructure at their work-place and whether they would like their siblings or kin to take up their profile in the present organization. The participants were not required mandatorily to disclose their identity (mobile number or e-mail ID), and some participants disclosed it voluntarily. These respondents helped the surveyor in verifying the authenticity and reliability of the responses on random basis. The participants were not selected consciously from any particular region or profile. A conscious effort was made so as not to ask participants directly about the job-satisfaction. The scale of measurement in this survey was nominal.

Results

The survey form was circulated to about 2000 heterogeneous veterinarians and out of these 471 responses received. As Government is the major employer in Veterinary and Animal husbandry related profiles, most of the participants in this survey are also employed in the government sector. This study was a sample type enquiry, mainly focusing upon the analysis of the characteristics of the job-satisfaction of the veterinarians.

Table 1: Qualification and Experience wise breakup of participants

Experience	Educational Qualification				Total	Percent
	B.V.Sc. & A.H.	M.V.Sc.	Ph.D.	Diploma after 10 or 10 +2		
0-5 years	61	83	6	2	152	32.3
5-10 years	39	57	8	4	108	22.9
10-20 years	40	40	8	3	91	19.3
>20 years	71	40	9		120	25.5
Total	211	220	31	9	471	100
Percent	44.8	46.7	6.6	1.9	100	

The details of the qualification and experience of the participants has been depicted in Table 1. The majority of the participants (32.3%; n=152) were serving since 0-5 years, followed by more than 20 years (25.5%, n=120), 5-10 years (22.9%), 10-20 years (19.3%; n=91). The above data inferred near uniform distribution of animal husbandry and veterinary professionals in the survey.

As can be seen from the above diagram, 44.8% of the respondents had graduation degree in veterinary sciences, while 46.7% had done their post-graduation in veterinary sciences, while 6.6% had Ph.D. in veterinary sciences, and 1.9% of the respondents were diploma holders.

In response to query about promotional opportunities, the overwhelming majority i.e. 76.6% of the respondents (Fig. 1) felt that adequate opportunities of promotion are not available in their cadre. Opportunities for promotion may prove to be a great moral-boosting and motivating factor for the employees. So, the veterinary and Animal Husbandry related qualified profiles in India are clearly at a disadvantageous position.

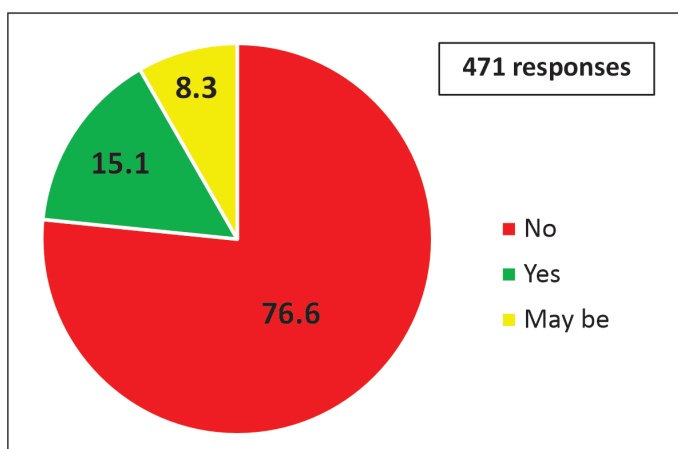


Fig. 1. Do you think that adequate opportunities for promotion available in your cadre (%)?

A huge majority of the respondents i.e. 85.4% expressed their concern about insufficient subordinate staff in their working conditions (Fig. 2). It is pertinent to note that in almost all of the states, the veterinarians have been given the rank of Gazetted Officer, thus lack of subordinate staff is certainly a demotivating factor.

Majority of the respondents i.e. 76.9% in this survey were clinicians followed by those occupying administrative posts, teaching faculty and the para-veterinary staff.

As can be seen from the above Pie diagram 69.2% of the respondents feel that even their duties are not well defined. When read with the results of the earlier question regarding non-sufficient subordinate staff, it clearly means that the veterinarians are made to perform the duties of their subordinate staff which again has a demotivating effect.

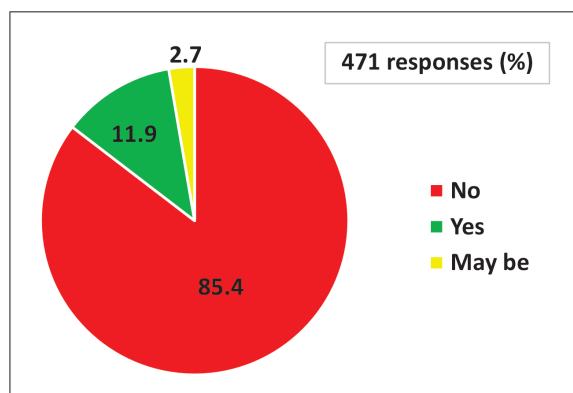


Fig. 2. Do you have sufficient subordinate staff?

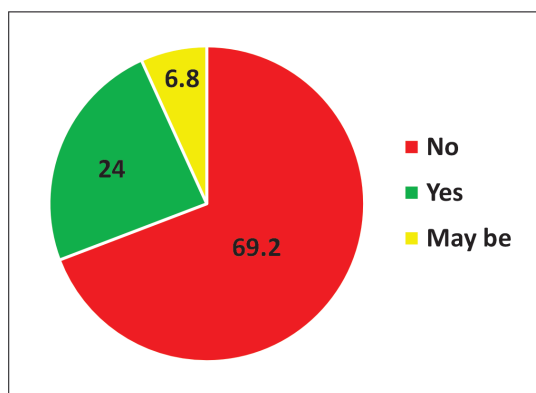


Fig. 3. Are your duties well defined?
Responses 471 (%)

As can be seen from the Pie diagram (Fig. 4), the 45% (n=212) of the respondents feel that their senior officers do not support and motivate them in their day-to-day working, while 21% (n=99) of the respondents were in doubt. Only 34% (n=160) of the respondents feel that their senior officers support and motivate them in day-to-day working.

The Fig. 5 again highlights the poor working conditions of the vets working in the Animal Husbandry sector in India. As per Fig. 5, the most of the senior officers do not encourage subordinate field staff to use modern technology for communication regarding sending reports or other correspondence or feed-back. So, on one side, the vets are carrying their duties (not well defined) with little or no subordinate staff and with very little infrastructure, and on the other side, they are to rush to offices to submit reports or other correspondence etc.

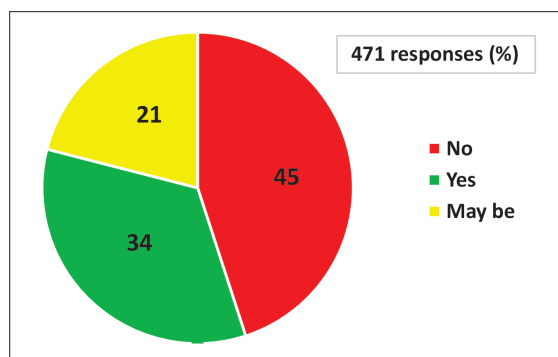


Fig. 4. Does your senior officers support and motivate you in day-to-day working?

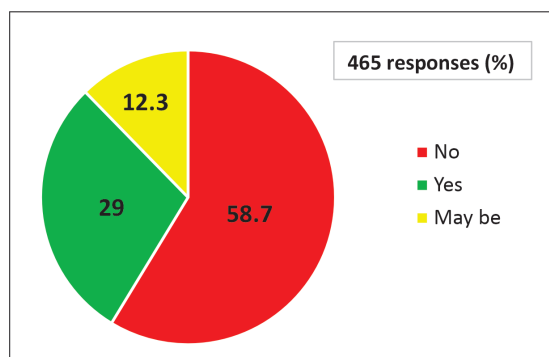


Fig. 5. Does your organization encourages the field staff to use modern technology in day to day communications?

This Fig. 6 also highlights another aspect of the working condition of vets in India. If a person is satisfied in his profile with the prevailing working conditions, he is more likely to recommend his near and dear ones also to go for the same. It is very apt and directly linked to job satisfaction. Here, only 36.7% of the vet-respondents declared that they will recommend their near and dear ones to take up their profile in their present organization.

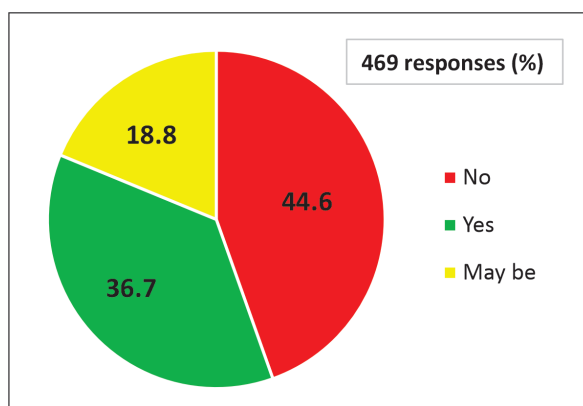


Fig. 6. Will you recommended your near and dear ones to take up your profile in your organization?

Contribution from the study: The findings of this study shall help the concerned and willing employers, legislators to draft new policies with respect to their current working scenario of veterinary employees so that they feel more motivated at their work-place and contribute more to the society and to the nation.

Limitation, and the direction of future research: In addition to Covid-19 situation, it is due to vastness of India also that it was not possible to contact the respondents personally. Therefore, the help of modern technology i.e. Google-form and WhatsApp has been taken in this survey. It might tilt the scales in favour of those vets who are more social and active on WhatsApp and are tech-savvy too. So voice of many other vets who are not-so-active on WhatsApp and not-so--tech-savvy might, if heard, could potentially alter the findings of this survey, either way.

IMPORTANCE OF LIVESTOCK SHOWS UNDER INDIAN CONTEXT

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Abstract

Organizing cultural / scientific melas are our tradition since ages. It has social, economical and scientific importance. Persons associated with livestock rearing especially the youth learn many life skills from such melas for success in their future endeavours. The success stories showcased in such events travel very far and in quick time in terms of great ideas adopted by innovative farmers, animals of highest genetic merit judged through bodily characters and happy bondings among different stakeholders established at the time of close meetings etc. Livestock shows also help in quick dissemination of technical and scientific know-how for genetic improvement, maintaining and improving livestock breeds, nutritional and health related practices and even marketing strategies. The existing as well as would-be livestock rearers get extraordinary encouragement and motivation from these livestock shows. The veterinarians are the key role players to encourage farmers to participate in such shows.

Keywords: Dairy farmer, livestock rearing, livestock show, mela

Events where livestock are exhibited and judged comparatively based on certain breed specific visual characters are known as livestock shows. Animal Husbandry Department of Punjab State organizes such shows regularly every year (Singh et al., 2018). These are usually held in every district of the state. The main aim of these shows is to strengthen various vocations associated with livestock. These shows act as a source of inspiration to budding livestock farmers. At the same time, these are worthy of providing newer experiences and noesis to senior farmers. The young farmers can imbibe such characters as patience, responsibility, self-confidence, team spirit etc. from such shows.

During these livestock shows, competitions are usually organized for buffaloes, cattle (exotic, zebu and crossbred), horses, camel, sheep, goats, pigs, dogs and poultry (chicken, duck, turkey etc.) in various categories. The animals are evaluated on the basis of external features visible to the eye and their behaviour in the ring. The animals topping in the category get suitably awarded.

Livestock farmers, other farmers, animal traders, milkmen, officials of Dairy Development Department, Animal Husbandry Department, Milk Cooperative Society,

other Cooperative Societies, Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana and other entrepreneurs associated with livestock farming participate in such shows. The farmers and general public from other states also take part in such events with great fervour.

Livestock farmers are the major beneficiaries of such events (Robin, 2009). The farmers who visit these shows are able to acquire the knowledge of characteristic features of a high producing buffalo/cow or a breeding bull. These farmers do not face major problems in future whenever they are engaged in purchasing similar animals.

There are certain occasions when a particular person faces a dilemma whether to start a commercial dairy farm or not. The farmers standing in exhibition ring of livestock show alongside their best animals become a motivating sight for such persons. This way, these shows trigger a spark in the mind of hesitant persons for starting a new venture in dairy farming.

The monetary value of animal that has been exhibited in a regional, state or national level livestock show gets increased significantly. Such animals are also responsible for raising the worth of their herdmates. If any animal gets first or second rank, its monetary worth gets skyrocketed automatically.

Whenever a livestock farmer brings his animal in the ring, it is but natural that he compares his own animal with that of others. If somehow, his animal appears to him a little bit weaker in comparison to others, he will automatically make his mind to manage the animal in a better way and bring the animal to the show next time with full preparedness. Better the management better will be the outcome in such shows. Better management will also bring heifers in heat earlier. This way they can produce more milk in their lifetime. The same farmer will, then, start keeping records of even minute things. Thus, livestock shows help the farmers in inculcating habit of record keeping.

The farmers who already possess the knowledge of best characters of a milch animal or who can easily identify the best breeding bull buy such animals from other parts of the country. They bring those animals to these livestock shows for competition. When the animal gets one of the top ranks, the self confidence of those farmers becomes limitless.

These livestock shows are also beneficial to the experienced judges who perform their duty with full honesty and unbiasedness. They are able to judge the animal in different categories with minute details. Thus, they can develop the habit of scrutinizing anything in detail.

Livestock show is a very good place for the budding judges to learn many things. Usually pictures of previously top ranked animals help a lot in comparative evaluation of

animals. Working with experienced judges also helps in acquiring many newer things.

Livestock shows can help in the identification of top ranked animals in various categories. The list of such animals can be helpful in some other way in future. Similarly, list of progressive farmers can also be prepared from the records of such shows.

The officials of the Animal Husbandry Department should keep track of the top ranked animal in each category. This will help in the genetic evaluation of that particular animal. Animals with high genetic make-up can be exploited fully through artificial means. This will make the whole dairy business of the state more viable economically.

Livestock shows also have a few more advantages that can't be analysed so easily. These shows help to develop social bondings among many farmers. The young farmers draw benefits out of these bondings in future. These shows also influence the behaviour of young farmers. These shows help them in developing habits of hard working, responsibility, decision making etc. At the same time, farmers also experience gentle behaviour whenever the animal is unable to fetch top honours. The farmers also learn alternate or newer management methods employed by other progressive farmers. Thus, livestock shows bring newer ideas for the farmers.

The livestock shows are also helpful for different companies associated with animals like those who deal in veterinary medicines, fodder seeds, fodder crop agronomic implements, animal management implements etc. These companies get huge clientele at one spot for advertising their products.

Thus, livestock shows bring something for all categories of people associated with livestock farming. We should encourage all types of farmers to participate in such events.

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