

RAKSHA TECHNICAL REVIEW

Publication of Indian Immunologicals Limited

September 2021

Free Private Circulation

**Epidemiology and Techno-economical
Aspect of Haemorrhagic Septicaemia
in India: A Review**

**Deep Stromal Corneal
Ulcer in a Pug:
Diagnosis and Treatment**

**Surgical Management of
Urinary Bladder Calculus
in a Female Maltese Dog**



INDIAN IMMUNOLOGICALS LIMITED

Reader's Desk



Milford Veterinary Clinic deeply appreciates the sharing of experiences with the field Veterinarians through the valuable portal, Raksha Technical Review. Animal Welfare is the cherished goal of the entire International Veterinary Fraternity. Dr Dale Clark and Dr Sabita Rakshit treat interesting cases in the companion animals, frequently. We value the sharing of the skills and knowledge, and hope that this bond will grow stronger in the days to come. Stay healthy and safe!



Thank you for sending me the copy of RTR Feb 2021 issue. I highly appreciate your concern towards field veterinarians. RTR is really helpful in upgrading oneself about various topics related to veterinary world.

Dr Prasanna Hegde
Mundgod, Uttara Kannada, Karnataka



It is an excellent magazine, with full information. Need more content regarding dairy animals.

Dr Rajesh Gupta
Jaipur, Rajasthan



RTR Journal is very good and informative.

Dr Pradeep Sharma
Krishi Nagar, Jabalpur



All products of Indian Immunologicals are very good and feedback of all products are very good. Good luck for Indian Immunologicals company.

Dr Kiran Khichar
Chittorgarh, Rajasthan

Editorial Board

Priyabrata Pattnaik

Dy Managing Director

Rajendra Lingala

Vice President - R & D

Karnati Srinivas

General Manager - AH QC

Honorary Members

Abdul Samad

Former Director & Dean
MAFSU, Nagpur

S Prathapan

Former Dean, TANUVAS
CVS, Tirunelveli

K Sadasiva Rao

Former Associate Dean,
NTR CVS, Gannavaram

H K Verma

Former Director of Extension
GADVASU, Ludhiana

Sub - Editorial Committee

V Surya Prasad

Sr Manager - Veterinary Services

Z Hasan

Manager - AH PMT

D Prabhakar

Manager – CRM & CSR

Sourced & Published by

S Sobhan Babu

Vice President - Animal Health

Contents

Reader's Desk	02
From the Editor's desk.....	04
Managing Director's Message.....	05

Large Animal Section

Epidemiology and Techno-economical Aspect of Haemorrhagic Septicaemia in India: A Review	06
Pelvimeretry - A Boon to Bovine Breeding	17
Omasal Impaction, Abomasal Impaction and Diffuse Peritonitis Disorders in Bovine	22
Management of Dicephalus, Monostomus, Dibrachius and Dipus monster in a Rathi Cow: A Case Report	25
Repeat Breeding Syndrome in Cows	26
Research Summary on Buffaloes in Marathwada Region: What We Know in Past 40 Years	29

Indigenous Breed

Mithun - An Animal of Indian Pride: A Review	32
Bargur Cattle: An Overview	37

Grazers and Browsers

Assam Hill Goat - An Indigenous Goat Breed of Assam	39
---	----

Companion Animal Section

Clinical Approach to Diagnosis of Ophthalmic Disease in Small Animals.....	40
Deep Stromal Corneal Ulcer in a Pug: Diagnosis and Treatment	44
Successful Surgical Treatment of Interdigital Follicular Cyst in a Dog.....	48
Surgical Management of Urinary Bladder Calculus in a Female Maltese Dog	51
Feline Upper Lip Eosinophilic Ulcer 'Rodent Ulcer': Diagnosis and Treatment.....	55
Nutritional Deficiencies in Guinea Pig.....	58

General Articles

Anti-microbial Resistance Crisis in COVID-19 Pandemic	61
Functional Role of Common Ingredients Used in Preparation of Value - Added Meat Products.....	63
Fluoride Toxicity: A Major Global Problem in Domestic Animals.....	67

Feedback	69
Guidelines to Authors	70



From the Editor's desk

Data is the most reliable tool to assess and take accurate measures in disease control and prevention to nullify the economic loss, especially in the dairy sector. It is necessary to gather information on a disease about the causative agent, historical aspects, epidemiological patterns, geographical distribution, the incidence in a country and around the world. The diagnostic methods are very crucial to identify the organism, pathology and clinical signs. Morbidity and mortality rates guide to proper treatment, control and prevention.

Haemorrhagic Septicaemia is an acute disease, causing a high economic loss in India, occurring due to stress such as transportation, shipping of cattle and other animals. The article *Epidemiology and Techno-economical Aspect of Haemorrhagic Septicaemia in India* have given elaborate information with data on the disease distribution, incidence, pathogenesis, treatment as well as prevention and control measures through available vaccines and medicines.

Certain types of fodder material cause disturbances in cattle in Indian feeding systems. The article *Omasal Impaction, Abomasal Impaction and Diffuse Peritonitis Disorders in Bovine* described those feed materials that impact the digestive system of the cattle in India and their management.

The Large Animal Section also contains articles *Pelvimetry – A boon to Bovine Breeding; Management of Dicephalus, Monostomus, Dibrachius and Dipus Monster in a Rathi Cow: A Case Report; Repeat Breeding Syndrome in Cows; and Research Summary on Buffaloes in Marathwada Region: What We Know in Past 40 Years*; covering several challenges faced in reproductive aspects of dairy cattle.

Indigenous Breed Section contains two interesting reviews on native breeds *Mithun – An Animal of Indian Pride: A Review* and *Bargur Cattle: An Overview*.

Grazers and Browsers Section brings an article *Assam Hill Goat - An Indigenous Goat Breed of Assam*.

Companion Animal Section contains the article *Clinical Approach to Diagnosis of Ophthalmic Disease in Small Animals* refers to ophthalmic issues of canines.

Milford Veterinary Clinic (MVC), USA also shared the clinical observations and surgical procedures on issues related to canine and feline veterinary practice. In the case of canines, MVC presents the articles *Deep Stromal Corneal Ulcer in a Pug Diagnosis and Treatment; Successful Surgical Treatment of Interdigital Follicular Cyst in a Dog*; and *Surgical Management of Urinary Bladder Calculus in a Female Maltese Dog*. In the case of felines, an article dealing with rodent ulcer, *Feline Upper Lip Eosinophilic Ulcer 'Rodent Ulcer' Diagnosis and Treatment*, was presented. The article *Nutritional Deficiencies in Guinea Pig* highlights poor diet and the need for proper supplementation of essential nutrients especially vitamin C.

General Articles Section brings focus on drug resistance during a pandemic in the article *Anti-microbial Resistance Crisis in COVID-19 Pandemic*. And value addition to meat by-products was explained in the article *Functional Role of Common Ingredients Used in Preparation of Value-Added Meat Products*. The article *Fluoride Toxicity: A Major Global Problem in Domestic Animals* points out fluoride toxicity impacts on environmental pollution, adverse effects and health hazards on domestic animals.

Stay Safe!! Stay Healthy!! Stay Vaccinated!!

Dr Priyabrata Pattnaik

Dy Managing Director

Managing Director's Message



Dear Patrons!!

Greetings!!

As we are learning to cope with the after-effects of World-wide lockdown over the past 18 months, it is heartening to note the resilience of the economies across the Globe. There is plenty of hope with the increasing availability of vaccines and vast coverage of vaccination by the respective Governments. It is very important that a significant number of people in our country get vaccinated to achieve herd immunity.

Rising to a request from the Government of India, the team at IIL worked tirelessly, and in a record time, to produce the much-needed COVID19 Drug Substance for supplies to Bharat Biotech. This effort was lauded by many including Shri Parshottam Rupala, Minister of Fisheries, Animal Husbandry and Dairying, Dr V K Paul, Member, NITI Aayog, Dr Renu Swaroop, Secretary, Department of Biotechnology etc.,

IIL will continue to contribute in whatever manner to ensure that we come out of this pandemic.

As we are approaching the end of 2021, we wish you all a very happy, healthy, and prosperous new year 2022.

Warm Regards

Dr K Anand Kumar

Epidemiology and Techno-economical Aspect of Haemorrhagic Septicaemia in India: A Review

Shreya Dubey, M N Brahmabhatta, R V Singh, Bhavana Gupta, Nidhi Sharma, Renuka Patel and Boreddy Manoj Reddy
Department of Veterinary Public Health and Epidemiology, College of Veterinary Science and Animal Husbandry, Anand, Gujarat.

Introduction

Hemorrhagic septicemia is an acute septicaemic disease occurring most often in cattle, buffalo, camel, sheep, goat and pig, usually following some form of stress, such as transporting, shipping and is characterized by high rise of temperature, depression and presence of multiple haemorrhages, pneumonia, pleuritis, pericarditis and arthritis. The clinical signs usually progress rapidly from fever and depression to death, within hours to a few days. Because the disease course is so short, few animals can be treated in time and recovery is rare. It is an important cause of livestock mortality (60%) in tropical regions of Asia, Africa and the Middle East. It also occurs in other animals and has caused severe outbreaks in endangered saiga antelope (*Saiga tatarica*) in Kazakhstan. One outbreak in 2015 killed more than half of saiga population. Genetically, buffaloes are more susceptible to it than cattle and the overall mortality in buffaloes is nearly three times more than in cattle. Young animals are more prone to this than adults (1) and outbreaks are particularly common during rainy weather, when the organism can spread readily causing morbidity and mortality between 50 and 100%. In areas where animals have no immunity, severe disease is expected to occur in all ages. There is a sharp drop in milk yield of the affected animals. It has emerged as a disease of considerable economic importance in India where cattle and buffaloes are large in number and are vital for milk production and draught power. It is the second most reported disease in India during 1991 to 2010 (NADRES-National Animal Diseases Referral Expert System) and is the cause of maximum number of the reported deaths. Around 97 per cent of the H.S. outbreaks reported by NADRES are in large ruminants. *Dutta et. al.*, (1990) (2) studied the epidemiological data of H.S. for a period of 13 years (1974-1986) and reported that H.S. was first in mortality and second in morbidity when compared to other epizootic diseases, namely foot and mouth disease (FMD), rinderpest, anthrax and black quarter. *Singh and Prasad* (2008) (3) have reported an average annual loss of 287.81 lakh due to H.S. in cattle in India based on the number of cases and deaths reported during 1991-2005 by the Department of Animal Husbandry, Dairying and Fisheries, Government of India. This loss might look small due to under-reporting of number of cases and deaths (4). The synonyms of disease are barbone,

haemorrhagic pneumonia, shipping fever, stockyard pneumonia.

Historical Aspect of Pasturellosis

Pasteurellosis of cattle was first described in 1878 by Bollinger in Germany and the causative agent was isolated by Kitt in 1885.

Causative Agent

The disease is caused by *Pasteurella multocida* type-I organism. *Pasteurella* is small, non-motile, facultative anaerobes, gram negative rods or coccobacilli, bipolar organism. The organism may be present in the respiratory tract as commensal and may not be able to produce disease alone. The Asian serotype B:2 and the African serotype E:2 (Carter and Heddleston system), corresponding to the newer 6:B and 6:E classification (Namioka-Carter system), Robert type-I to IV (previous classification) based on mice protection test, are mainly responsible for the disease. Other serotypes, namely A:1, A:3, have been associated with a HS-like condition in cattle and buffaloes in India with mainly pneumonia leading to death.

P. multocida are identified based on the difference in capsular polysaccharides (5) and are designated as A, B, D, E and F. The capsule of type A and B is composed of hyaluronic acid, type D contains heparin and type F contains chondroitin. *Pasteurella multocida* is divided into following subspecies: *P. multocida* subsp. *multocida*, *P. multocida* subsp. *septica* and *P. multocida* subsp. *gallicida* based on fermentation of sorbitol and dulcitol. *P. multocida* subsp. *multocida* strains ferment sorbitol but do not ferment dulcitol; *P. multocida* subsp. *septica* strains exhibit negative reactions for both sorbitol and dulcitol fermentation, whereas the *P. multocida* subsp. *gallicida* strains ferment both sorbitol and dulcitol (6).

Scientific Classification of Bacteria

Phylum	Protobacteria
Class	Gammaproteobacteria
Order	Pasteurellales
Family	Pasteurellaceae
Genus	<i>Pasteurella</i>
Species	<i>P. multocida</i> , <i>P. haemolytica</i>

Survivability of Organism

P. multocida is susceptible to mild heat (55°C). Moist conditions prolong its survival thus disease tends to spread more during the wet season. Rice cultivation during monsoon season enhances movements of animals, work stress in work animals etc, all these favour the precipitation of outbreaks (7). *P. multocida* can survive for hours and possibly days in damp soil but viable organisms are not found in the soil or pastures after 2–3 weeks.

Disinfection

P. multocida is susceptible to many common disinfectants, as well as to mild heat (55°C / 131°F) or UV light. Specific agents reported to be effective against *Pasteurella* sp. include 0.5% phenolic disinfectants, 70% ethanol, 1% sodium hypochlorite, iodophors, formaldehyde, glutaraldehyde and peracetic acid.

Geographical Distribution

The disease was first reported as early as the 1880s in Malaysia. Based on the distribution of the disease, three distinct categories of countries can be identified in the Asian continent (8).

Category A

The disease is reported to be endemic and is of

utmost economic importance in Bhutan, China, India, Indonesia, Mongolia, Myanmar, Philippines, Sri Lanka and Malaysia (except in Sabah region where only a low sporadic or exceptional occurrence). Most of these countries have large cattle / buffalo populations, in respect to the existing infrastructures and the limited operational capabilities of their veterinary services. Furthermore, livestock rearing is largely extensive, and management is rather poor in view of the significance of ruminants in the respective national economies.

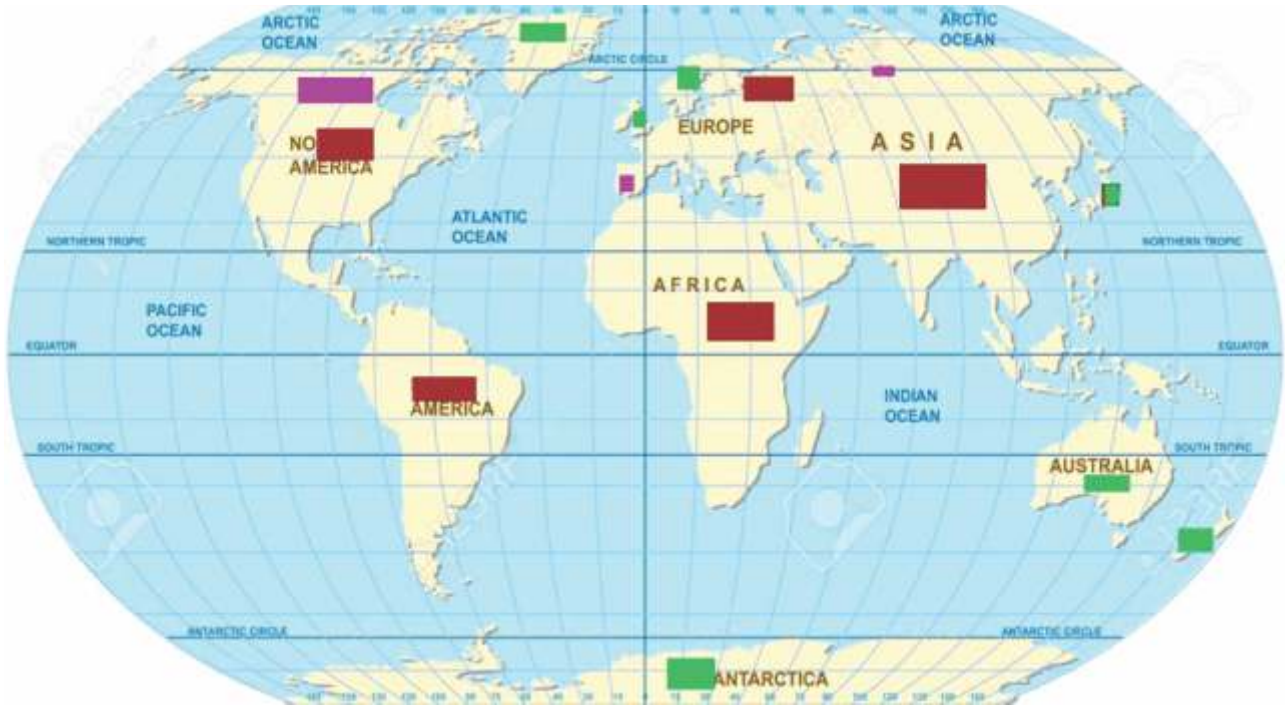
Category B

In this category of countries, although sporadic outbreaks of the disease have been suspected clinically, its presence has not been confirmed by isolation of the agent and serotyping. Example: Kuwait, Qatar. These countries are net livestock importers and therefore, may see the disease introduced occasionally through cattle imports.

Category C

In this group of countries which are spread throughout all the continent, clinical reports of the disease in the past are on record, but presently, these countries are free of the disease (Singapore, Chinese Province of Hong Kong, Israel) or the disease has never been reported (Japan, Jordan).

Disease Endemic	Low Sporadic	Suspected but not Confirmed	Probably Existed Now Free	Never Occurred
ASIA - Bhutan, China, India, Indonesia, Malaysia, Mongolia, Myanmar, Philippines, Sri Lanka	Malaysia (Sabah)	Kuwait, Qatar	Hong Kong (1984), Israel (1948), Singapore (1930)	Cyprus, Japan, Jordan
AFRICA - Central African Republic, Chad, Ivory Coast, Ghana, Guinea, Kenya, Mozambique, Niger, Nigeria, South Africa, Zimbabwe	Angola	Botswana	Algeria, Republic of Congo, Egypt (1970), Eritrea (1993), Mauritius (1989), Morocco, Namibia (1988), Seychelles, Sudan	Cape Verde Libya
AMERICA - Argentina, Brazil (part), Ecuador, Falkland Islands, Honduras, Nicaragua, Venezuela	Canada, Jamaica, U.S.	El Salvador (1990), Paraguay (1985), St Kitts and Nevis	Barbados, Belize, Bolivia, Chile, Cuba, Haiti, Mexico	
EUROPE	Estonia, Latvia, Portugal, Russian Federation, Spain	Albania, Austria, Croatia, Greece	Finland (1993), Germany (1986), Iceland, Italy, Luxembourg, Macedonia, Malta, Poland (1985), Romania (1993), Sweden, Switzerland, Isle of Man, Yugoslavia	Czech Republic, Denmark, France, Iceland Lithuania, Moldova, Siovania, United Kingdom



**Fig. 1 Showing Global Distribution of Disease
Category A Countries – Brown Color, Category B Countries - Pink Color,
Category C Countries – Green Color.**

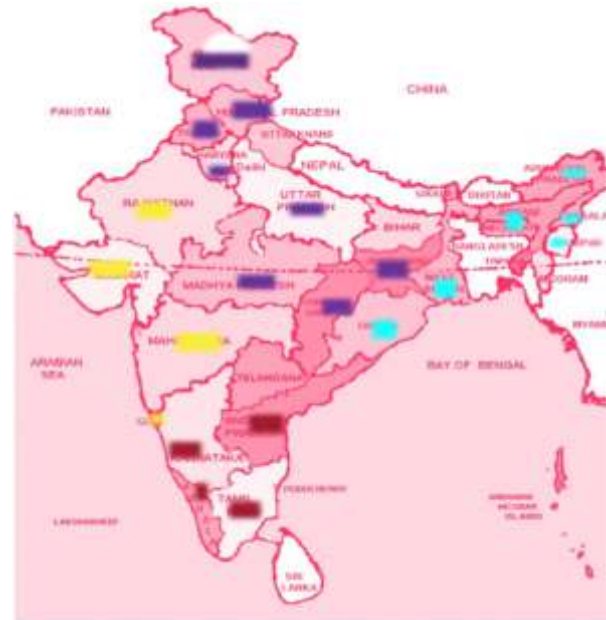
Distribution and Incidence of H.S. in India

In India, each year 53,000 bovine deaths are attributed to pasteurellosis, which during 1974 – 1986 remained the cause of highest mortality and second highest morbidity as compared to foot and mouth disease, rinderpest and black quarter. Seroprevalence of H.S. in Assam was 35.7% (9). In Karnataka, the H.S. outbreaks are regularly reported since 1987 and from 2011 onwards there is slight reduction in the outbreaks. In Andhra Pradesh and Telangana, the outbreaks were more severe from the year 1992 until 2006. However, there was reduction in number of H.S. outbreaks from the year 2007 onwards and there were very few outbreaks in the year 2016. The reduction in number of outbreaks may be due to effective and planned vaccination in the Andhra Pradesh and Telangana.

Incidence of H.S. in India

The highest cases of cattle infection were reported from the eastern region (Arunachal Pradesh, Manipur, Meghalaya, Nagaland, Tripura, West Bengal, Odisha) (43.71%), followed by western (Gujarat, Maharashtra, Rajasthan, Goa, Lakshadweep) (27.06%), southern (Andhra Pradesh, Karnataka, Tamil Nadu, Pondicherry) (22.42%) and northern (Jammu & Kashmir, Himachal Pradesh, Punjab, Haryana, Uttar Pradesh, Madhya Pradesh, Chhattisgarh, Jharkhand) (6.81%) regions. The incidence pattern

of H.S. in buffaloes was different; it was highest in southern region (69.93%), followed by northern (20.97%), western (8.51%) and eastern (0.58%) regions. In terms of the period, the effect of H.S. was observed throughout the year but in terms of



**Fig. 2 Showing Distribution of Disease in India
Eastern Region - Blue , Northern Region -
Purple, Western Region - Yellow ,
Southern Region - Brown.**

season, the H.S. infection (number of cases) in cattle was more during rainy season (40.85%), followed by summer (30.6%) and winter (28.55%) seasons. In the case of buffaloes, the number of H.S. infection was more in the rainy season (64.65%), followed by winter (23.42%) and summer (11.93%) seasons. Average annual morbidity and mortality rates of H.S. during 2007 to 2011 were 12.14 per million and 3.40 per million, respectively in the cattle and 7.83 per million and 3.30 per million, respectively in buffaloes.

Epidemiological Patterns

1. Host of Disease

a) Principal Hosts - Cattle and water buffaloes (*Bubalus bubalis*) are most susceptible species. Young growing cattle within the age group of 6 months to 2 years are most often affected (10), although outbreaks of hemorrhagic septicaemia have been reported in sheep, goats, swine and infrequently in deer, camels, elephants, horses, donkeys and yaks. The horse is comparatively immune. Of laboratory animals, rabbit and mice are highly susceptible and guinea pig and rats are resistant. Latent carriers are those animals in which organisms are lodged in the tonsils. Active carriers are those animals where organisms are detectable in the nasopharynx (7).

b) Reservoir Hosts - Mammals, birds and humans serve as reservoir host.

2. Sources of the Agent

a. Blood Septicaemia - in H.S. occurs at the terminal stage of the disease, therefore, blood samples taken from sick animals before death may not always contain *P. multocida* organisms.

b. Nasal Secretions - It was observed that 7% cattle and 3.5% buffaloes carried this organism into their nose. Organisms are also not consistently present in sick animals.

3. Mode of Infection

a. Direct Contact - *P. multocida* is transmitted by direct contact with infected animals.

b. Indirect Contact - Infection occurs by inhalation or ingestion of *P. multocida* bacteria and sometimes by the bite the arthropod vector.

4. Factor Favours the Transmission

a. Climatic Condition - Higher incidence of Haemorrhagic Septicaemia is associated with moist, humid conditions.

b. Population of Susceptible Animal - High buffalo population density and extensive free grazing system of management, where large herds graze freely in common pastures and are paddocked together at night.

c. Age of Animal - Young animals are more susceptible, this is because in adults there is naturally acquired immunity resulting from the so-called nonfatal infection which largely controls the mortality and morbidity patterns (11).

d. Breed of Animal - Cattle and water buffaloes (*Bubalus bubalis*) are the principal hosts of hemorrhagic septicaemia, and it is widely considered that buffaloes are the more susceptible. A study showed indigenous breeds having higher sero-positivity (33.5%) compared to crossbred animals (18.5%), which can be explained by these mainly being kept in rural areas.

e. Location of Farm Animals - Rural farms (47.4%) were more affected as compared to the urban farms (23.6%).

f. Type of Countries - Countries where occasional sporadic outbreaks occur low in some region the mortality may be very low, unlike endemic countries where regular, seasonal outbreaks occur, where losses and mortality rate may be high.

Pathogenesis

P. multocida infections are endogenous. The organisms, which are normally commensals of the upper respiratory tract, may invade the tissues of immunosuppressed animals. Exogenous transmission can also occur either by direct contact or through aerosols. Several adhesins have been described including fimbriae, surface fibrils and filamentous haemagglutinins. These are important for adhesion and antiphagocytosis activity. Capsule of types A and B is composed of hyaluronic acid these compounds are like those found naturally in the host species, this molecular mimicry limits the development of a strong immune response to the organism. The availability of iron is important for colonization and multiplication of the organism and *P. multocida* has several iron acquisition systems. The major virulence factor of *P. multocida* strains of type D, which produce atrophic rhinitis in pigs, is PMT toxin (*Pasturella multocida* toxin), a cytotoxic protein that stimulates cytoskeletal rearrangements and growth of fibroblasts. In septicaemic pasteurellosis, severe endotoxaemia and disseminated intravascular coagulation cause serious illness which can prove fatal.

The major adhesins of *M. haemolytica* appear to be two outer membrane proteins, OmpA and Lpp1 (12) although the organism also has fimbriae and enzymes such as sialoglycoprotease and neuraminidase which may enhance colonization of the respiratory tract (13). Once established in the lung, the organism produces leukotoxin, which has potent effects on ruminant neutrophils whereas neutrophils of other species are resistant. The specificity is based on the ability of the leuko-toxin to

bind to the transmembrane receptor CD18, a subunit of beta2-integrin. This receptor, when associated with CD11a, is expressed as the heterodimeric glycoprotein lymphocyte-function-associated antigen 1 (LFA 1) which is responsible for the high affinity binding of leukotoxin to ruminant leukocytes (13). Binding initiates a series of intracellular events which impair mitochondrial function and leads to the release of cytochrome c and ultimately cell death (12). The importance of viral infections in predisposing to infection with *M. haemolytica* is well known and recent work has shown the possible mechanism behind this observation. In vitro studies have shown increased expression of LFA-1 on bovine peripheral blood mononuclear cells infected with BHV- 1, thus making them more susceptible to the effects of *M. haemolytica* leukotoxin (14).

5. Zoonotic Potential

Pasteurella species are Hazard Group 2 organisms. Two cases of laboratory acquired infections have been reported, both of which were associated with working with laboratory animals (15). *Pasteurella* species reported to have caused human infections are as follows - *P. aerogenes*, *P. bettyae*, *P. canis*, *P. dagmatis*, *P. multocida subspecies gallicida*, *P. multocida subspecies multocida*, *P. multocida subspecies septica*, *P. pneumotropica*, *P. stomatis*, *P. caballi*, *P. oralis*. However, *P. multocida* is an opportunistic pathogen in humans and precautions should be taken to avoid unnecessary exposure. This organism most often causes soft tissue infections, but it is occasionally involved in other conditions such as osteomyelitis, endocarditis, meningitis, respiratory infections or septicemia, especially in people who are immunocompromised or have other underlying diseases.

6. Diagnosis of Disease

Diagnosis of Haemorrhagic Septicaemia is based on a combination of clinical signs, gross pathological lesions and a consideration of relevant epidemiological parameters and other similar diseases prevalent in the locality.

a. Based on History - There may be a history of exposure to stress arising from transportation or overcrowding.

b. Clinical Signs - Incubation period in subcutaneous infection is around 12-14 hours, in aerosol or oral infection 7 and 2 days, respectively and in naturally exposed animals 46-80 hours after initial contact.

In Cattle and Buffalo - Temperature elevation, loss of appetite, nasal discharge, salivation and laboured breathing, with swellings in the submandibular region spreading to the brisket area and even down to the forelegs are common clinical sign. The clinical

syndrome may broadly be divided into three phases as follows,

Phase 1 is dominated by increased temperature, loss of appetite, general apathy, and depression. If closely monitored, a rise in rectal temperature to 40-41°C is recorded, which lasts throughout the course, dropping to subnormal levels during the terminal phase, a few hours before death.

Phase 2 is dominated by a respiratory syndrome. There will be an increased respiration rate (40-50 / minute), laboured breathing, clear nasal discharge and salivation. Submandibular oedema may also begin to show during this phase. As the disease progresses, the nasal discharge becomes opaque and mucopurulent.

Phase 3 is dominated by recumbency. The respiratory distress becomes more acute, the animal lies down, terminal septicaemia sets in and death follows.

In Sheep and Goat - In lambs and kids, pneumonic pasteurellosis can be acute, characterized by fever, lethargy, poor appetite and sudden death. Sheep and goats that survive the acute stage may recover or become chronically affected showing reduced lung capacity and weight gain efficiency and sporadic deaths may occur.

In Pig - High temperature 104°F to 107°F usually ranges in pig. Dullness, depression, anorexic and copious nasal discharge is seen in pig. Pasteurellosis in pig is usually seen in acute form (16).

Morbidity and Mortality

Khan et. al., (2006) (17), reported that the young stock of both buffaloes and cattle have higher morbidity (21.19%), mortality (95.25%) and case fatality rates as compared to the older ones. The exhaustion of the maternal immunity against H.S. after the 60th day of life and delayed vaccination might be attributed to the higher susceptibility of the young calves (18). Once clinical signs appear, case fatality is nearly 100%. A wide range of mortality rate (5 to 90%) has been reported in different outbreaks in India (19) and Philippines (20).

Clinical Pathology

Shoaib Ashraf et. al., (2009) (21), reported there was no significant difference between haematology of diseased and healthy calves, except total leukocytic count. Endotoxin exposure in cattle produces a consistent and profound leukopenia within 1 hour, followed by a rebound leukocytosis within 24 hours (22). Both neutropenia and lymphopenia are observed, with the neutropenia being more severe (22). Sodium ions (Na⁺) concentration increased during disease in all the animals than the normal (reference value: 130-135

mEq/L), chloride ions (Cl⁻) concentration also showed the same trend as sodium ions (reference value: 95-105 mEq/L). A decrease was noted in the potassium ions (K⁺) concentration during disease condition in all the animals (23).

Haptoglobin (Hp) is α 2-globulin, synthesized in the liver. It is one of acute phase proteins whose serum levels increase in acute infections. Skinner *et. al.*, (1991) (24) evaluated serum Hp in inflammations and introduced Hp as an inflammatory indicator in cattle. In apparently healthy animals (mixed breed and age), serum Hp reference value was 0.35 g/L (25).

Postmortem Lesions

Subcutaneous edema (a gelatinous mass with straw - colored or bloodstained fluid) can usually be found in the submandibular region and neck, sometimes extending to the brisket or legs. Similar swellings may occur in the musculature and petechiae may be detected in the subcutaneous connective tissues. The thoracic and abdominal cavities and pericardial sac often contain blood-tinged, serous to serofibrinous fluid and petechiae are common throughout the internal organs, especially on the serosal surface. Ecchymotic hemorrhages may also be found, particularly on the heart. The lungs are diffusely congested and edematous and may contain hemorrhages. Foam is often found in the airways. The digestive tract of some animals may be hyperemic and congested and the abomasum and intestinal mucosa sometimes contain petechiae and ecchymoses. The brain is rarely examined, but meningitis was found in cattle in one outbreak and a group of experimentally inoculated buffalo calves, which additionally received corticosteroids, developed hemorrhagic encephalitis.

Laboratory Diagnosis

A provisional diagnosis is important since preventive measures to control the spread of the disease are required immediately, without waiting for the necessary laboratory confirmation. At the earliest opportunity, however, appropriate material should be collected and dispatched to the nearest laboratory.

Sample Collection

Heparinized blood, nasal swab and affected tissue from live animals as *P. multocida* is not always found in blood samples before the terminal stage of the disease and is not consistently present in nasal secretions. Blood sample should be collected from tips of ears (from live animal only). Ribs and tip of the ear, affected organs such as lung, liver, trachea, gastrointestinal tract from dead animal.

A. Direct Examination - The organism is stained bipolar when smear of blood or tissue are

stained with giemsa's stain or Leishman's stain.

B. Isolation and Identification - The routine medium for isolation of *Pasteurella multocida* are as follows,

- (a) Ox/ sheep blood agar 5-10% CO₂ at 35-37°C for 16-48 hour.
- (b) Casein, sucrose and yeast agar (CSY agar) with 5% blood.
- (c) CGT medium (clindamycin, gentamicin, potassium tellurite and amphotericin B) with 5% horse-blood agar.
- (d) Mac Conkey agar
- (e) CLED (Cystine-Lactose-Electrolyte-Deficient) agar
- (f) BGA (Brilliant Green Agar)
- (g) Tryptose-tryphone agar containing 0.1% sucrose

It produces large, mucoid, non haemolytic colonies with sweetish odour. It does not grow on MacConkey agar but *Pasteurella haemolytica* produces pinpoint red colonies indicating the tolerance of the bile salts in the medium.

C. Biological Screening - Mouse isolation may prove necessary as samples are often heavily contaminated when they reach the diagnostic laboratory. Type B pasteurellae that cause H.S. are highly virulent for mice, with a 50% lethal dose (LD₅₀) of 1-10 viable organisms. Thus, this method is extremely useful in situations where the material has only a few viable organisms.

D. Animal Inoculation - Rabbit is most susceptible lab animal for these bacteria. Mice and guinea pigs are also used generally for this purpose. Material is inoculated through subcutaneous or intraperitoneal route, animal dies within 24 hours; having haemorrhagic lesion.

E. Detection of Plasma Fibrinogen - Level of plasma fibrinogen are raised, and in parallel body temperature of animal is also increasing. Young cattle with clinical sign of acute respiratory disease and fibrinogen concentration greater than 0.7 g/dl and temperature greater than 104°F are suspected to have pasteurellosis.

F. Rapid Slide Agglutination - This test is used for detection of capsular antigen.

G. Indirect Haemagglutination - This test is used for detection of capsular antigen. An antibody titre of 1:160 or above in an indirect haemagglutination test is indicative of recent exposure to the pathogen.

H. Agar Gel Precipitation Test - It is used for

detection of somatic antigen for identification of *P. multocida* and determination of serotypes. For this purpose, the agar gel was prepared by dissolving 0.9 g of Special Noble agar in 100 ml of 0.01 M phosphate-buffered saline (pH 7.2) containing 0.05% sodium azide. Five milliliters of the melted agar were placed on a slide (25 by 75 mm). Nine wells (3 mm in diameter, 8 mm from center to center) were cut and arranged in a circular pattern, with one well in the center and eight wells in the periphery. Usually, antiserum was placed in the central well and testing antigens were placed in the outer wells. The results were recorded after 72 hours of incubation of the slide at room temperature in a humidified chamber. Known positive and negative sera and known antigens were included as controls.

- I. **ELISA Test** - has recently been developed in Australia, however it fails to differentiate between the Asian (B:2) and African (E:2) types but can be a good test for screening a large number of cultures.
- J. **Acriflavine Flocculation Test** - An acriflavine flocculation test was used to identify capsular type D *P. multocida* because these organisms are the only ones that flocculate and precipitate in 0.1% aqueous solution of neutral acriflavine. Known capsular type A (strain PM 1062), type B (strain 656), type D (strain KOBE 6), and type E (strain Bunia II) *P. multocida* isolates were included as controls (26).
- K. **Hyaluronidase Test** - A staphylococcal hyaluronidase inhibition test was used to identify capsular type A - *P. multocida*. Type A - *P. multocida* organisms manifest a diminution in colony size in the area surrounding staphylococcal streaks and loss of capsular hyaluronic acid, whereas types B, D and E *P. multocida* organisms are not affected.
- L. **Penicilline Sensitivity Test** - Pasteurella species are typically penicillin susceptible and so a zone of inhibition around a 10-U penicillin disc may aid differentiation from other Gram-negative bacilli (27).
- M. **Nucleic Acid Amplification Tests (NAATs) / PCR** - PCR is usually considered to be a good method as it is simple, sensitive and specific. PCR detects ToxA protein, which is produced by toxigenic *P. multocida*. It is an essential virulence factor for progressive atrophic rhinitis, but it is not produced by nontoxigenic *P. multocida* isolates (28).
- N. **Ribotyping** - This technique involves the fingerprinting of genomic DNA restriction fragments that contain all or part of the genes

coding for the 16S and 23S rRNA. This technique is having an epidemiological significance because they enable strain differentiation within serotypes and hence some epidemiological inferences, for investigations extending beyond routine diagnosis.

- O. **Multi-Locus Sequence Typing (MLST)** - More accurate method to define the causative organisms. This technique has been suggested as an alternative to 16S rRNA gene analysis and recommended for improving identification at the species level of members of the Pasteurellaceae.
- P. **Matrix-Assisted Laser Desorption / Ionisation** - Time of Flight (MALDI-TOF) Mass Spectrometry - rapid and powerful tool because of its reproducibility, speed and sensitivity of analysis. The advantage of MALDI-TOF as compared with other identification methods is that the results of the analysis are available within a few hours rather than several days. The speed and the simplicity of sample preparation and result acquisition associated with minimal consumable costs make this method well suited for routine and high-throughput use (29).
- Q. **Biochemical test** - Catalase positive, Oxidase positive, Indole positive, H₂S production positive, Nitrate reduction positive, Ornithine decarboxylase production positive.

Differential Diagnosis

Actinobacillosis, lightning, snakebites, blackleg, rinderpest, anthrax and black quarter.

7. Approaches to treatment, prevention and control

Treatment

P. multocida are resistant to streptomycin, tetracycline, oxacillin and trimethoprim (30), therefore, while treating H.S. case such drugs should be avoided. The infected animal should be placed in a warm, well ventilated stall and all healthy cattle should be segregated from infected animal. The body of the infected individual should be maintained warm by blanketing and collapse from shock anticipated. If this occurs two or five ml. of adrenaline chloride solution (1:1000), should be administered subcutaneously and repeated if necessary.

1. The oldest therapy recommended was intravenous treatment with sulfonamides. Intravenous infusion of sulfadimidine sodium 33.33% at a dosage of 1 ml. per 5 pounds bodyweight (about 2.3 kg) has been practiced. In initial phase of disease intravenous administration of sulfonamides i.e. sulfamethazines @ 150mg/kg for three days or

sulfadimidines @150mg/kg for three days is recommended. Side effects of sulfonamides are crystallization in urinary tract, cutaneous eruption, hypothyroidism and idiosyncratic toxicosis.

2. Arif Zafar *et. al.*, (2009) (23) reported 80% recovery rate when treated H.S. with hypertonic saline solution that is administered once (only on the first day of treatment) in the buffaloes while ceftiofur HCl and ketoprofen were administered after every 12 and 8 hours, respectively, for five consecutive days.
3. Shoaib Ashraf *et. al.*, (2009) (21) reported that combination of florfenicol at the dosage rate of 20 mg/kg body weight I.M. with flunixin meglumine at the dosage rate of 2.2 mg/kg B.W gave better results with survival rate of 90%.
4. Oxytetracycline can be used @5-10 mg/kg for 3 days. Besides these treatments may extend with chloramphenicol @10 mg /kg or ampicillin@10 mg (21).
5. Fluid administration is the corner stone for the management of patients suffering from septicaemia (32). Along with these anti-inflammatory drugs are also given e.g. betamethasone @ 1mg/5kg or dexamethasone @ 1mg/5kg, prednisolone @10mg/kg (should not be given in pregnant animal, may abort) in serious condition. The non-steroidal anti-inflammatory drugs (NSAIDs) such as, aspirin, paracetamol and phenylbutazone, meloxicam, nimesulide, ketoprofen, flunixinine @ 1mg/kg body weight is safe for animals.

Prevention and Control

A. Measures During an Outbreak

Vaccination - Vaccination of H.S. for prophylactic purpose must be carried out preferably two to three months before the season (33). During an outbreak, immediate whole herd vaccination is necessary, irrespective of previous vaccination history (33). Vaccination is a major control measure in the face of a new epidemic.

Inactivated Vaccines

H.S. vaccine prepared from a whole broth culture of *Pasteurella multocida* type B and Mannheimia haemolytica and *P. trehalosi* (208 germs / ml), killed by 0.3% formalin and precipitated by 1% aluminium potassium sulphate. For best results vaccinate animals at least 21 days before the haemorrhagic septicemia season.

Dose Rate - Adults and Calves with 2 ml S/C or as recommended by manufacturer.

Duration of Immunity - Immunity appears in 10 days after vaccination and lasts for 6 to 8 months.

1. Commercial H.S. Inactivated Vaccines - H.S. vaccines contains formalin inactivated cultures of *Pasteurella multocida* (strain P52). The inactivated cultures are further purified and concentrated before adjuvantation with aluminium hydroxide e.g. Raksha-HS vaccine.

Examples of Compound Vaccines of FMD / HS / BQ are as follows

- a) **Raksha Biovac (FMD+HS)** contains FMD inactivated antigens against O, A, and Asia-1 strains and formaldehyde inactivated *Pasteurella multocida* culture mixed together in oil emulsion, dose - 3 ml, midneck, deep i/m, primary vaccination – 4 months, booster dose – 1 month after primary vaccination in primo vaccinates, revaccination – Every 6 months.
- b) **Raksha Triovac (FMD+HS+BQ)** contains FMD inactivated antigens against O, A, Asia-1 and formaldehyde inactivated *Pasteurella multocida* culture, inactivated *Clostridium chauvoei* culture mixed together in oil emulsion, dose - 3 ml, mid-neck, deep i/m, primary vaccination – 4 months, booster dose – 1 month after primary vaccination in primo vaccinates, revaccination – Every 6 months.
- c) **Raksha (HS+BQ)** contains formaldehyde inactivated cultures of *Pasteurella multocida* and *Clostridium chauvoei* adsorbed on aluminium hydroxide gel, dose- 3 ml, mid-neck region, s/c, primary vaccination - 6 months and above, booster dose - 1 month after primary vaccination in primo vaccinates, revaccination – Annually and in adverse climatic conditions like unseasonal rains and cyclones, etc.

Live Attenuated Vaccines - A live vaccine prepared using an avirulent *P. multocida* strain B:3,4 (Fallow deer strain) has been used for control of the disease in cattle and buffaloes over 6 months of age in Myanmar since 1989; it is administered by intranasal aerosol application. The vaccine has been recommended by the Food and Agriculture Organization of the United Nations (FAO) as a safe and potent vaccine for use in Asian countries; however, there is no report of its use in other countries and killed vaccines are the only preparations in use by the countries affected with H.S.; a trial of the vaccine has been completed in Indonesia.

2. Use of Hyperimmune Serum - It is recommended for immediate control of disease during an outbreak.

3. Disease Management and Sanitation - includes (a) avoidance of stress to animal by providing proper shelter, space, feed, water, transport and workload, (b) Proper and quick disposal of dead animals. Sanitary measures also include early detection and

isolation of new cases and their immediate treatment with antibiotics, deep burial of carcasses or incineration and the prevention of movements of animals to disease free areas.

B. Measures in Endemic Countries / Zones

1. Vaccination on a routine prophylactic basis preferably two to three months before the high-risk season (monsoon).
2. Awareness of the disease among farmers backed up by a good disease reporting/disease information system.
3. Segregation of animals from endemic and nonendemic areas to avoid contact with carriers.

C. Measures for Prevention of Spread Across Borders

1. By ensuring that the animals originate from a region where no outbreaks of H.S. have occurred for a minimum period of one year or by holding animals under observation for 2–3 weeks before transport.
2. Quarantine them for the same period upon arrival to destination and by vaccinate all animals at the end of the quarantine period, whenever animals are exported from H.S. endemic countries, even after observing all the above precautions.

8. Technoeconomical Aspect of H.S. in Asia

Asia has 423 million cattle and 145 million buffaloes, which is about 33% of world’s cattle and 95% of the world’s buffaloes populations. Cattle and buffaloes are reared mainly for use as draught animals in the rice fields, but in India and, to a lesser extent, Pakistan, milk production is also important (8). In Asia as a whole, buffaloes contribute 37% of milk production but in India, where the production of milk

is the highest in Asia, nearly 50% of the milk is produced from buffaloes.

Most of the cattle and buffaloes are used as draught animals in the rice fields and rice is the staple diet in many countries. Thus, the high population of buffalo in Asia, the high susceptibility of buffalo to H.S. and the high case fatality, all point to the significance of the economic losses due to the disease (8).

Technoeconomic Importance in India

India has a sizeable cattle and buffalo population of 192 million and 70 million, respectively. From 1936 to 1944, there were an average of 700 reported outbreaks of H.S. and 40,000 deaths per year. During the 1950s, the average reported deaths per year ranged from 30,000 to 60,000. In the 1960s and 1970s this figure dropped to approximately 4000 per year, presumably due to improved control measures (1). *Dutta et al.*, (1990) (2) estimated that in India during the past four decades H.S. has accounted for 46-55% of all bovine deaths. They also reported that from 1974 to 1986, H.S. accounted for 6.3 deaths per year for every 1,00,000-bovine population. This is accounted to 58.8% of the aggregate of bovine deaths due to the five epidemic diseases FMD, rinderpest, black quarter, anthrax and H.S. Before 1939, the corresponding figure ranged from 13.4 to 20.9%. Thus, it appears that H.S. has increased in economic importance in relation to the other four epidemic diseases.

Economic Losses

First Type of Classification - The total economic loss due to H.S. can be divided into two parts (i) direct losses and (ii) indirect losses. Direct economic loss is due to mortality and reduction in milk output due to H.S. The contribution of direct losses to the total economic losses was 80.32%. The mortality

Table - Annual Economic Loss due to H.S. in Few Important Milk Producing Countries of Asia

Country	Economic importance
Pakistan	The annual financial loss due to disease was estimated as PKR 19 million, of which PKR 6.8 million was attributed to H.S.
Nepal	Ranked HS as the second most important infectious disease after rinderpest. <i>Thakuri et al.</i> , (1992) (34) found that parasitic diseases and infectious diseases accounted for 54% and 24% of all cases, respectively and HS was the most frequent infectious disease.
Sri Lanka	15.2 % of buffaloes and 8.1 % of cattle die of HS each year thus estimate of economic loss was 90 million Sri Lankan rupees (SLR), equivalent to US\$6 million at that time (35).
Bangladesh	H.S. is second most importance economic disease
Indonesia	H.S. was ranked fourth, after fascioliasis, trypanosomiasis and FMD

loss contributed 95.69%, whereas milk loss contributed 4.31% to total direct loss.

Indirect economic losses were milk loss due to increased abortions and elongation of calving interval, the value of calves that could have been born, reduction in growth, reduction in work power, cost on animal treatment and the opportunity cost. The contribution of indirect loss to the total economic losses was 19.68 per cent. Among different components of indirect losses, the contribution was highest of reduction in growth (59.56%), followed by opportunity cost (14.57%), treatment cost (11.67%), total milk loss (7.60%), drop in work power (6.07%) and values of the calves (0.53%).

Second type of classification - Based on the age groups of the animals, the economic losses due to HS can be divided into two groups: (i) calves, and (ii) adults. In the case of calves, the maximum loss was due to mortality (78.46%), followed by reduction in growth (15.67%), opportunity cost (3.05%), treatment cost (2.68%), and total value of calves (0.14%). The calves contributed 74.80% to the total economic loss. In the case of adults, the mortality loss contributed 72.13% to total economic loss in adults, followed by total milk loss (19.67%), drop in work power (4.74%), opportunity cost (2.31%) and treatment cost (1.15%). The adults contributed 25.20% to the total economic loss.

Conclusion

Haemorrhagic septicaemia (H.S.) is an acute and highly fatal septicaemic disease accounting for 23% of total losses in terms of morbidity and rest 77% are due to mortality of animals. The total economic loss per infected animal due to H.S. in India has been estimated as ₹ 6816 in case of cattle and ₹ 10901 in buffalo. It is estimated that H.S. causes economic loss of ₹ 5255 crore in India thus it should be considered as an important disease from policy perspective. This involves investment in research on H.S. and implementation of vaccination schedules as preventive measures.

References

1. FAO (1991). Proceedings of the Fourth International Workshop on Haemorrhagic Septicaemia, February 1991, Kandy, Sri Lanka. FAO-APHCA Publication No. 1991/13.
2. Dutta, J., Rathore, B. S., Mullick, S. G., Singh, R. and Sharma, G. C. (1990). Epidemiological studies on occurrence of haemorrhagic septicaemia in India. *Indian Veterinary Journal*. 67(10): 893.
3. Singh, B. and Prasad, S. (2008). Economic evaluation of important cattle diseases in India. *Indian veterinary journal*. 85(11): 1207-1210.
4. Ahuja, V., Rajasekhar, M. and Raju, R. (2008). Animal health for poverty alleviation: A review of key issues for India. Background Paper Prepared for "Livestock Sector Review" of the World Bank.
5. Carter, G. R. (1955). Studies on *Pasteurella multocida*. A hemagglutination test for the identification of serological types. *American journal of veterinary research*. 16(60): 481.
6. Mutters, R., Ihm, P., Pohl, S., Frederiksen, W., Mannheim, W. (1985). Reclassification of the Genus *Pasteurella* Trevisan 1887 on the basis of deoxyribonucleic acid homology, with proposals for the new species *Pasteurella dagmatis*, *Pasteurella canis*, *Pasteurella stomatis*, *Pasteurella anatis* and *Pasteurella langaa*. *International Journal of Systematic Bacteriology*. 35: 309.
7. De Alwis, M. C. L., Wijewardana, T. G., Gomis, A. I. and Vipulasiri, A. A. (1990a). Persistence of the carrier status in haemorrhagic septicaemia (*Pasteurella multocida* serotype 6: B infection) in buffaloes. *Tropical animal health and production*. 22(3): 185.
8. FAO-WHO-OIE (1994). Yearbook 1994.
9. Shome, R., Deka, R. P., Sahay, S., Grace, D. and Lindahl, J. F. (2019). Seroprevalence of hemorrhagic septicemia in dairy cows in Assam, India. *Infection Ecology & Epidemiology*, 9(1): 1604064.
10. Blood, D. C., Rasostits, O. M. and Henderson, J. A. (1983). *Veterinary Medicine*. 6th edn. Balliere-Tindall. p 591.
11. De Alwis, M. C. L. (1990b): Haemorrhagic Septicaemia. ACIAR Monograph No. 57, p. 36.
12. Czuprynski, C. J. (2009). Host response to bovine respiratory pathogens. *Animal health research reviews*. 10(2): 141.
13. Rice, J. A., Carrasco-Medina, L., Hodgins, D. C. and Shewen, P. E. (2007). Mannheimia haemolytica and bovine respiratory disease. *Animal Health Research Reviews*. 8(2): 117.
14. Leite, F., Kuckleburg, C., Atapattu, D., Schultz, R. and Czuprynski, C. J. (2004). BHV-1 infection and inflammatory cytokines amplify the interaction of Mannheimia haemolytica leukotoxin with bovine peripheral blood mononuclear cells in vitro. *Veterinary immunology and immunopathology*. 99(3-4): 193.
15. Pike, R. M. (1976). Laboratory-associated infections: summary and analysis of 3921 cases. *Health Lab Sci*. 13:105.
16. Kumar, H. V., Mahajan, S. and Sharma, S.

- (2007). Concurrent pasteurellosis and classical swine fever in Indian pigs. *J Swine Health Prod.* 15(5): 279.
17. Khan, A. U., Saddique, R., Ahmad and Khan, H. (2006). Serosurveillance of Haemorrhagic septicaemia in cattle and buffaloes in district Malakand, NWFP. *J Agric Biol Sci.* 1: 11.
18. Mahmood, A. K., Sheikh, M. A., Akhtar, S., Nabi, G. and Rashid, H. B. (2007). Duration of maternally derived antibodies against *Pasteurella multocida* in cow calves. *Pak Vet J.* 27: 92.
19. Saini, S. S., Sharma, D. R., Gill, B. S., Kwarts, M. S., Singh, J., Sarma, J. K., Dhillon, S. S. and Ramneek, R. (1991). Re-emergence of haemorrhagic Septicaemia in Punjab. *Indian J. Anim. Sci.* 61: 1178.
20. Molna, H. A., Dela Pena, E., Camer, G. and Padilla, M. A. (1994). An outbreak of haemorrhagic septicaemia among carabaos in Northern Samar. *Philippine J. Vet. Med.* 31: 27.
21. Shoaib Ashraf., Awais Omer., Muhammad Ijaz., Umer Naveed Chaudry. and Muhammad Muddassir Ali. (2009). Efficacy of Florfenicol Against Haemorrhagic Septicaemia in Buffalo Calves, Pakistan *J. Zool. Suppl. Ser. No. 9:* pp. 119.
22. Griel, L. C., Zarkower A., Eberhart R. J. (1975). Clinical and clinical pathological effects of *Escherchia coli* endotoxin in mature cattle. *Can. J. Comp. Med.* 39: 1-6.
23. Arif Zafar, M. G., Muhammad, Z. I. and Riaz, M. (2009). Effects of Hypertonic Saline Solution on Clinical Parameters, Serum Electrolytes and Plasma Volume in the Treatment of Haemorrhagic Septicaemia in Buffaloes. *Pakistan Veterinary Journal.* 30 (2): 95.
24. Skinner, J. G., Brown, R. A. L. and Roberts, L. (1991). Bovine haptoglobin response in clinically defined field conditions. *The Veterinary Record.* 128: 147.
25. Horadagoda, A. P. D., Eckersall, J. C. Hodgson, H. A. G. and Moon, G. M. (1994). Immediate responses in serum TNF alpha and acute phase protein concentrations to infection with *Pasteurella haemolytica* A1 in calves. *Research in Veterinary Science.* 57: 129.
26. Carter, G. R. and Subronto, P. (1973). Identification of type D strains of *Pasteurella multocida* with acriflavine. *Am. J. Vet. Res.* 34: 293.
27. Holmes, B., Pickett, M. J., Hollis, D. G., Pasteurella. In: Murray, P. R., Baron, E. J., Pfaller, M. A., Tenover, F. C., Tenover, R. H. (1999). *Manual of Clinical Microbiology.* 7th ed. Washington DC: American Society for Microbiology. p. 632.
28. Lichtensteiger, C., Steenbergen, Lee, S. R., Polson, D. and Vimr, E. (1997). Direct PCR analysis for toxigenic *Pasteurella multocida*. *Journal of clinical microbiology.* 34: 3035.
29. Barbuddhe, S. B., Maier, T., Schwarz, G., Kostrzewa, M. and Domann, E. (2008). Rapid identification and typing of listeria species by matrix-assisted laser desorption ionization-time of flight mass spectrometry. *Appl Environ Microbiol.* 74: 5402.
30. Wassenaar, T. M. and Silley, P. (2008). Antimicrobial resistance in zoonotic bacteria: lessons learned from hosts specific pathogens. *Anim. Hlth. Res. Rev.* 9: 177.
31. Singh, A. K. (2010). Haemorrhagic septicaemia in cattle, a case of worry to farmers. www.cabdirect.org/abstract.
32. Rivers, E., Nguyen, B., Havstad, S., Ressler, J., Muzzin, A., Knoblich, B., Peterson, E. and Tomlanovich, M. (2001). Early goal-directed therapy in the treatment of severe sepsis and septic shock. *New Engle J Med.* 345: 1368.
33. Benkirane, A. and De Alwis, M. C. L. (2002). Haemorrhagic septicaemia, its significance, prevention and control in Asia. *Vet. Med. Czech.* 47: 234.
34. Thakuri, K. C., Mahato, S. N. and Thakur, R. P. (1992). Diseases of cattle and buffaloes in the Koshi hills of Nepal. A retrospective study. *Veterinary Review (Kathmandu).* 7(2): 41.
35. De Alwis, M. C. L. and Vipulasiri, A. A. (1980). An epizootiological study of haemorrhagic septicaemia in buffaloes and cattle in Sri Lanka. *Ceylon Veterinary Journal.* 28(1/4): 24.

Pelvimetry - A Boon to Bovine Breeding

Kalyaan U S, R Rajalakshmi, S Rangasamy, R Gna Devi and T A Kannan
Livestock Farm Complex, Dept. of Veterinary Anatomy, VC and RI (Udumalpet) and Dept. of VGO,
MVC and Education cell, TANUVAS, Chennai, Tamil Nadu.

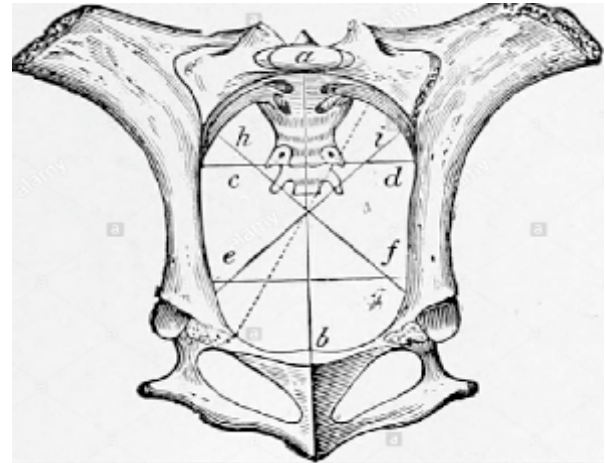
Introduction

Difficulty in calving or dystocia in pregnant cows leads to a major economic loss to cattle owners due to various reasons viz., delay in next estrus, decreased conception rate. In some cases, it may even end up in loss of either calf or cow if left unattended. It also lowers calf weaning weight and market value (1). Common factors associated with calving difficulty in cattle are large sized fetus, small pelvic size of the dam, prolonged gestation, heavy birth weight, frame score and genetics of sire used for breeding, too thin or too fat dam and abnormal fetal presentation at calving. In bovine breeding, the major dystocic determinant is, the fetomaternal disproportion (relation between the calf size/weight at birth and the cow's birth canal/pelvic area) (2, 3). In such circumstances, pelvimetry serves as an excellent tool in predicting dystocia, thereby increasing the survivability of the calf by directly opting for C-section and also it is an useful tool for selection during purchase of cows.

Pelvimetry is defined as the measurement of dimension of the bony pelvis, for assessment of pelvic area, to determine whether the animal can give birth normally or will require a caesarean section. In human obstetrics, pelvimetry has been a routine part of the prenatal management. In veterinary obstetrics pelvimetry is nowadays being commonly carried out to reduce the incidence of dystocia (4).

Fig.1 Illustrates the Pelvic Inlet of Bovine Pelvis with Measurements Needed for Pelvimetry.

Fig.1 - Inlet of Bovine Pelvis



ab: Sacro-pubic diameter;

ed: Superior bisiliac diameter;

ef: Inferior bisiliac diameter;

ei, fh: Sacro-iliac diameters(oblique)

(Image courtesy: Dalrymple, 1907) (5)

Types of Pelvimetry

I. External Pelvimetry: In this method, pelvic area of the animal is determined indirectly by measuring the distance between the selected external points on the animal where measurements are taken using a sliding / hinged pelvic calliper (Fig.2). But is not so appropriate when compared to that obtained via internal pelvimetry.

Pelvic Area Calculation via External Pelvimetry Method:

S. No.	Measurements	Formula
1.	Transverse diameter of pelvic outlet	$\frac{1}{4} (A+B)$
2.	Vertical diameter of pelvic outlet	$\frac{3}{4} \times C$
3.	Transverse diameter of pelvic inlet	$12.2/10 \times (\text{Transverse diameter of pelvic outlet})$
4.	Vertical diameter of pelvic inlet	$13/10 \times (\text{vertical diameter of pelvic outlet})$

Note: Distance between the external angle of the ilium (cm): A; Distance between the ischial tuberosity (cm): B; and Distance between the summit of the croup and the hip joint (cm): C.

Pelvic area: Transverse diameter of pelvic inlet x Vertical diameter of pelvic inlet(cm²)
(Dhaliwal et. al., 1981) (6)

Fig.2: Graduated Sliding Pelvic Caliper



Designed by:
Dr S Balasubramanian, Dr D Kathiresan, Dr S A Asokan,
Department of Veterinary Gynaecology and Obstetrics,
Madras Veterinary College, Chennai, Tamil Nadu.

ii. **Internal Pelvimetry:** In this method, pelvic dimensions of the animal are obtained directly via per-vaginal or per-rectal measurements where the measurements are taken using Rice pelvimeter (Fig.3) or Krautmann pelvimeter (Fig.4) and (Fig.5).

Fig. 3 Rice Pelvimeter

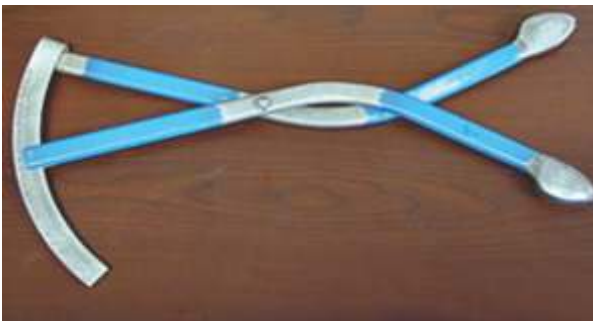


Fig. 4 Krautmann Pelvimeter



Fig. 5 Height and Width of Internal Pelvic Opening



(Image Courtesy: Drost, 1975) (7)

Importance of Pelvimetry in Bovine Breeding

- **Heritability**

The heritability of pelvic area is estimated to be moderate to high ranging from 40-60% indicating the fact that this trait will respond rapidly to selection (2, 8, 9).

- **Pelvic Area and Body Weight**

There is a highly significant positive correlation between the heifer body weight and pelvic area at the prebreeding period, precalving and at three stages of gestation. As a result larger dams with greater body weight possess greater pelvic dimensions or larger pelvic areas. In some cases, two heifers of equal weights can have considerably different pelvic areas. So, body weight is not always a good indicator. Hence selecting heifers for large pelvic area, rather than by body weight, would be highly advantageous (10, 11, 8, 12).

- **Skeletal Size and Pelvic Area**

Due to breed variation, the ratio of pelvic area to wither height is considered to be a more equitable measurement for comparing the pelvic area between herds. Significant differences in this ratio between herds and within breed proves that pelvic size is more influenced by weight gain than by skeletal size (withers height). But in buffalo heifers, maturity in terms of pelvic dimensions appears earlier than sexual maturity, so in an optimally developed buffalo heifer, dystocia due to inadequate pelvic size is not so common (13, 14).

- **Heifer Culling and Pelvic Area**

Selection of heifers based on pelvimetry helps alleviate the dystocia rate for two reasons. First, the correlation of calving difficulty with small pelvic areas, where heifers with smaller pelvic areas have substantially more dystocia problems than heifers with larger pelvic areas (2, 12). Secondly, pelvic area growth is linear in young heifers and measurements taken even as early as pre-breeding can be used to accurately predict the animal's pelvic area at calving.

If pelvic measurements are obtained before

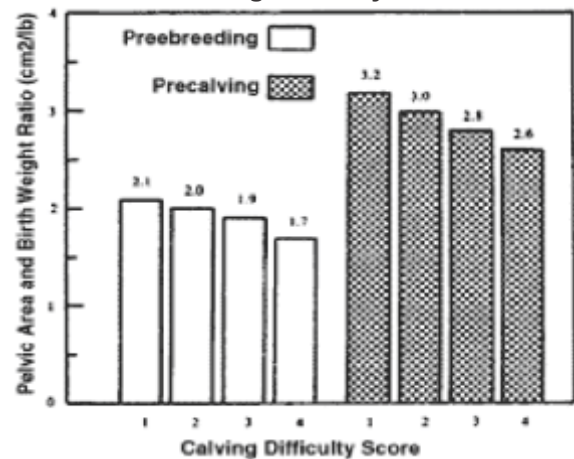
breeding, heifers with a small pelvic size can be culled from the herd. The pelvic measurements should be taken two to three weeks before the breeding season and can be incorporated into a total heifer management programme. This programme helps that, a high percentage of the heifers are cycling and could become pregnant early in the breeding season thereby reducing the incidence of dystocia in bovine. It also aids in estrous synchronization and artificial insemination by determining the percentage of cycling heifers. If heifers are measured at the time of pregnancy examination, heifers with narrow pelvis could be culled/sold (2, 15).

• **Pelvic area and calf birth weight relationship**

Research shows that calf birth weight in relation to the cow's pelvic area determines the degree of calving difficulty. Heifers with ratios of 2:1 or greater before breeding had little or no calving difficulty, while heifers with ratios of 1:9 or less required substantial assistance using a calf puller (Fig.6). These ratios are useful in predicting which heifers may require assistance delivering a certain size calf.

Pelvic measurements obtained on a heifer before breeding and the pelvic area can be divided by a

Fig 6 Pelvic Area and Calf Birth Weight Ratios in Relation to Calving Difficulty



Scores: 1 - No Assistance; 2 - Slight Assistance; 3 - Moderate Assistance; 4 - Major Assistance / C-section. (Image courtesy: *Deutscher, 1987*) (2)

ratio (factor) of 2.1 to estimate the calf birth weight the heifer can deliver without having substantial difficulty. Pelvic measurements can be obtained at the time of pregnancy examination also but the ratio (factor) of 2.7 should be used (Table 2).

Time of Measurement	Heifer Age (months)	Heifer Weight (lb)	Pelvic Area (cm²)	Pelvic Area / Birth Weight Ratio	Estimated Calf Birth Weight (lb)
Before Breeding	12-13	600	z	2.1	67
			160	2.1	76
			180	2.1	86
Pregnancy Examination	18-19	800	180	2.7	67
			200	2.7	74
			220	2.7	82

If the heifers vary considerably in weight at the time of obtaining the measurements, different ratios should be used for various weights and ages of heifers (Table 3). These ratios appear to be good indicators of dystocia, with an accuracy of about 80% (16, 17, 2).

Heifer Weight (lb)	Age at Measurement (months)			
	8-9	12-13	18-19	22-23
500	1.7	2.0	-	-
600	1.8	2.1	-	-
700	1.9	2.2	2.6	-
800	-	2.3	2.7	3.1
900	-	2.4	2.8	3.2
1000	-	2.5	2.9	3.3
1100	-	-	-	3.4

Breeding Strategy and Heifer Replacement

A group of replacement heifer can be divided into three groups based on pelvimetry results as, heifers with large enough pelvic areas bred to herd's average-calving bulls; heifers with too small pelvic area to be bred to the regular herd bull and heifers with very small pelvic diameters that should be culled from the herd. Alternatively, heifers with minimum pelvic area can be picked up for breeding with a bull selected for low birth weights. This practice aids in reduction of calving difficulty at herd level (1).

- ii. Difficulties faced in animal in restraint of some animals for measuring the pelvic dimensions.
- iii. Physical factors like dung in the rectum, rectal straining, and fatigue of the veterinarian on continuous measuring of increased number of animals (19).
- iv. Imprecise structural landmarks for pelvic measurements suggest that pelvic dimensions can only be measured with moderate accuracy (20).
- v. Pelvic measurements are always inaccurate because significant differences in mean

Bull Age (Months)	Bull Weight (lb)	Average Pelvic Area (cm ²)
12-15	900-1200	150-170
23-25	1300-1500	190-210

Bull Pelvic Measurements

Pelvic size can be transmitted from the sire to the resulting progeny. This indicates that selection for large pelvic size in bulls will result in increased pelvic size of daughter offspring (2). Pelvic area in bulls is significantly influenced by the age and weight of bulls (Table 1).

Other traits having positive correlations with the bull's pelvic area are hip height, frame score and scrotal circumference whereas body weight exhibits a poor correlation to pelvic area. Hence all the above criteria must be collectively included in the sire selection for optimum pelvic size of daughter progeny. The best time to measure bulls is when they are yearlings, or at the end of their performance feeding test. The measurements can be obtained by a veterinarian in combination with the breeding soundness exam (fertility evaluation) (1).

Relation Between External and Internal Pelvic Measures

Pelvic angles and slope of rump measurements are generally not related to calving difficulty or internal pelvic area, whereas external pelvimetry together with age of the cow yielded reliable information about internal pelvic variables, that could in turn prove useful in cases of dystocia. This can be confirmed through CT (computed tomography) pelvimetry. External pelvic measures are also well correlated with the internal pelvic measures and serve as good indicators of pelvimetry (11, 18).

Failure of Pelvimetry in Predicting Dystocia

- i. Difference in technical knowledge among the veterinarians.

prebreeding pelvic area between heifers with and those without dystocia were no longer significant at calving (19).

Conclusion

Pelvimetry aids in timely culling and replacement of heifers from the herd at prebreeding and precalving period thereby reducing the incidence of dystocia at herd level. It additionally helps in sire selection too, so as to produce heifer calves with suitable pelvic area or to increase the pelvic area of heifer offspring. Since, pelvimetry is an easy test to perform, with moderate accuracy, it can be applied as a mandatory protocol in bovine breeding for heifer and sire selection and as a preliminary obstetric approach in the handling of any dystocic case, thereby ruling out one of the major maternal cause (fetomaternal disproportion) of dystocia.

References

1. Anderson, P. (2012). Cow/Calf Management: Minimizing calving difficulty in beef cattle. University of Minnesota Extension. Americans with Disabilities Act, 1-10.
2. Basarab, J. A., Rutter, L. M. and Day, P. A. (1993). The efficacy of predicting dystocia in yearling beef heifers: I. Using ratios of pelvic area to birth weight or pelvic area to heifer weight. Journal of animal science. 71(6): 1359.
3. Bellows, R. A., Gibson, R. B., Anderson, D. C. and Short, R. E. (1971). Precalving body size and pelvic area relationships in Hereford heifers. Journal of animal science. 33(2): 455.
4. Dalrymple, W. H. (1904). Veterinary Obstetrics: A Compendium for the Use of

- Students and Practitioners. WR Jenkins. pp 7.
5. Dhaliwal, A. S., Dugwekar, Y. G. and Sharma, R. D. (1981). In vivo pelvimetry in buffaloes (*bos bubalus*). *Theriogenology*. 15(5): 501.
 6. Daly, Russell F. and Riese, Richard L. (1992) "Pelvic Measurements: Applications in Beef Cattle Practice Today," *Iowa State University Veterinarian*: Vol. 54 : Iss. 1 , Article 14.
 7. Deutscher, G. H. (1987). G87-895 Pelvic Measurements for Reducing Calving Difficulty. Historical Materials from University of Nebraska-Lincoln Extension, 325.
 8. Drost project, (1975). Visual Guides of Animal Reproduction (VisGAR).
 9. Holm, D. E., Webb, E. C. and Thompson, P. N. (2014). A new application of pelvis area data as culling tool to aid in the management of dystocia in heifers. *Journal of Animal Science*. 92(5): 2296.
 10. Johnson, S. K., Deutscher, G. H. and Parkhurst, A. (1988). Relationships of pelvic structure, body measurements, pelvic area and calving difficulty. *Journal of Animal Science*. 66(5): 1081.
 11. Laster, D. B. (1974). Factors affecting pelvic size and dystocia in beef cattle. *Journal of Animal Science*. 38(3): 496.
 12. Meijering, A. (1984). Dystocia and stillbirth in cattle—A review of causes, relations and implications. *Livestock Production Science*. 11(2): 143.
 13. Morrison, D. G., Williamson, W. D. and Humes, P. E. (1986). Estimates of heritabilities and correlations of traits associated with pelvic area in beef cattle. *Journal of animal science*. 63(2): 432.
 14. Noakes, D. E., Parkinson, T. J. and England, G. C. W. (2001). *Arthur's Veterinary Reproduction and Obstetrics*. 8th ed. London. Saunders. Elsevier. pp 205.
 15. Price, T. D. and Wiltbank, J. N. (1978). Predicting dystocia in heifers. *Theriogenology*. 9(3): 221.
 16. Ramin, A. G., Daniel, R. C. W., Fenwick, D. C. and Verrall, R. G. (1995). Pelvic parameters, growth rate, puberty, and their interrelationships in young dairy heifers. *Reproduction in Domestic Animals*. 30(3): 117.
 17. Singh, G., Singh, G. B., Sharma, S. S. and Sharma, R. D. (1984). Body size and pelvic area relationships at puberty and two-month-pregnancy in buffalo-heifers. *Theriogenology*. 22(5): 563.
 18. Sloss and Dufty (1980). *Handbook of Bovine Obstetrics*. The Williams and Wilkins Company, Baltimore, USA. pp 155.
 19. Tsousis, G., Heun, C., Becker, M. and Bollwein, H. (2010). Application of computed tomography for the evaluation of obstetrically relevant pelvic parameters in German Holstein-Friesian cows. *Theriogenology*. 73(3): 309.
 20. Van Donkersgoed, J., Ribble, C. S., Booker, C. W., McCartney, D. and Janzen, E. D. (1993). The predictive value of pelvimetry in beef cattle. *Canadian Journal of Veterinary Research*. 57(3): 170.

Omasal Impaction, Abomasal Impaction and Diffuse Peritonitis Disorders in Bovine

Mithilesh Kumar

Dept. of Surgery and Radiology, Bihar Veterinary College, Patna, Bihar.

Introduction

In cattle and buffalo digestive system, omasal impaction, abomasal impaction and diffuse peritonitis are found due to feeding practices. Fodder scarcity in the dry months also lead to various diseases in digestive tract of ruminants in India. Chopped wheat straw is also known as "Bhusa" fed to the animals. This fodder settles in between the omasal leaves and causes omasal impaction by interfering with movements of the organ.

Case History

The animal had vague history of complete anorexia (n=9) or partial anorexia (n=6) suffering from reticulophrenic adhesions (n=15). The anorexia might be a result of altered reticular motility due to perireticular adhesions. All the animals, suffering from omasal impaction, diffuse peritonitis and many organ disorders, showed complete anorexia.

All were fed on wheat straw, green grass and grain in varying concentrations. Interestingly, all the animals suffering from omasal impaction were fed with machine made wheat straw which is very fine variety of wheat straw. It can be hypothesized that because of fine texture, the feed might have got entrapped in between the omasal leaves leading to omasal impaction which might have reduced the gastrointestinal tract motility with increased omasal pressure resulting from omasal impaction. Earlier studies have also implicated the finely cut wheat straw as a possible cause of omasal impaction (1, 2). Sudden change in diet was not reported in animals of any group.

Animal showed either scanty defecation or complete cessation of feces which may be due to involvement of the forestomach, perireticular adhesions and reduced GIT motility due to diffuse peritonitis.

Regurgitation was observed in very few animals suffering from various reticular disorders. However, regurgitation was recorded. This could be attributed to inability of ingesta to pass further down the digestive tract due to reticular/omasal malfunction and involvement of vagus (3). This may also be associated with disfunction of low threshold receptors and fibrosis of wall of reticulum with no involvement of vagus (4). Animals were in either lactation, recently parturated or were in advanced

pregnancy. The pregnancy status however could not be related to the disease in any of these groups. Pain was not recorded in majority of the animals. However, some amount of pain evidenced by flinching on palpation, looking at the flank, frequently getting up and getting down was observed in animals suffering from many functional disorders. Pain observed in these animals may due to the presence of metallic foreign bodies.

Suspended rumination was observed in majority of animals suffering from many organ disorders. Animals showed hypermotile rumen. Recurrent tympany was observed in 7 animals from many functional disorders, while it was absent in 4 animals. The rumen pH was slightly acidic. This may be attributed to abomasal reflux associated with the forestomach disorders. This was consistent to findings of Toor (2) in cases of reticular abscess and omasal impaction. The protozoa was absent in majority of animals suffering from many functional disorders. This may be due to stagnation of GIT contents and indiscriminate antibiotic therapy prior to presentation of case. The animals were in general alert and had wet muzzle. However, three animals which showed dry muzzle and had moderate degree of dehydration suffered from omasal and abomasal impaction. The rectal temperature, heart rate and respiration rate at time of presentation were within normal physiological limit. This was in accordance to the earlier finding of (5, 1, 2).

Result and Discussion

Elevated TLC, with neutrophilia and consequent lymphopenia were recorded in the blood parameter. These findings (6, 7) suggested that the increase in TLC and neutrophilia associated with lymphopenia might be in response to stress. In these groups the haemoglobin and packed cell volume were within normal range.

In animals, total plasma protein was observed which may be attributed to increase in globulin fractions due to inflammatory process in the body (8), dehydration and infection. In general, the total plasma protein was elevated in animals which may be fair indicator of the disease process along with dehydration. Hypalbuminaemia was recorded. This may be secondary to sequestration of protein into extravascular i.e. hepatic failure, malnutrition, malabsorption, maldigestion and inflammatory

response. The plasma fibrinogen remained within normal range.

The animals showed normal to slightly elevated plasma calcium levels. This was consistent to findings of (9) who reported no change in plasma calcium levels in various digestive disorders. The plasma sodium values were recorded to be lower than normal. These findings were consistent to those recorded by earlier workers (1, 2). The plasma potassium values were within normal range. This was in contrast to findings of earlier worker who reported lower potassium levels in these disorders (1, 10, 11, 2). The plasma chloride levels were recorded to be normal. The rumen chloride was recorded to be higher than normal reported values which may give a fair indication of the abomasal reflux. However, rumen chloride did not appear diagnostic in any of the combination. In contrast, (5, 1, 2) reported reduced chloride values in forestomach disorders in bovines.

Radiograph

Survey radiographs were not helpful in diagnosis. However metallic foreign bodies in the reticulum could be visualised on radiographs.

Ultrasound

The reticulum was scanned at 7th intercostal space starting from the level of right elbow slowly scanning down to the ventral midline with crescent shaped 3-5 MHz transducer. The reticular wall was evident as a typical crescent shaped structure having a smooth contour (12, 13, 14) or undulating wall. However, the biphasic reticular contractions were not seen, and the reticular motility was recorded as nil all the animals. At times, the reticular wall appeared to move along with each respiration



Accumulation of fluid with fibrinous adhesions: Diffuse Peritonitis (Ultrasonography)

thereby giving a fair indication of reticulophrenic adhesions. The reticular wall in real time B-mode appeared to slide along the skin and muscle layers and at no time the reticulum contracted away from the transducers. These features strongly suggested reticulophrenic adhesions which was confirmed upon exploratory laparo-rumenotomy. Similar ultrasonographic features have been reported by Braun et al (12, 15) who stated that in cases of traumatic reticuloperitonitis the frequency.

Diffuse peritonitis was visible ultrasonographically as accumulation of fluid with or without fibrinous adhesions covering the entire peri-reticular area and at times involving the whole peritoneal cavity. Depending on fibrin or cell contents of the fluid the echotexture was anechoic to hypo echogenic. Sometimes bands of fibrin were seen within the peritoneal effusion. Similar findings have been reported in cases of peritoneal effusions and diffuse peritonitis by (6, 8). Ultrasound guided abdominocentesis was done in all the animals and diffuse peritonitis was confirmed. Ultrasound was found to be reliable in diagnosis of diffuse peritonitis.

Surgical Management

The intra-operative finding in animals suffering from many functional disorders/multi-organ involvement (n=11), exploratory laparotomy revealed rumen adhesions in 4 cases. The rumen contents were semi-solid (n=4) or impacted (n=4), in general, which suggested generalised atony. However, in three animals the rumen contents were watery. Reticular adhesions were recorded in nine animals which confirmed the ultrasonographic findings of reticulophrenic adhesion in eight cases. Potentially penetrating or non-penetrating metallic foreign bodies were recorded in six animals. The increase in peritoneal fluid was recorded upon laparotomy in eight animals. This again confirmed the ultrasonographic findings of increased peritoneal fluid recorded prior to laparo-rumenotomy. The omasum was impacted in two cases while in one case abomasum was impacted. In one animal both omasum and abomasum were impacted while in another animal omasum and abomasum were distended but not impacted which suggested generalised atony and gastrointestinal stasis.

In animal suffering from many functional disorders the individual disorders like omasal impaction / abomasal impaction / rumen impaction were corrected upon laparo-rumenotomy. In case of rumen impaction, the rumen was evacuated and filled with one third of water before closing the rumen. The cases of omasal impaction were treated as described above. The cases of abomasum distention / impaction were treated by passing small diameter tube through reticulo-omasal orifice into

the abomasums and flushing with high pressure tap water. After abomasums reduced to normal size, two litres of mineral oil were passed into the abomasums through same pipe. Majority of animals were relieved by surgical treatment and were discharged after two day follow up.

References

1. Brar, G. S. (2004). Clinical studies on surgically treated reticulo-diaphragmatic and omasal disorders in bovines. M.V.Sc. Thesis, Punjab Agricultural University, Ludhiana, India.
2. Toor, A. S. (2003). Clinical studies on the surgically treated ruminoreticular, omasal and abomasal disorders in bovines. M.V.Sc. Thesis, Punjab Agricultural University, Ludhiana, India.
3. Leek, B. F. (1969). Reticulo-ruminal function and dysfunction. Vet Rec. 84: 238.
4. Behl, S. M. and Krishnamurthy, D. (1996). Clinical and experimental studies on reticulo-omasal and pyloric functional disorders in buffaloes. Thesis abstract A review. Indian J Vet Surg. 17: 134.
5. Behl, S. M., Singh, J. and Krishnamurthy, D. (1997). Functional stomach disorders (vagus indigestion) in buffaloes. A clinical investigation. Indian J Vet Surg. 18: 73.
6. Blood, D. C., Radostitis, D. M., Arundel, J. H. and Gay, G. C. (1989). Diseases of the alimentary tract. Special examination in: Veterinary Medicine. 7th edn. Pp. 160. Bailliere and Tindal, London.
7. Coles, E. H. (1974). Veterinary Clinical Pathology. 2nd edn. Pp. 426. W. B. Saunders

Company, Philadelphia, London.

8. Fubini, S. L., Normand, G., Ducharme, Hollis, N., Erb, N. H., Donald, F. S., Williams, C. and Rehbum. (1989). Failure of omasal transport attributable to perireticular abscess formation in cattle, 29 cases (1980-1986). J Am Vet Med Assoc. 194: 811.
9. Avery, T. B., Nagraja, T. G. and Frey, R. A. (1986). Blood, urine and ruminal fluid changes associated with metabolic alkalosis induced by duodenal obstruction. Am J Vet Res. 47: 890.
10. Sobti, V. K., Ramkumar, V. and Kohli, R. N. (1982). Certain biochemical studies on diaphragmatic hernia in buffaloes-effects of herniation, laparo-rumenotomy and herniorrhaphy. Indian Vet J. 59: 358.
11. Tagra, S. K., Sharma, D. K., Singh, J., Krishnamurthy, D., Behl, S. M. and Gupta, S. L. (2001). Correction of critical electrolyte deficit in cases of diaphragmatic hernia in buffaloes with abomasal reflex. India J Vet Surg. 22: 92.
12. Braun, U. (2003). Ultrasonography in gastrointestinal disease in cattle. Veterinary Journal. 166: 33.
13. Braun, U. and Gotz, M. (1994). Ultrasonography of the reticulum in cows. Am J Vet Res. 55: 325.
14. Braun, U., Gotz, M. and Marmier, O. (1993b). Ultrasonographic findings in cows with traumatic reticuloperitonitis. Vet Rec. 133: 416.
15. Braun, U., Fluckiger, M. and Gotz, M. (1994). Comparison of ultrasonographic and radiographic findings in cows with traumatic reticuloperitonitis. Vet Rec. 135: 470.

Choose

Gousac[®]

Power

Rumen derived Lyophilized Probiotic Culture



Composition:
Each gram of Gousac[®] Power* contains:

<i>Saccharomyces cerevisiae</i> var. <i>bouardii</i>	: 3.0 Billion CFU
<i>Bacillus subtilis</i>	: 1.5 Billion CFU
<i>Bacillus licheniformis</i>	: 1.5 Billion CFU
Excipients	: q.s.

*Prebiotics added
*All the strains used in the formulation are lyophilized

Dosage:
Daily Supplementation:

5-10 grams per animal mix in feed or
10 grams per 100 litres of water.
Mix 250 grams in every 1000 Kg cattle feed

Available in:
1 kg pack

Management of Dicephalus, Monostomus, Dibrachius and Dipus Monster in a Rathi Cow: A Case Report

Sandeep Dholpuria, Tapender Kumar, Shailendra Mahala, Tirlock Gocher and Rajender Mehra
Department of Veterinary Obstetrics and Gynaecology, College of Veterinary and Animal Science,
RAJUVAS, Bikaner, Rajasthan.

Introduction

The duplication of cranial part of the fetus is more common than the caudal parts (1). During early embryonic development, certain factors such as physical, chemical and viral infections were responsible for occurrence of fetal anomalies (2). The occurrence of double headed monster was reported more frequently in ruminants (3, 4, 5). A successful vaginal delivery of dicephalus, Monostomus, Tetraophthalmus, Dibrachius and Dipus monster reported in Rathi cow is reported.

Case History and Clinical Observation

Approx. seven-year pluriparus Rathi cow was in third parity presented to Veterinary Clinical Complex of College of Veterinary Animal and Science, Bikaner. Owner reported that approx seven months pregnant cow given discharge from vagina and straining after fighting with other cows in herd from last 10-12 hours. Previously that cow delivered two normal calves after complete gestation. The cow was alert with wet muzzle and good body condition. The rectal temperature was recorded to be 102° F. Reddish discharge was noticed from vulva. On vaginal palpation, it was found that external os and internal os dilated three and two finger respectively. Vulva was edematous and vaginal mucus membrane was wet and congested.

Treatment and Discussion

Cow was handled after parental therapy of hormone estrogen and calcium, wait for dilation of cervix. The cervix was examined every 2 hours interval. After 10 hours cervix was dilated in sufficient proportion. On per vaginal examination, revealed that fetus was double headed in anterior longitudinal presentation, dorso-iliac position. The similar type of dicephalus monster has been reported previously (6, 5). The fetus has not given any living response during palpation. Before handling dystocia, lubricated the birth canal with liquid paraffin, then apply slight traction on head. In the present case the two heads were continued to a single neck leading to single thorax, two forelimbs, two hind limbs and one tail delivered without damaging the birth canal.

However, only one scrotum with two testes was present. In postmortem examination of the fetus

revealed that structure was duplicated upto pharynx whereas there was only one oesophagus. All other visceral organs e.g. lungs, heart, liver, kidneys, genitalia were of single fetus.

On the basis of gross examination, the fetus was classified as dicephalus, monostomus, dibrachius and dipus monster.

References

1. Robert, S. J. (2004). Veterinary Obstetrics and Genital Diseases. 2nd Indian reprints, CBS Publishers and Distributors, Delhi.
2. Prabakaran, V., Jayakumar, K., Kathirvel, S., Selvaraju, M. and Dharmaceelan, S. (2013). Dystocia due to bull dog calf in a nondescript buffalo. Shanlax Int. J. Vet. Sci. 1(1):43.
3. Thangadurai. and Selvaraju, M. (2015). Dystocia due to double headed monster fetus in Murrah buffalo under field condition. Indian Vet. J. 92(11):61.
4. Acharya, C., Kalita, M. K., Mahanta, Chhetri, N. B., Barman, U. (2019) . Dystocia due to a Dicephalus Monster Fetus in Holstein Friesian Cow. Int. J. Curr. Microbiol. App. Sci. 8(2):3024.
5. Periyannan, M., Selvaraju, M., Varudharajan, V., Fazil, A., Gopikrishnan, D. and Senthilkumar, K. (2021). Dicephalus tribrachius dipus dicaudatus monster in a jersey crossbred cow. The Pharma Inno. J. 10(5): 372.
6. Chauhan, P. M., Nakhshi, H. C., Suthar, B. N. and Parmar. V. R. (2012). Dicephalus, Monostomus, Tetraophthalmus, Dipus, Dibrachius, Dicandatus monster in a Kankrej Cow Vet. World. Vol.5(1): 38.



Repeat Breeding Syndrome in Cows

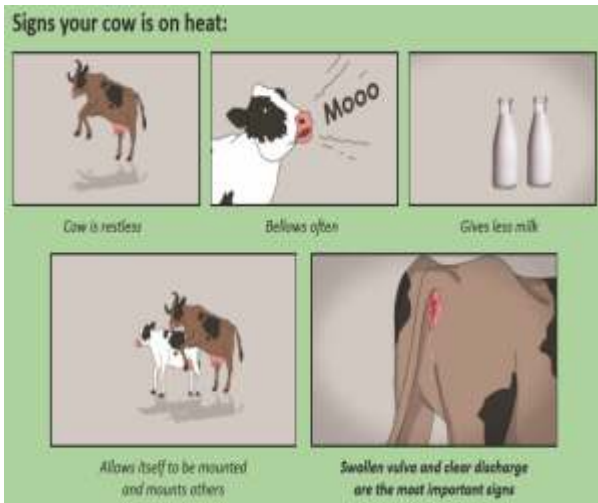
J Vinodhini and S Kantharaj

Rajiv Gandhi Institute of Veterinary Education and Research, Puducherry.

Introduction

The repeat breeding cow is the one that has normal or nearly normal estrous cycles and estrus periods and has been bred 2 or 3 or more times to a fertile bull or with fertile semen yet failed to conceive. It is also defined as one that

- Has returned to estrus after a third fertile service,
- It is less than ten years of age,
- Calved at least once,
- Doesn't have any abnormalities of the reproductive organs detectable by physical examination,
- Has no abnormal discharges from the genital tract,
- Has normal inter-estrus intervals.



The Repeat Breeding Syndrome (RBS) is more common in cows and high yielders but rare in heifers. In recent years, the problem of repeat breeding leading to infertility in cattle has been reviewed. Over the years, landless poor farmers faced severe financial crisis due to infertility caused by repeat breeding. It is a significant fact that the larger the herd the greater the infertility or incidence of repeat breeding. A survey of the problem of repeat breeding has shown that over 50% of the farms have this average incidence of affected animals. A survey conducted in few villages has also shown that 20 to 25% of the cows in a village are suffering from repeat breeding syndrome. Thus, finding the causes and treating the animals for RBS

have become imperative. This in turn, helps to reduce the economical loss and in scientific management of dairy cattle.

Causes Based on Reproductive Physiology

1. Failure of Fertilization by preventing the union of the sperm cell and the ovum. So, animal will not get conceived and suffer from RBS.

2. Early Embryonic Death (EED) It denotes the death of fertilized ova or embryos upto the end of implantation. It occurs early in few days of pregnancy (mostly from 11 to 42 days). In repeat breeders, the critical period for embryonic death is when the morula is developing into the blastocyst at 6 or 7 days especially the later day. Embryonic deaths occurring at 8 to 16 days after breeding usually do not affect the normal cycle length of about 18 to 24 days but embryonic deaths occurring at 16 to 25 days often result in prolonged periods between service and next estrus.

Pathological & Managerial Causes for Repeat Breeders

Genetic or Congenital Anatomical Defects of the Genital Tract

These defects include principally the anomalies or segmental aplasia of oviduct, uterus, cervix and vagina. These abnormalities largely cause failure of fertilization by preventing the union of sperm cell and ovum. Bilateral defects usually result in complete sterility. These defects are usually present in heifers. In the less severe conditions like uterus



unicornis, repeat breeding may occur but conception and normal pregnancy may follow. These defects are infrequent causes for repeat breeding, and many can be diagnosed by a careful rectal and vaginal examination.



Genetic /Congenital / Acquired Defects of the Ova, Spermatozoa or Early Zygote

These defects result in failure of fertilization or early embryonic death culminating in repeat breeding. Defective ova in older cows and post ovulatory aging of mammalian ova often result in failure of fertilization or polyspermy and early embryonic death. Aging of ova may occur due to breeding cows too late following the end of estrus. In-breeding has a deleterious effect on conception rate and embryonic survival. Daughters of certain bulls are apparently more fertile than daughters of other bulls. This may be partly due to reflection of chromosomal constitution of the ova. Crossbred cattle have superior reproductive performance. Aging of spermatozoa in the female reproductive tract occurs in a matter of hours and it appears that the fertilizing capacity of the sperm cell is lost before



Vulvovaginitis

changes occur in genetic materials.

Infectious or Traumatic Inflammatory Processes Affecting Genital Organs

It results in repeat breeding due to early embryonic death. Failure of fertilization may occur in - inflammatory processes resulting in unilateral / bilateral obstruction of genital tract; cows with severe metritis, cervicitis/vaginitis characterized by inflammatory exudates+bacteria (which is toxic to sperm resulting in loss of motility and death). Subclinical endometritis can cause failure of fertilization. Post coital pyometra due to pathogenic organisms such as trichomoniasis can cause infertility and repeat breeding due to EED probably secondary to an endometritis.

Endocrine Dysfunction as a common cause of repeat breeding is unlikely since the estrous cycle in these cows are normal except for prolonged post service periods when early or late embryonic deaths occur; anovulatory cycles result in failure of fertilization, while delayed ovulation may result in failure of fertilization (if sperm becomes poorly viable within 24-48 hours in cattle) or EED (aged sperm fertilize the ova and ends up in EED); Luteal insufficiency will certainly cause repeat breeding. Because progesterone is necessary for maintenance of pregnancy until 150-200 days or even to term, the main source of progesterone is corpus luteum(CL). If CL is not completely formed, pregnancy fails or terminates due to insufficient progesterone. Main luteotrophic hormone is lutenizing hormone(LH). Thus, if LH activity is enhanced (by injecting hCG/GnRH) after ovulation, proper development and function of CL may result. To increase the conception rate in cattle, GnRH can be given at the time of artificial insemination (AI). It synchronizes ovulation & increases the lutenization of follicular cells.



Managerial and Nutritional Deficiencies

It lowers the fertility rate and cause repeat breeding. Poor observation of cows, poor timing of AI, poor nutrition, poor handling of semen (from collection ,

processing to preparation of semen straw and poor AI technique), palpation of ovary at the time of AI which may cause accidental manual rupture of follicle. In addition, transportation and heat stress contributes to repeat breeding.

Treatment and Prevention

In case of anatomical defects and oviductal obstruction, culling is the only better choice economically. Because farmer/owner will not get any benefits from maintaining an infertile animal. Besides, owner should spend more money for availing treatment to the animal.

- Animal should be maintained in positive energy balance(example- providing mineral mixture supplements to cows, nutrients supplementation). Cow should maintain a good body condition score (neither thin nor obsessed).
- For delayed ovulation- double AI at 12-24 hours interval or multiple AI during estrus can be done. GnRH at the total dose rate of 10 microgram or hCG 1000-1500 IU can be given through the intramuscular route at the time of AI.
- For luteal insufficiency , progesterone supplementation (500mg) at 5th day after AI has been done. Intravenous antibiotics , antiseptics can be used in case of inflammatory conditions and infectious process. Intrauterine douching can also be done .This helps to increase the blood flow to the affected part and gradually increases the healing rate.
- Proper handling of good quality semen are more important factors to be considered when it comes to AI. AI should be done at the ideal time without damaging the internal organs.

Estrus synchronization protocol such as ovsynch can be initiated. The other method is to skip the estrus, give PGF2 alpha at 9th or 10th day followed by fixed time AI at 72 and 96 hours after PGF2 alpha injection.

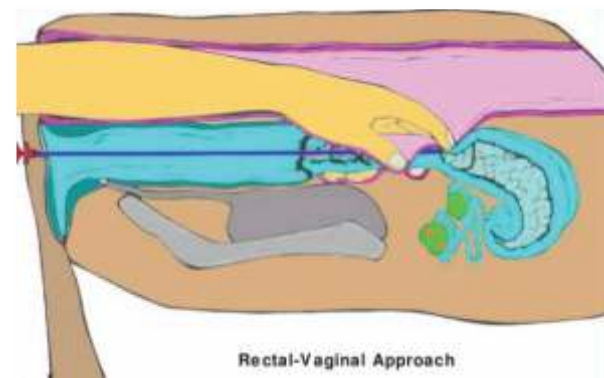
Conclusion

The general and broad physiological and pathological causes of repeat breeding in cattle have been outlined. Repeat breeders should be carefully evaluated in order to define the most probable reason for the failure to conceive (early repeats) or failure in pregnancy maintenance (early and late repeats). Because of the multiplicity of causes for repeat breeding that varies from herd to herd, cow to cow, estrus to estrus, diagnosis and therapy by the veterinarians requires great diagnostic skill and knowledge. Many excellent studies on various aspects of RBS have been

made and advances have been achieved in the past. Yet further studies on the causes and possible rational therapy for repeat breeding cow are needed.

References

1. Robert's, S. J. (1986). Veterinary Obstetrics and Genital Diseases, Third Edition, CBS Publishers and Distributors, India.
2. Senger, P. L. (2003). Pathways to Pregnancy and Parturition. Second revised edition, Current conceptions Inc., Pulman, WA99163-5607.
3. Sloss, V and Dufty, J. H. (1980). Hand book of Bovine Obstetrics, Williams and Wilkins, Baltimore /London.
4. Zemjanis, R. (2002). Diagnostic and Therapeutic Techniques in Animal Reproduction, Greenworld Publishers, Lucknow.



AI TECHNIQUE

<p>Lack of shade in feeding area There is no protection (shade) against solar radiation over the feed bunk</p>	<p>Open Mouth Breathing due to Heat Stress Open mouth breathing in lactating dairy cows without shade during hot summer weather. Cows have very few sweat glands</p>	<p>Environmental Management of Heat Stress Use of a combination of misters and fans to cool cows during hot summer weather</p>
<p>Environmental Management of Heat Stress Use of fans to cool lactating dairy cows</p>	<p>Environmental Management of Heat Stress Use of sprinklers to cool lactating dairy cows during the hot summer months</p>	<p>Cooling Pond Cows like to stand in the cooling pond during the hot summer months</p>

MANAGEMENT OF HEAT STRESS

Research Summary on Buffaloes in Marathwada Region: What We Know in Past 40 Years

A K Wankar, N M Markandeye and K U Tekale

Dept. of Veterinary Physiology, Dept. of Gynaecology and Obstetrics, College of Veterinary and Animal Sciences [MAFSU], Parbhani, Maharashtra.

Introduction

The College of Veterinary and Animal Sciences, Parbhani (Established – 1972) which is currently part of Maharashtra Animal and Fishery Sciences University (MAFSU), Nagpur, has conducted a coordinated research (for post graduate and doctoral degrees) on different breeds of buffaloes, including the Marathwada buffalo, for establishing baseline data covering various aspects such as, normal physiology, metabolism and nutritional needs, management of common bacterial, viral and parasitic diseases, management of parasites and pests by traditional and mainstream medicine, reproductive functions etc., The objective of this study was to improve performance of Marathwada buffaloes and to achieve maximum production in the times to come.

The buffalo owners of Marathwada region are not educated beyond the high school and mostly are marginal farmers with less than 2 hectares of land. An expansive study was conducted to serve and support these marginal farmers by reviewing the following aspects:

- Practices for animals housing, feeding, seasonal management, waste, farm and shed management
- Breeding strategies, heat detection, herd productivity, and milk production
- Cost of feed, farm economics, animal nutritional requirements at different growth and production levels
- Prevention of diseases and treatment, vaccination schedule, parasites and pest management
- Animal insurance, marketability of their milk and milk products

Few excerpts of the above study are presented herewith.

Marathwada Buffalo Breed

The Marathwada buffalo breed is an ancient buffalo breed of Marathwada region (Beed, Parbhani, Jalna, parts of Nanded, Latur and Osmanabad). These buffaloes here are mainly reared for milk and as draught animals. Other breeds in the region include Pandharpuri, Murrah, Jafrabadi, Nagpuri etc. District wise population of buffalo was mapped by NDDB [1]. Amongst different breeds of buffaloes, the Marathwada buffalo is very well adapted to the local climatic conditions and shows better adaptability than others [2]. The animals are reared under semi-

intensive management system, which is commonly adapted for cattle, but the population of Marathwada buffalo is constantly reducing with time [3, 4]. Grazing along with little sorghum, paddy straw, sugarcane leaves/tops, grasses and other farm waste mainly constitute their feed.

A study was conducted to evaluate the genetic resource of the Marathwada Buffalo breed. It was revealed that both male and female buffaloes of the breed had species and breed specific genetics. Any kind of abnormality was not observed in the hereditary material. It was concluded that the breed is having potent genetics, optimum vigour and better adaptability.

Milk Nutrition

Another study on selected buffaloes from 1st, 2nd and 3rd lactation had established the baseline data for blood and milk components. As per the study, there was an increase in serum glucose and chloride. Whereas Urea, Total Protein, Total Lipids, Cholesterol, Calcium, Phosphorus and Sodium have decreased significantly with the lactations and parity. Milk pH, specific gravity, chloride, urea, cholesterol and sodium in whey increased significantly with parity. Whereas, milk conductivity, lactose, total lipids, phosphorus and potassium decreased with subsequent lactations. Concentration of total lipids, calcium, phosphorus and potassium were higher in milk. It was concluded that the milk of Marathwada buffalo contains optimum levels of macro-nutrients and micro-nutrients (minerals/electrolytes) and should be regularly incorporated in human food. Also, as the animals mature the nutrients (total lipids, calcium, phosphorus and potassium) are maintained constantly at optimum levels and they do not decrease.

Parasites and Pest Management

Studies conducted on parasites and their prevalence in the region revealed that flukes, tapeworms, lice are amongst the most common ecto and endo-parasites in buffaloes. The fluke infestation was maximum during rainy season, then decreased during winter and was least during summer seasons, respectively.

The fluke prevalence had overall rate of 38.17 % in buffaloes, out of which Fasciola sp were maximum at 17.08 %; followed by Amphistomes sp, 13.07 %; Schistomes sp, 4.27 % and mixed fluke population at 3.81 %. Primary host for the fluke infestation was identified as snail, whose population increases in its breeding season i.e., monsoon. Adult buffaloes (>36

m) had maximum infection rate at 48.73 %, followed by growers (12-36 m) at 37.60 % and least for calves at 19.01 %. Sex-wise prevalence was also noted and males had higher infection rates as compared to females. Breed-wise studies revealed that, Marathwada buffalo was least infected by flukes amongst Murrah > Non-Descript > Nagpuri > Pandharpuri > Jafrabadi breeds.

Similarly, more number of animals were infected by tapeworms in monsoon (39.39 %) season as compared to summer (15.68 %) and winter (7.04 %). Both roundworms and helminths had same prevalence rates in buffaloes at 20.23 %. It was found that as compared to growing animals (< 3yrs) and adults (> 3yrs) the calves (0-12 m) had more infection rates at 36.45 %, 8.95 % and 41.55 %, respectively. Sex-wise prevalence was also noted and males had higher rates of infection as compared to females at 29.02 and 18.20 %. The different roundworms like Ascarid sp, Stongyloides sp, Trichuris sp, Mixed worm sp, Strongyles sp were prevalent at following rates, 4.8 %, 1.18 %, 3.29 %, 5.12 % and 5.58 %, in buffaloes. Management of snails during rainy season, which are the primary host for these parasites is very important as the infected animals have poor growth, reproductive performance and reduced production efficiency.

Regional prevalence of lice revealed, that Jalnadistrict (17.50 %) had the highest infection rates, followed by Parbhani (15.96 %), Beed (8.30 %) and Nanded (3.41 %) districts of Marathwada region. Here the most common lice species identified was Haematopinus Tuberculatus sp, with prevalence rates of 32.88, 35.12, 28.50 and 32.21 %, in Parbhani, Nanded, Jalna and Beed districts, respectively. The lice infection was maximum during winter months of November, December, January and February, than the rest of year. As effective treatment against lice, several herbal preparations like karanj oil (83-100%), eucalyptus oil (85-92%), Neemazol / neem oil (86-100%) were prepared and tested. It was observed that, all the herbal formulations were safe to use, killed lice efficiently and were potentially more economical to use.

Handling of Metabolic Diseases

Some of the important metabolic diseases associated with pregnancy and lactation were also studied and, clinical mastitis was prevalent in parbhani district at 12.37% rate. Incidences of mastitis were more during monsoon (14.47%), than other seasons (Summer-11.20 % & winter-10.86 %). Similarly, with parity and lactations the animal is predisposed to mastitis and highest incidences were during 3-5 lactation as compared to 1-2 parity. The buffaloes in first and second lactation had low mastitic incidences at; 20.00 % and 23.52 %, respectively while, as the parity increased from third (31.18 %), fourth (32.43 %) and to fifth (32.43 %) all types of mastitis were more pronounced. Sub-acute type of mastitis was most commonly recorded at 35.71 %, followed by chronic

(28.57 %), interstitial (21.42 %) and chronic granulomatous mastitis (14.28%). To prevent mastitis which adversely affects milk production and animal health and performance, it is pertinent to take proper care of lactating females during monsoon season. Shelter management also plays crucial, especially during wet seasons for prevention of mastitis. It is pertinent to take care of female buffaloes after second parity till senescence or end of active sexual life, to prevent mastitis, reduce production losses and to make the milk business sustainable for dairy owners.

Other diseases like, clinical ketosis, hypocalcaemia and hypophosphatemia were also studied in depth in buffaloes and it was noted that cases of clinical ketosis were highest in early lactation period. While, incidences of hypocalcaemia were highest at early lactation and in dry females. Hypophosphatemia didn't affect early and late stages of lactation but, mid lactation stage and dry animals were affected. Haemoglobinuria, another disease had highest incidences in advance pregnant (75 %) and recently calved females between 3-5th parity (76.92 %). These metabolic diseases are very common in moderate-high yielder. Lack of optimum nutrition, especially during last trimester of gestation, drains the important nutrients (Macro-Micro minerals) and predisposes the animals to various metabolic disorders. Metabolic stress during pregnancy and its continuance, thereafter, is a major reason for development of these conditions. Hence, it is advisable to provide optimum balanced feed in the end of last trimester and early to mid-lactation to buffaloes.

Clinical condition	Lactation Stage	Prevalence (%)
Ketosis	Early	50
	Mid	20
	Late	10
	Dry	00
Hypocalcaemia	Early	20
	Mid	10
	Late	10
	Dry	20
Hypophosphatemia	Early	00
	Mid	20
	Late	00
	Dry	40

Zoonotic Diseases

The study of zoonotic pathogenic diseases like brucellosis and rotavirus were also undertaken in the region. The brucellosis seroprevalence was very limited in Parbhani district and brucellin and ELISA were identified as most easy and accurate diagnostic confirmatory tests for brucellosis. Similarly, rotavirus prevalence was very less in both humans and buffaloes in the entire region. Rotavirus primarily

targets intestinal tract and produces persistent diarrhoea. Highest incidences of the rotavirus in buffaloes was established in December, January, February months and again decreasing from March/April onwards. While in human population the same virus was more active during April and December, January, February months of the year. All the zoonotic diseases are serious problem for both animals and humans, due to their high morbidity or mortality and affect the economics of farming significantly. It remains a constant challenge to reduce the human animal interaction, to prevent zoonotic diseases.

Reproductive Disorders and Management

To improve the reproductive performance in buffaloes, common reproductive disorders and infections were also studied extensively in the region. Several treatments and protocols were tested and established, to resolve the common reproductive problems like anestrus, estrus synchronization, repeat breeding, silent heat and endometritis. Most effective treatments were GnRH and OVSYNCH protocol in repeat breeders. It was found out that these two treatments had success rates of 75.00 % and 66.70 % in treatment of non-infectious repeat breeding buffaloes.

Another protocol developed to ameliorate the problem of silent estrus. The dosage were 50 mg progesterone/day/buffalo and 2.5 ml GnRH at the time of artificial insemination. The treatment resulted in higher estrus expression (>80 %), increased the average estrus duration to 25.37 hrs and improved the conception rate and overall conception rates to 87.50 % and 70.00 % respectively. It was concluded that the combination protocols were more effective in improving the common reproductive problems in buffaloes.

Others tried, treatment of endometritis and repeat breeding in buffaloes by combination therapy. Different herbal, chemical and bacterial compounds were used for the treatment like; neem seed extract (@ 20%), lugols iodine (@ 4 %), neem extract + levamisole, lugols iodine + levamisole, lipopolysaccharide E. coli and lipopolysaccharide E. coli + levamisole. Lowest efficacy for repeat breeding and endometritis were found for neem extract (55.55%), neem extract + levamisole (55.55 %), followed by lugols iodine (66.67 %) and highest were for E.coli LPS (88.89%) and its combination levamisole+ E.coli LPS (88.89%). New treatment protocols like, combination of chemical + herbal + immune modulators, are very effective and economic way to treat common reproductive disorders and infections in buffaloes.

Housing

The effect of different housing systems viz., with or without shade nets was also observed. The normal physiological responses i.e., heart rate, pulse rate and respiration rates were higher in shade net house, while rectal temperature was higher in animal house with no

shade net. In the same study, haemogram picture of animals in shade net housing showed higher haemoglobin, total leukocyte count, total erythrocyte count, lymphocytes, neutrophils, packed cell volume, mean corpuscular volume, mean corpuscular haemoglobin concentration. From the haemogram it can be said that shade net housing is more favourable for animal's normal haematological functions. Whereas the biochemical variables i.e., serum cholesterol, protein, albumin, globulin were significantly higher in shade net house. Although the physiological functions alter marginally, normal haematopoietic and biochemical functions are optimum in shade net houses as compared to non-shade net animal houses. Also, the shade net housing system provided protection from ectoparasites and pests, increasing the comfort levels of housed animals.

Feed

To improve animal nutrition, economics and production, the newly developed varieties of paragrass and Yashwant were tested. It was found that, feeding Yashwant instead of traditional paragrass, improved dry matter intake/body weight and significantly increased body weight gain (higher growth), nutrient digestibility, milk yield, milk fat % and reduced feed cost substantially. New Yashwant variety was established as a superior fodder in nutrient quality and lowers cost of milk production and hence should be incorporated instead of paragrass in buffalo ration.

Conclusion

In order to achieve the target of sustainable and profitable dairying, the findings of such studies are required to be incorporated into Government policies and the dairy farmers should be encouraged to learn, appreciate and apply the results of such studies in their farm management.

References

1. National Dairy Development Board, Dairying in Maharashtra, A statistical profile. 2015.
2. Hadi, M.A. (1965). A preliminary study of certain productive and reproductive characters of Marathwada Buffaloes of Maharashtra State. Indian Veterinary Journal. 42(9): 692.
3. Agri-IS, animal genetic resources of India, <http://14.139.252.116/agris/breed.aspx>.
4. Government of India, Ministry of Agriculture and Farmers Welfare, Department of Animal Husbandry, Dairying and Fisheries, Breed Survey. 2012-13.

Mithun - An Animal of Indian Pride: A Review

Danveer S Yadav, Sandeep Nanavati, Rashmi Choudhary and M S Jamara
College of Veterinary Science and Animal Husbandry, (NDVSU), MHOW, Madhya Pradesh.

Introduction

Mithun, also known as “Cattle of Mountain”, is an important bovine species of north-eastern hilly region of India and also of China, Myanmar, Bhutan and Bangladesh. This magnificent massive bovine is presently reared under free-range condition in the hill forests at an altitude of 1000 to 3000 m above mean sea level. Mithun plays an important role in the socio-economic and cultural life of the local tribal population. Presently, this animal is mainly reared for meat, which is considered to be more tender and superior over the meat of any other species. Mithun milk though produced less in quantity is of high quality and can be used for preparation of various milk products. Leather obtained from this species has been found to be superior to cattle. With the dwindling population of Mithun over the years and gradual denudation of free-range area along with the biotic and abiotic stress there is urgent need of scientific intervention for proper management as well as conservation of this beautiful hill animal through implementing an effective conservation program.

The North-East India is the hotspot of floral and faunal biodiversity and the habitat of a number endemic species. Of these species, Mithun (*Bos frontalis*) is an important one which needs support for healthy propagation. However, due to denudation of free range along with the biotic and abiotic stress, there is urgent need of scientific intervention for proper management as well as conservation of this beautiful hill animal through implementing an effective conservation programme. Biotechnologies such as cryo-preservation of semen and embryos, coupled with artificial insemination and embryo transfer are important potential tools for the preservation of animal biodiversity. Frozen semen technology offers a very potent means of in vitro conservation of male germplasm. Mithun is a massive semi-domesticated rare ruminant species mainly reared for meat. This strongly built hill animal of Southeast Asia plays an important role in the socio-economic and cultural life of the local population (1). In India, Mithun meat is considered to be more tender and superior over the meat of any other species. At present, Mithun farmers rear this animal at an altitude of 1000 to 3000 meters above mean sea level under free grazing condition in its natural habitat. Recently initiatives are being taken to popularise economic Mithun farming under semi-

intensive condition with controlled breeding. While wild Indian Gaur, the ancestor of Mithun is in a vulnerable position (2). Mithuns are subject to non-cyclical population decrease and local-regional contractions indicative of this species or its population that are not yet endangered but may become so in the near future. With a decline in population size inbreeding can occur which in turn can reduce reproductive fitness including fecundity and survivability (3). Furthermore the present free ranging Mithun rearing system permits grazing of limited number of these animals in a particular hill pocket without any migration to other locations and vice versa that results in considerable inbreeding in this species. When genetic variation is maintained increased population vigour and the ability to adapt to environmental change are enhanced. Grazing of local cattle together with Mithun in the same forest area increased the chance of crossbreeding with the local cattle that may result in a loss of species uniqueness and specialized adaptive and fitness traits and failure to respond in captivity owing to behavioural problems associated with confinement. In addition, very recently slaughtering of Mithun on regular basis for meat imposed further threat on the future population size of this species.



Mithun Inhabited Areas in India

The total Mithun in the country is 3.9 Lakhs in 2019, increased by 30.0% over previous livestock Census (2012). Mithun, Yaks, Horses, Ponies, Mules, Donkeys and Camels taken together contribute 0.23% of the total livestock. In the North Eastern Hill Region (NEHR) of India, Mithuns are distributed in four different States namely Arunachal Pradesh, Nagaland, Manipur and Mizoram.

Table 1: Mithun Population 2012 and 2019

Category	Population (In Lakh) 2012	Population (InLakh) 2019	% Change
Mithun-Total	3.0	3.8	26.66
Male	1.3	1.7	30.75
Female	1.7	2.1	23.52

2: Mithun Population 2012 & 2019 of Major States

S.No	States	Population (In numbers) 2012	Population (In numbers) 2019	% Change
1.	Arunachal Pradesh	249000	350154	40.62
2.	Nagaland	34871	23123	-33.69
3.	Manipur	10131	9059	-10.58
4.	Mizoram	3287	3957	20.38

Peculiar Features of Mithun

This animal prefers cold and mild climate. They are browsers like goats and can utilize coarse fodders, which are generally not consumed by other livestock. This is a very fertile animal, which can produce one calf in a year with age at puberty varying from 22 to 30 months. Their productive life ranges from 16 to 18 years. This is an underutilized animal and has a great potential for quality meat, milk and leather production. The quality of meat, milk as well as leather of this animal is very good and there is a great scope to promote this animal as an organic meat and milk producer. This animal is also used as bridal gift as well as in barter trade and the milk producing capacity of this animal also needs to be explored.

Types of Mithun Strains

As described by *Verma (1996) (4)*, two distinct types of Mithuns are available in India and they were named after the name of the State where they belong (Nagami and Arunachali). These two distinctive types have also been reported by (5). However, *Bhusan et. al., (2000) (6)* have identified four distinct strains of Mithun and named them as Arunachalee, Mizorami, Nagami and Manipuri



strain. The names indicate their home tract in north eastern States of India. As per the survey conducted by the National Research Centre on Mithun to identify different strains of Mithun in North Eastern Hills of India, four different strains have been identified. Characterization of these four different strains has been done based on 37 phenotypic characters and genetic characterization was done through RAPD. Results were suggestive of genotypic difference among four different strains.

Socioeconomic Importance of Mithun

Mithun is considered as the pride of North Eastern Hilly region of India. This animal plays an important role in the social, cultural and economic life of the local tribal population. The ownership of Mithun is considered to be the sign of prosperity and superiority of an individual in the society. Farmers mainly rear Mithun for meat purpose. Besides, this animal is also used as marriage gift and sacrificial animal for different social and cultural ceremonies. Though at present farmers do not consume its milk, this animal produces highly nutritious milk.

Being a meat animal the growth rate of Mithun is the prime concern of farmers. With adequate feeding the growth rate of this animal varies from 300 to 600 g/ day, which is comparable with cattle and buffalo. However, the plasma growth hormone concentration (30-90 ng/ ml) (7, 8, 9) is much higher in Mithun than in any other domesticated animals. The consumption of Mithun meat is not a regular practice in tribal society. These animals are sacrificed for meat only during important social ceremonies and festivals. However, there is a great demand for Mithun meat and consumers consider this meat as more tender and superior over the meat of any other species except pork. The dressing percentage in Mithun varies from 48 to 54 % in different age groups. However, to achieve an optimum dressing percentage, it is suggested to slaughter Mithuns at 4 to 5 years of age. There is a great scope to utilize this meat to make some value-added meat products. The National Research

Centre on Mithun has already standardised the process of making some value-added products of meat like meat nuggets, meat powder, meat Patties as well as meat block. The organoleptic test conducted by the institute on these products revealed high scoring of 6-7 in the scale of 1-8.

Presently, the consumption of Mithun milk is not an accepted practice among its rearers. Mithun produces around 1 to 1.5 kg milk per day. However, Mithun milk is nutritionally superior to any other domesticated species as it contains high fat (8 to 13%), solid-not-fat (18 to 24%) and protein (5 to 7%). Hence, Mithun has a scope to be promoted as moderately good milk animal for home consumption in these hilly areas. Due to high fat and protein content in Mithun milk, it may be used for the preparation of different value-added milk products such as paneer, various sweet products, ghee, cream, curd and cheese. The National Research Center on Mithun, the premier Institute of Indian Council of Agricultural Research, has successfully standardized the process of making paneer, barfi, rasgulla, curd and lassi from Mithun milk.

The quality of Mithun hide is found to be superior in comparison to the traditional cow hide (10). The National Research Centre on Mithun has successfully processed different varieties of leather from Mithun hide. Mithun hide has been found to be very good for the production of upper shoe leather, bag leather and garment leather. Bag leather has been found to be much superior to cow leather. Besides, Mithun hide with hairs could be an excellent exotic outer cover for a sofa.

Scientific Rearing System

Currently farmers rear Mithun under free-grazing condition in the forest area without any additional housing or feeding facilities. Occasionally, farmers bring back the female Mithun just before parturition and send it back to the forest following parturition. However, it is suggested that even under a free-range system, a temporary housing structure using locally available materials can be constructed in some strategic locations in the Mithun rearing area. Mithun can also be trained to come to the shed at a particular time every day by providing little bit of concentrate and salt. This will be helpful for farmers to supervise, provide additional feeding and medication to their animals. Besides, farmers will also get an opportunity to look after the individual animal regularly for any kind of discrepancy or disorder. If farmers opt for semi-intensive system of rearing, they should go for housing structures with feeding and watering provisions and they can also tie the animals at night once they come back from the forest after grazing. The supervision of individual animals, additional feeding, watering and medication can be done there in late evening or

early morning.

Feeding Management

Mithun thrives on the jungle forages, tree fodders, shrubs, herbs and other natural vegetations (11). Farmers do not provide any additional feeding. Though the animals are owned by the farmers, they are kept under natural forest in a semi-wild condition. However, farmers occasionally provide common salt, especially at the time of restraining for some purposes. Each individual owner can identify his Mithuns even though they do not bear any identification marks and similarly each Mithun knows his owner which is reflected in the fact that the Mithun approaches the owner periodically for salt. In other words, the owner does not have to invest anything in his Mithun as they are simply let loose in the forest which constitutes around 50 percent of total land area of the region. Owners generally keep Mithuns in community herd in fenced hilly jungle area and village councils assign Mithun grazers to take care of their animals.

As Mithun entirely depends on the locally available jungle fodders, special care should be taken in terms of mineral supplementation for better performances (11). In steep hilly slope, the leaching of mineral elements is a common phenomenon especially during rainy season. Therefore, in a particular hilly grazing gradient the soil will be deficient in some important mineral elements. In that case the vegetation of that particular area will also be deficient in some of the mineral elements, which may induce mineral deficiency. The only option to correct this situation is mineral supplementation. However, the salt licking behaviour as well as drinking of mineral water sources in the hills is the natural way to meet the requirement of minerals in these animals (12).

During the lean season, when availability of jungle fodders goes down, additional concentrate supplementation may be required. It is advisable that during the flush season when abundant fodders are available in the jungle, the salt and mineral mixture together may be fed additionally to the animals to avoid mineral deficiency. Whereas, during lean season additional concentrate feed (15% CP and 70% TDN) fortified with salt and mineral mixture (1 to 2 kg per animal daily up to 2 years and 2 to 4 kg per animal daily above 2 years) may be offered to maintain optimum performances (11). For lactating Mithun, as it produces less quantity of milk, no additional feeding is required. In free-range Mithun, these feed supplements may be provided to the animals in the shed constructed in strategic location in the grazing area. Whereas, for animals under semi intensive system the feed supplements may be provided in the shed in late evening or early morning whenever the animals are

ried. It has been found that the drinking water requirement for Mithun is approximately 9% and 12% of body weight, during winter and summer respectively. Therefore, the provision of adequate drinking water according to this specification is highly essential.

Breeding Management

Like cattle, Mithun is a poly-estrus animal. The healthy adult female Mithun show repeated estrus cycles at an interval of 19 to 24 days unless it is pregnant. The Mithun breeds throughout the year and no definite breeding season is observed in this species. The length of gestation period, service period and calving interval in Mithun varies from 270 to 290 days, 50 to 100 days and 350 to 400 days, respectively. The age at puberty and age at first calving varies from 27 to 36 months and 40 to 48 months, respectively. The Mithun bulls become mature to breed at 3 to 4 years of age. Under free-range system, a practical approach for selective breeding in Mithun is the introduction of superior and tested bulls (1 bull for 10 breedable females) in the herd and simultaneous culling of the unwanted bulls from the herd. Efforts should be made to replace breeding bulls preferably once in five years to avoid inbreeding depression. Under semi intensive system, the female can be detected in heat to be bred with superior bulls either through natural service or artificial insemination.

The expression of estrus behaviour is silent in Mithun (13). Unlike cattle, it is difficult to detect heat in Mithun through visual observations. Among all the behavioural signs of estrus, the mounting of Mithun bull over estrus cow is the best indicator of estrus followed by standing of estrus cow to be mounted by Mithun bull. Congestion of vulval mucous membrane and swelling of vulva are also important signs of estrus in Mithun cows. In contrast, other signs like mucous discharge, restlessness and alertness, tail raising, frequent urination and loss of appetite were found to be less prominent estrus signs in Mithun cows (13). Bellowing is not generally observed in Mithuns during estrus. The genital organ of Mithun cows during estrus reveals relaxed and open os externa of cervix, turgid uterus and ovaries having palpable follicles. However, it is suggested to use healthy Mithun bulls to detect heat. In Mithun, ovulation occurs between 20 to 31 hours after the onset of estrus (14, 15, 16).

Conservation of Mithun

There are three ways for the conservation of Mithun genetic resources: i) through cryopreservation of genetic material like living ova, embryos or semen; ii) preservation of genetic information as DNA; and iii) conservation of live population (in situ

conservation).

The need for parallel conservation of Mithun genetic resources along with live animal conservation as raw material for future breeding programmes should be recognized and has become an important issue in planning of Mithun husbandry. Conservation is of particular concern in the Mithun inhibited regions where there is effort for agricultural change, thereby the risk of gradual replacement of indigenous stocks and farming methods by new techniques. Those areas where climatic extremes and particular parasitic conditions may result in genetically modified and unique local stocks which are able to survive under extreme conditions need to be given proper attention. Such conservation efforts are particularly important in the light of predicted global climate change and the ability of microbial and insect parasites to evolve and adapt to modern chemical control methods.

Ex situ versus In situ Methods of Conservation for Mithun

Ex situ preservation involves the conservation of Mithun in a situation removed from their normal habitat. It is used to refer to the collection and freezing in liquid nitrogen of animal genetic resources in the form of living semen, ova or embryos. It may also be the preservation of DNA segments in frozen blood or other tissues. Finally it may refer to captive breeding or other situations far removed from their indigenous environment. This is the storage of animal genetic resources which farmers are currently not interested in using. It includes cryogenic preservation and the maintenance of breeds from domesticated species as live-animal populations in parks, zoos and other locations away from the environment in which they are being developed. The global programmes on ex situ conservation strategy is still being developed but it is based on the use of live-animal populations wherever practicable supported by cryopreservation where technology exists or can be developed combining within-country gene banks with global repositories of last resort. This strategy is in keeping with the Convention on Biological Diversity. The technology required for storing both male and female gametes of all species of interest is not yet developed. Of course, interested governments, non-governmental organizations, research institutions and private enterprises will be encouraged to maintain in vivo samples of breeds at risk with national inventories being established and kept up to date so that the genetic resources are readily available for use and study.

In situ conservation is the maintenance of live populations of animals in their adaptive environment or as close to it as is practically possible. For domestic species the conservation of

live animals is normally taken to be synonymous with in situ conservation

Conclusion

Mithun husbandry in North Eastern hill region of India is an important component of the livestock production system. Scientific rearing of this species will not only support the need of protein but also help to generate extra income to the poor peoples. The recent success in the field of artificial insemination, estrus synchronization coupled with timed AI and embryo transfer technology will definitely help to go a long way to achieve the target of propagating quality germplasm in the farmers' field.

References

- Mondal, M., Karunakaran, M., Lee, K. and Rajkhowa, C. (2010). Characterization of Mithun (*Bos frontalis*) ejaculate and fertility of cryopreserved sperm. *Animal Reproduction Science*. 118: 210.
- Baillie, J. and Groombridge, B. (1996). IUCN Red List of Threatened Animals. IUCN, 1996; Gland, Switzerland.
- Ralls, K. and Ballou, J. D. (1986). Preface to the proceedings of the workshop on genetic management of captive populations. *Zoo Biology*. 5: 81.
- Verma, N. D. (1996). Mithun- a magnificent Bos species. In Dairy India (Ed) Gupta PR, A 25 Priyadarshini Vihar, Delhi. pp. 180.
- Arora, C. L. (1998). Less used animal: Yak and Mithun- an overview: *Indian Journal of Animal Science*. 68 (8, special issue): 735.
- Bhusan, S., Sharma, D., Bujarbaruah, K.M. and Singh, R.V. (2000). Annual Report, National Research Centre on Mithun, Jharnapani, Nagaland 797106: 4.
- Mondal, M., Rajkhowa, C. and Prakash, B. S. (2006a). Exogenous GH-releasing hormone increases GH and LH secretion in growing Mithuns (*Bos frontalis*). *General and Comparative Endocrinology*. 149: 197.
- Mondal, M., Rajkhowa, C. and Prakash, B. S. (2006c). Determination of effective dosage of GH-releasing factor for blood GH responses in Mithun (*Bos frontalis*). *Journal of Animal Physiology and Animal Nutrition*. 90: 453.
- Mondal, M., Rajkhowa, C. and Prakash, B. S. (2006d). Plasma growth hormone concentration in Mithun (*Bos frontalis*) of different ages: relation to age and body weight. *Journal of Animal Physiology and Animal Nutrition*. 91:68.
- Das, K.C., Mukherjee, G., Baruah, K.K., Khate, K. and Rajkhowa, C. (2011). Study of growth performance and leather quality in Mithun (*Bos frontalis*) and its comparison with local cattle (*Bos indicus*) fed on tree leaves-based ration. *Livestock Research for Rural Development*. Volume 23, Article #104. Retrieved August 17, 2012, from <http://www.lrrd.org/lrrd23/5/das23104.htm>.
- Das, K.C., Haque, N., Baruah, K. K., Rajkhowa, C. and Mondal, M. (2010). Comparative nutrient utilization, growth, and rumen enzyme profile of Mithun (*Bos frontalis*) and Tho-tho cattle (*Bos indicus*) fed on tree-leaves-based ration. *Tropical Animal Health and Production 2010 Online First* DOI:10.1007/s11250-010-9676-1.
- Prakash, B., Rathore, S. S., Khate, K. and Rajkhowa, C. (2013). Nutrient composition of forest based foliages consumed by Mithun (*Bos frontalis*) under Imphal district of Manipur. *Livestock Research for Rural Development*. Volume 25, Article #187. <http://www.lrrd.org/lrrd25/10/prak25187.htm>.
- Mondal, M., Karunakaran, M., Rajkhowa, C. and Prakash, B. S. (2008). Development and validation of a new method for visual detection of estrus in Mithun (*Bos frontalis*). *Applied Animal Behaviour Science*. doi: 10.1016/j.applanim.2008.02.009.
- Mondal, M., Rajkhowa, C. and Prakash, B. S. (2006b). Behavioral estrous signs can predict the time of ovulation in Mithun (*Bos frontalis*). *Theriogenology*. 66: 1391.
- Mondal, M., Rajkhowa, C. and Prakash, B. S. (2006e). Relationship of plasma estradiol-17 β , total estrogen and progesterone to estrous behavior in Mithun (*Bos frontalis*) cows. *Hormones and Behavior*. 49:626.
- Mondal, M., Rajkhowa, C. and Prakash, B. S. (2006f). Timing of Ovulation in Relation to Onset of Estrus and LH Peak in Mithun (*Bos frontalis*) Cows. *Reproduction in Domestic Animals*. 41: 479.

Bargur Cattle : An Overview

C Pugazharasi, M Abarna, T Sarath, N Arunmozhi and Cecilia Joseph

Department of Clinics, Madras Veterinary College, Tamil Nadu Veterinary and Animal Sciences University, Chennai, Tamil Nadu.

Introduction

Cattle breeds evolve based on the agro-climatic conditions and socio-economic status of the people especially in countries like India whose backbone is agriculture, draught breeds play vital role in contributing to the livelihood of landless and marginal farmers (1). One such popular draught breed belonging to Tamil Nadu is Bargur cattle with its native tract at Bargur forest hills of Anthiyur Taluk of Erode District in western Tamil Nadu in India which is on an average 1000m above the mean sea level. They are wild and semi-hardy to handle (2). They are also called as "Semmarai" in their native tract and are reared by a group of people called "Lingayats" (1). They are famous for their power of endurance and unsurpassed speed in trotting (3).

These are medium sized animals and are compact in size. These cattle have a typical brown colour with white patches with variation of coat colour from cherry red to light red and the hair is short, straight and fine. They have moderately broad forehead with long head which tapers towards the muzzle. Hump is well developed in bulls and moderately developed in bullocks. In adult bulls, bullocks and cows, height at withers, body length and chest girth are 119, 114 and 119; 126, 126 and 139; 116, 115 and 124 cm respectively. Muzzle, eyelashes, horns and hooves are brown with short and horizontal ears (2).

Population Status

Population of the Bargur cattle in the native tract was estimated to be 95,400 in 1977, which got reduced to 46,600 in 1982 and 10,102 in 2003. Recent enumeration of purebred Bargur cattle in its entire native tract in the year 2007, revealed that the presence of about 2500 heads with only 1100 breedable females. Thus, there has been decline of more than 93 % in the breedable female population size of Bargur cattle from 1977 to 2009. The population size falls under "Endangered" category as per prescribed standards for conservation (<http://www.tanuvac.ac.in/bcrs.html>).

Bargur cattle are the only native Indian cattle which has recently experienced the genetic bottleneck in their population. It can be due to several reasons including mechanization of agriculture and reduction of market demand for draught cattle, restrictions imposed on cattle grazing inside the forests, lack of systematic breeding, lack of

awareness among the herdsmen to breed true to the type animals, etc and this bottleneck event could have led to loss of some rare alleles in the species and thus conservation becomes conspicuous (4). Hence, TANUVAS has established Bargur Cattle Research Station in Bargur, Erode district with an aim to conserve this breed with total budget of the project 600 lakhs. However, "Community-based conservation" is essential for recovering the population status of the breed.

Rearing System

When group of Kannada speaking people named "Lingayats" migrated to Tamil Nadu they came along with their cattle and buffalo. Every month they offered the minor millet named "Varagu" in tamil, "Bargu" in kannada to their community deity and hence their home tract got the name "Bargur" and so the cattle. These cattle are maintained under zero input condition by grazing and they are kept penned in forest for most parts of the year and are mostly maintained for dung and draught in their hometract (1).

They are taken to forest during the month of July/August (after sowing) and are kept penned there and the places vary based on the availability of fodder and water. The animals from 25-30 households were collected and were sent to deep forest. Temporary enclosures were constructed in the forest for the stay of herd man and the animals during night time. These cattle camps are called as "Pattis" in local language. During the month of January/February, they are taken back to village to graze the remains in the harvested fields (4).

Reproductive Parameters

The average age at first oestrus and first calving is 30.00 ± 1.36 months and 40.61 ± 1.21 respectively (2). Average calving interval is 12-18 months (2). Cows are poor milkers and produce an average of 0.5 to 3 litres milk per day after suckling by calves (5). The average body weight of breeding bulls at the time of semen collection is 183.33 ± 2.4 Kg. The mean semen volume is 3.708 ± 0.125 ml and the semen is creamy white in colour with a mass activity of 0.93 ± 0.18 . The semen concentration is around 1221.0 ± 56.09 (6). In most cases natural services were practised and no charges were paid for the natural service done by the bull. Only about 6.7% farmers followed artificial insemination using Kangayam bull semen. The age at selling for utility

is to 2 years and for culling it is 14 years (1).

Summary

Bargur cattle are hilly draught breed and are second recognized after Kangayam of Tamil Nadu. They are famous for their endurance and their unsurpassed speed in trotting. They are maintained under zero input grazing system and are maintained mainly for their dung and draught alternatively in forests and harvested lands. This breed has experienced genetic bottleneck in their population due to drastic reduction in their population size to more than 93% and thus, the Tamil Nadu government has established Bargur Cattle Breeding Station in Bargur, Erode district and several strategies were adapted to recover the population from "Endangered" category.

References

1. Kavithaa, N. V., Vimal Rajkumar, N. and Mathialagan, P. A. (2019). A Social Assay on Bargur Cattle Farming System in Tamil Nadu. *Int. J. Livest. Res.* 9(8): 154.
2. Ganapathi, P., Rajendran, R., Subramanian, A. and Meenakshisundaram, S. (2013). Bargur Cattle: Characterization and Management Practices. *Indian Vet. J.* 90(11):09.
3. Nivsarkar, A. E., Vij, P. K. and Tantia, M. S. (2000). *Animal Genetic Resources of India: Cattle and Buffalo*. Directorate of Information and Publications of Agriculture, ICAR, New Delhi.
4. Ganapathi, P., Rajendran, R. and Kathiravan, P. (2012). Detection of occurrence of a recent genetic bottleneck event in Indian hill cattle breed Bargur using microsatellite markers. *Trop. Anim. Health. Prod.* 10.1007/s11250-012-0171-8.
5. Pundir, R. K., Kathiravan, P., Singh, P. K. and Manikhandan, V. A. (2009). Bargur cattle: status, characteristics, and performance. *Indian J. Anim. Sci.* 79(7): 681.
6. Ganapathi, P., Manokaran, S. and Prakash, S. (2019). Establishment of Seminal Parameters Values for Endangered Bargur Cattle. *TNV-35th VCRI-CA-Dec-2019-AAIO-18*.



Assam Hill Goat - An Indigenous Goat Breed of Assam

Kandarpa Boruah and Goutam Bordoloi

Lakhimpur College of Veterinary Science, Assam Agricultural University, Joyhing, North Lakhimpur, Assam

Goat is a most profitable and demandable livestock farming component in present context of livestock farming in India particularly in Assam. The goat population in Assam as per the 20th livestock census is 4315.17 thousand (1). The majority of goat population in Assam is Assam hill goat, although Black Bengal, Sirohi, Beetal, Jamunapari are available in some parts of the state.

The Assam Hill goat is the 31st recognized indigenous goat breed of Assam by National bureau of Animal genetics Resource, Karnal, Haryana. It is very well adapted to the native climatic condition and resistant to diseases. They are mostly found in the plains of the Brahmaputra and Barak valley and in the hilly tract of Dima Hasao and Karbi Anglong districts of Assam and in the adjoining hilly tract of Meghalaya state (2).

The Assam Hill Goat breeds are mainly reared for meat purpose and they produce a good quality chevon. The goats have been also utilized for their milk, hair, skin and manure and it can provide ready liquidity cash value. It is a poor milk yielder. It has been noticed that this breed was utilized by the poor farmers of Assam and its adjoining areas for animal protein as well as their rural livelihood since long back.

These animals are short legged with small body size. Their body size is smaller than many other goat breeds of the country. The body coat color of Assam Hill goats are white, brown, black, grey or black and white and occasional black patches on backline and legs. Ears are generally erect, medium size, horizontally placed with pointed tips. Most of the animals are having bearded and have short cylindrical horns, which are curved backwards. Sometimes with long hair have covered on their body.

The reproductive performance of Assam Hill Goat is very high and commonly known for their high prolificacy, fertility and fecundity coupled which is evident from higher percentage of twinning and even triplet, however, in many instances, quadruplets are also recorded. The birth weight of male and female kid is 1.28 and 1.13 kg respectively. Adult body weight ranges from 15 to 26kg; the average dressing percentage is 48-50% (3). The meat of this goat breed is highly delicious, more juiciness, tenderness as compared to other goat breed meat. It has noticed that all sections of the society irrespective of caste and religion of this region eat the goat meat.

The age at sexual maturity of female ranges between 7 to 8 months and age at first kidding between 13-14 months. The Service period, kidding interval of Assam

Hill goat breeds are 94.41 ± 5.41 and 247.73 ± 4.54 days respectively. Average litter size is 1.6. (4)

The average body length, body height and heart girth of this breeds are 65.21 ± 0.55 , 49.94 ± 0.40 and 56.57 ± 0.55 cm respectively.

This breed of Assam Hill goat has tremendous scopes for commercialized farming among the farmers of this region specially Assam to make a livestock entrepreneurship. Although there are some constraints particularly conservations of local germplasm due to unscientific breeding practices by the farmer's community. In this regard proper scientific intervention as well as mass awareness campaign is utmost necessary. Recently, the department of Animal husbandry and Veterinary, government of Assam has taken some initiative measure to conserve the local germplasm of this indigenous breed, for that a breeding policy of goat has been introduced in the state, where selective breeding of the Assam hill goat breed by high genetic merit animals is recommended.

References

1. 20th livestock Census. (2019). Department of Animal Husbandry, Dairying and Fisheries. Government of India.
2. <https://nbagr.icar.gov.in/en/assam-hill>
3. Assam goat breeding policy. (2019). Department of Animal husbandry and Veterinary, Government of Assam.
4. Indian Council of Agricultural Research - Annual Report (2019-20). Department of Agricultural Research and Education Ministry of Agriculture and Farmers Welfare, Government of India.



Fig 1. Assam Hill Goat Buck



Fig 2. Assam Hill Goat Doe

Clinical Approach to Diagnosis of Ophthalmic Disease in Small Animals

Mehak Mahant and Shashi Kant Mahajan
Dept. of Surgery and Radiology, COVS, GADVASU, Ludhiana, Punjab.

Introduction

For successful and satisfactory clinical results, early and accurate diagnosis of ocular disorders is necessary. This can be achieved by thorough and detailed history taking, careful physical examination and selection of specific diagnostic tests. Basic components of ophthalmic examination are history taking, hands-off examination and hand-on examination.

Protocol for Examination

- Proper order of examination should be followed; proceed from external of eye to internal, so that the fundus is examined at last.
- Both eyes and adnexa should be examined.
- Not every diagnostic procedure is required on each animal.
- Ophthalmic examination can be summarised as 'hands off' in the light, then 'hands on' in the light without any source of examination.
- Examination in light is followed by examination in the dark, using first a source of light and magnification followed by ophthalmoscopic examination.

Vision Evaluation The animal should be observed walking into examination room. A blind animal may show signs of high stepping, collision with objects or reluctance to move in strange environment. Note patient's response to hand movements, bright light or cotton ball tossed in air. (1).

Ocular Examination The examination should begin with adnexa and external ocular structures and progress to internal ocular structures so that all areas of eye are examined (1).

- 1. Orbit and Adnexa:** Evaluate the orbit for symmetry or enlargements. Presence or absence of nystagmus and strabismus is noted. Globe size and position should be noted. Eyelids are examined for abnormalities such as lagophthalmos, ptosis, trichiasis, dystichiasis, ectropian, entropian, lid neoplasms etc.
- 2. Conjunctivae, Nictitating Membrane and Sclera:** Examine both palpebral and bulbar conjunctivae for anaemia, icterus, chemosis,

foreign body, ectopic cilia etc. Note for eversion of cartilage of nictitans, prolapse of the gland, foreign bodies etc. The sclera should be evaluated for change in colour, abnormal masses and tears or laceration.

- 3. Cornea:** Normal cornea is transparent, avascular, moist and unpigmented. It should be carefully examined for loss of transparency, opacity, vascularisation, pigmentation, dryness, lacerations and ulcers.
- 4. Anterior Chamber, Iris and Lens:** Examine the anterior chamber for aqueous flare with focal light source. Iris is examined with focal light for colour, shape, pupil size and movement. The lens, which is normally transparent avascular structure, should be examined for opacities (Cataract), position, presence and size.
- 5. The Vitreous:** The vitreous humor is normally a clear gel. The anterior portion can be examined using focal illumination and posterior portion is examined by ophthalmoscopy or the slit lamp microscope.
- 6. The Fundus:** It is examined at last with direct or indirect ophthalmoscopy.

Schirmer Tear Test: This test determines whether the eyes produce sufficient tears to keep it moist. It should be done prior to application of any topical anaesthetics. The test is performed with sterile, individually packed strips of absorbable paper with notch 5 mm from one end. Each strip is folded at notch and hooked over the middle to lateral third of lower lid for 60 seconds. The distance from notch to end of the moist part of paper is noted. Normal STT values in various species:

- 1. Canines :** 15-25 mm/ min
- 2. Felines:** 18 mm/ min
- 3. Equines:** 31-32 mm/ min

Phenol Red Thread Test: In the PRT tear test the thread is 75 mm long and is impregnated with phenol red, a pH sensitive indicator. A 3 mm indentation at the end is inserted into the inferior conjunctival sac for 15 seconds. The wetted length of thread (as indicated by colour change from yellow to red) is measured in millimetres from end of thread

A. Schirmer Tear Test Strip



B. Placement of Schirmer Tear Strip



(not the bend) and is recorded in mm/seconds. In dogs the mean length of absorption using PRT tear test is 29.7-38.6 mm/15 seconds. The normal range in cats is 18.4-27.7 mm/15 seconds.

Neuro-ophthalmic Examination

1. Pupillary Light Reflex: PLR is performed in dark using a strong light source. PLR is sub cortical reflex and is not an indicator of vision. Both direct and indirect PLR are noted. An indirect PLR is reported as from the illuminated eye to the contra-lateral constricting eye. Positive indirect PLR indicates the continuity of a neurological pathway connecting the eyes. PLR is regulated by CN 2, 3.

2. Palpebral Reflex: This reflex consists of partial or complete closure of eyelids in response to touching the eyelid skin. The reflex should be tested through stimulation at both medial and lateral canthus. Afferent arm of neural arc includes sensory fibres of trigeminal nerve and efferent arm

includes motor fibres of facial nerve and muscles of eye.

3. Menace Response: It is elicited by threatening hand gesture heading towards the eye. The test should be performed by covering the contra-lateral eye. This is learned response and is not present in dogs and cats below 3-4 months of age. It is cortical response that requires the entire peripheral and central visual pathways, as well as visual cortex and facial nucleus of CN 7 to be intact (2).

4. Dazzle Response: It is manifested as partial or complete eyelid closure on the illuminated (sometimes on both sides) when bright light is directed at eye. The reflex is absent in presence of severe retinal, optic nerve, optic tract or facial nerve lesion.

5. Neuro-localization of Lesions:

- **Cerebral and Thalamic Dysfunction:** A patient with forebrain dysfunction have contralateral postural, sensory and menace deficits with normal pupil light response.
- **Brainstem (Midbrain to Medulla) Dysfunction:** Postural Dysfunction is ipsilateral to lesion Damage to CN 8 leads to nystagmus, positional strabismus. Lesions in CN 6 cause medial strabismus and failure to retract globe.
- **Cerebellum:** Lesions of cerebellum are manifested as nystagmus, positional strabismus and ipsilateral menace deficit.
- **Vestibular Disease:** Signs with peripheral vestibular disease causes nystagmus with fast phase on opposite side of head tilt and rate more than 60 beats per minute. Positional strabismus is on the same side as that of head tilt.

Ophthalmic Dye Tests:

- **Tear Film Breakup Test:** It is done to assess the stability of pre corneal tear film. Fluorescein stain is applied to cornea and eyelids are immediately closed until viewed with source of magnification and blue light source. Observe the fluorescein stained pre corneal tear film. TFBUT is the time of eyelid opening until the tear film breaks up and dark spot appears. Average TBFUT in dogs is 20 seconds and in cats is 17 seconds.
- **Fluorescein Dye Test for Corneal Ulcers:** Fluorescein dye is retained by hydrophilic stroma whenever there is loss of corneal epithelium. Intact cornea is lipophilic and doesnot allow the uptake of dye. Apply the tip of strip impregnated with fluorescein dye against bulbar conjunctiva for 2 seconds. Wash eye with ophthalmic solution and examine with

Transilluminator. Corneal ulcers stain bright green. With descemetoceles the centre of ulcer will fail to take up stain and appears black with halo.

- **Jones Test / Fluorescein Passage Test:** This test investigates lachrymal outflow through naso-lacrimal duct. The examiner instils the fluorescein into the conjunctival fornices. The appearance of stain at nares confirms patency of naso-lacrimal duct on that side(3).
- **Seidel Test:** This test assesses for the presence of aqueous humor leakage from the anterior chamber due to defect in cornea or sclera. Moisten the fluorescein dye strip with normal saline and apply above the lesion or at the superior conjunctival fornix. Visualize the eye under cobalt blue light. Seidel test is positive when fluorescein dye dilutes in the aqueous humor and causes to fluoresce bright green.

Ophthalmoscopy: It is needed to visualize posterior segment of eye (vitreous, retina, optic nerve and their associated blood vessels. There are three methods of ophthalmoscopy – direct, indirect and monocular indirect (4).

- **Direct Ophthalmoscopy:** It provides direct and upright image of fundus. There is more magnification with less field of view.
- **Indirect Ophthalmoscopy:** It provides inverted

and virtual image. The magnification is usually less, and the field of view is greater than that achieved with direct ophthalmoscope. Usually +20 D or + 30 D lens are used.

- **Monocular Indirect Ophthalmoscopy:** It provides upright image of moderate magnification and moderate field of view.

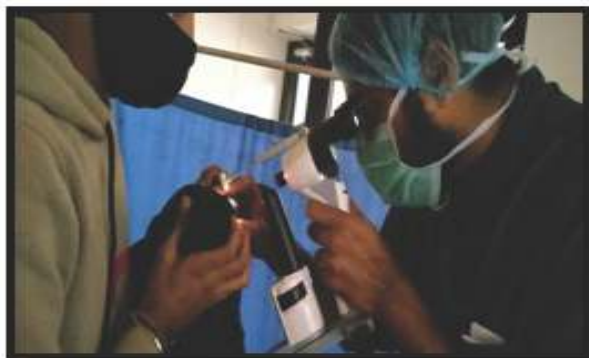
Tonometry: It is measure of intra-ocular pressure. Normal IOP in dogs is 10-20 mm of Hg. It is helpful in diagnosing glaucoma, anterior uveitis etc.

- **Indentation Tonometry:** The schiottz tonometer relies on the principle of indentation. A standard force is applied to anaesthetize cornea and the distance the rod indents the cornea is measured and is inversely related to IOP.
- **Applanation Tonometry:** The principle of applanation tonometry is that the force required to flatten a given area of sphere is directly proportional to pressure within the sphere. Most commonly used applanation tonometer is Tono-Pen.
- **Rebound Tonometry:** It ejects small probe at a fixed distance from cornea. Motion of probe is assessed as it returns to instrument after striking cornea. Eyes with higher. IOP results in shorter return time of probe

Gonioscopy: Refers to examination of iridocorneal angle utilizing corneal contact lens. The width or narrowness of the iridocorneal angle and abnormalities associated with congenital



C. Ophthalmoscopic Examination

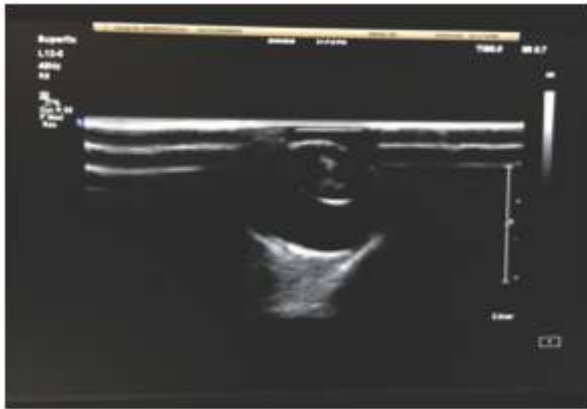


D. Measurement of Intra Ocular Pressure Using Tonopen

malformations, trauma, tumours and inflammation can be detected. This technique is helpful to differentiate between open and closed angle glaucoma (5).

Electroretinography (ERG): It measures the electric potential occurring in retina in response to light stimulation. It is commonly used to diagnose retinal disorders before cataract extraction or in cases of unexplained visual loss without any ophthalmoscopic abnormalities (6).

Ultrasonography: It is used to diagnose the conditions which lead to opacity of media (which is usually clear) that diminishes the visualization of posterior structures. B scan USG produces two dimensional cross section of eye and orbit. A scan USG assists in measurement distances within the eye. This is helpful to diagnose many conditions of eye like retinal detachment, uveitis, vitreal haemorrhage, optic neuritis, cataract etc (7).



E. Ultrasonography of Cataract

Pachymetry: It aids in measurement of corneal thickness. Corneal thickness is an indicator of corneal health. Measurement of corneal thickness helps in diagnosing corneal disorders and surgical planning.

References

1. Brooks, D. E. (2005). Ophthalmic Examination Made Ridiculously Simple. World Small Animal Veterinary Association World Proceedings.
2. Moore, P. A. (2001). Examination Techniques and Interpretation of Ophthalmic Findings. Clinical Techniques in Small Animal Practise. Vol 16, No. 1.
3. Guzek, J. P., Ching, A. S. and Hoang, T. A., et al. (1997). Clinical and radiologic lacrimal testing in patients with epiphora. 104 (11):1875.
4. Turner, A. (2010). Fundoscopic Examination: What to Look For. World Small Animal Veterinary Association World Congress Proceedings.
5. Gelatt, K. N. and Ladds, P. W. (1971). Gonioscopy in dogs and cats with glaucoma and ocular tumours. Journal of Small Animal Practise. 12: 105.
6. Lee, J., Kim, K., Jang, H., Lee, B., Kim, J. Y. and Jeong, S.(2009). The normal electroretinogram in adult healthy Shih Tzu dogs using the HMsERG. Journal of Veterinary Science. 10(3): 233.
7. Moore, D. and Lamb, C. (2007). Ocular Ultrasonography in Companion Animals: a pictorial view. In Practice. 29: 604.

Deep Stromal Corneal Ulcer in a Pug: Diagnosis and Treatment

Sabita Rakshit, Dale Clark, Kabita Roy and I C Datta

Milford Veterinary Clinic, 110 Canal Street, Milford, USA and College of Veterinary Science (NDVSU), Jabalpur, Madhya Pradesh.

Abstract

A young spayed female Pug was presented to the Milford Veterinary Clinic with visible inflammation in the left eye. On physical examination, all the vitals were within the normal limits. Occasional squinting on the affected eye was noticed by the owner for the past 4-5 days. The case was immediately referred to an ophthalmologist after palliative treatment. The patient's eye problem was diagnosed with fluorescein stain test as deep corneal stromal ulcer in the left eye, related to the highly deleterious ectopic cilia in the upper eye lid. Specialized medical treatment was given in the Ophthalmology specialist's centre with detailed advisory on discharge. Constant use of protective neck collar to preempt traumatic eye injury till the recheck was emphasized. Progressive improvement in the eye health status is highly encouraging.

Introduction

In companion dogs a corneal ulcer, superficial or deep discontinuity in the corneal surface, is one of the most common eye disorders (1) and a major cause of blindness (2). The most common agent is traumatic insult. Several other factors are also involved. These include entropion and distichiasis, conditions in which one or more eyelashes rub against the cornea. Damaged sensory nerves in the eyelids resulting in impaired blink reflexes may culminate in ulcerated cornea (3).

In keratoconjunctivitis sicca, the dry unhealthy cornea may become ulcerated. Ocular microbial infections (bacterial, viral, fungal) may cause painful corneal ulcer(4). Squinting (blepharospasm) and reddish affected eye are often observed(5).



Occasionally, neurotropic factors, e.g. facial or trigeminal nerve paralysis may be involved (6). Excessive lacrimation is a common clinical symptom. Any eye injury should be treated as an emergency mandating (i) urgent primary veterinary care (ii) well-judged referral to veterinary ophthalmologist for specialized treatment. The superficial corneal ulcers may heal uneventfully

without medical aid. However, if the ulcer is deep, the treatment time lag is prolonged, and significant infection is present, vision may be affected with progressive corneal scarring, thinning and occasional perforation, as in the case under report. Genetic predilection, closely linked to the craniofacial geometry and eye lid conformation (7, 8) is a high-risk factor. The highest prevalence of corneal ulcer in a retrospective study in the United Kingdom was recorded in Pug, Boxer, Shih Tzu, Spaniel, and Bulldog (9).

Case History

A young spayed female Pug, a little above one year in age, was presented to the Milford Veterinary Clinic on February 16, 2021 with an inflammation in her left eye. Anamnesis revealed occasional squinting on the affected eye, noticed by the owner during the past 4-5 days. The patient exhibited discomfort, attempting to lick her genitalia. Physical examination showed all vitals within the normal limits: rectal temperature 101.9° F, heartrate 96 beats/ minute, respiration rate 34/ minute, capillary refill time (CRT) <2 seconds, visible mucous membranes pink, and body condition score (BCS) 3/5. The conspicuous 'hole' in the cornea resembled a descemetocoele (**Fig. 1a,b**).



Fig. 1a. Note the "hole" in the left eye

Fig. 1b. Encircled Descemetocoele

A drop of proparacaine (local anaesthetic) was administered in the affected left eye, which immediately relieved the squinting reflex. Urgently needed primary medical care: analgesic Meloxicam [5mg/ml] @ 0.2 mg/kg was injected S/C. Broad-spectrum topical eye antibiotic, Tobramycin @ 2 drops BID into the affected eye, was dispensed for home use for 10 days. The well-briefed owner was advised referral to the veterinary ophthalmologist for advanced diagnosis and treatment. In the home clinic, MVC the patient's turgid anal glands were carefully expressed manually. Meloxicam oral suspension 1.5 mg/ml was dispensed to be given daily with food for pain relief.

Ophthalmology Specialist's Report

On 18th February 2021 the patient's eye problem was diagnosed as corneal deep stromal ulcer. The eye pressure Oculus Dexter (OD), right side eye 13mm Hg, and Oculus Sinister (OS), left side eye 9mmHg. Fluorescein stain test: OD-ve, OS +ve. Ectopic cilia: present in the upper eye lids, both eyes.

Ophthalmology specialist's prescribed home medications: Ofloxacin ophthalmic 0.3% solution 5 ml: apply 1 drop in the affected left eye 4 times a day, until the reddish tinge disappears. Atropine 1% ophthalmic solution 2 ml: apply 1 drop in the left eye (OS) for 5 days, then discontinue. Doxycycline 50 mg tablet: give 1 tablet BID, PO with food for 14 days. Gabapentin 100 mg capsules: 1 capsule BID, PO with food for pain relief. Meloxicam oral suspension 1.5 mg/ml, to continue as directed. Protective neck collar mandatory, until recheck.

Specialist's Comment

The dog patient, Nora has developed a deep stromal corneal ulcer in the left eye, presumably because of patchy loss of epithelial lining cells by the bacterial proteolytic enzymes. Unrestricted deeper perforation may lead to blindness from draining of the ocular fluid. Definitely, the problem originated from the two ectopic cilia in the upper eyelid, penetrating the inner conjunctival lining, inducing recurrent episodes of corneal ulceration, pigmentation and scar tissue formation.

The ectopic cilia are epilated, and the initial medical treatment aims to restrict the resulting ulceration. Ofloxacin, penetrating deep into the corneal stroma, is preferred over Tobramycin to eliminate the primary aetiology: pathogenic bacterial population. Oral administration of Doxycycline is aimed to prevent further stromal collagen degeneration.

I. To do

1. Seek veterinary attention immediately if your dog suffers an eye injury. Delays in treatment may lead to irreparable damage to the eye.
2. Use simple measures to prevent corneal ulcers. If you bathe your dog at home, ask your veterinarian to recommend a protective ointment that you can place in the pet's eyes beforehand to avoid irritation of the cornea. Inform your veterinarian if the pet has been on any medications.
3. Give medication precisely, as directed, and if you are concerned about any suspected adverse side effects, discuss with your veterinarian immediately. Seek a second opinion if the condition is not improving/resolving.
4. Watch for signs of illness: lethargy, weakness, decreased appetite, hiding more than usual, aggressiveness, and other behavioral changes. Signs of problems with the eye(s): increased tear production, squinting, red eyes, swollen eyelids, itching/pawing the eyes, drainage of white or greenish material from the eye, and acting in pain or defensive posture, when the area around the eye is touched.

II. Not to do

1. Do not permit your pet to peep out from the window of the moving motor vehicle.
2. Do not remove the protective Elizabethan neck collar on your own, without consulting the veterinarian.
3. Do not attempt to remove any foreign body lodged inside the pet's eye yourself; it could be damaging.
4. Do not apply any topical medicines in the affected eye, not intended for ophthalmic use.

Useful tips to dog owners

Caution

- (i) No attempt should be made to open the eyelids manually to administer the topical medicines. Instead, tip the patient's nose upward and allow only one drop to fall into the eye gently, at one time.
- (ii) Never touch the tip of the antibiotic bottle with the eye surface, as it will contaminate the contents.

Home Care Advisory

Elizabethan or lampshade collar is recommended to prevent the chances of self-inflicted trauma. Though superficial, uninfected corneal ulcers generally heal uneventfully, deep ulcer (instant case), or persistent/ 'indolent' ulcers require long-

term treatment with scheduled visits to the clinic.

Discussion

A brief resume of the complex biomechanisms, responsible for induction of corneal ulcer will be pertinent. Matrix metalloproteinases (MMPs) group of zinc-dependent extracellular endoproteinases is importantly involved in corneal ulcer pathogenesis in humans and animals (10). Originating from the corneal fibroblasts and the supportive epithelium, MMP-2 and MMP-9 are the primary degrading agents (11, 12). A strong functional refractive lens is supported by a combination of corneal and pre-corneal tear film. The corneal extra-cellular matrix (ECM) is stabilized by well-balanced collagen and ECM synthesis and concurrent degradation by the proteinases, responsible for constant surveillance and scavenging of non-functional/ dead epithelial cells (13-15). The MMPs play a key role in wound healing, but their over expression is highly deleterious because of accelerated ECM degradation with tissue destruction, and eventual irreparable loss of vision (16).

In pet practice, the clinician must make a sincere attempt to evaluate the precise cause of the corneal ulcer, and the best remedial strategy. To treat bacterial infections, topical ophthalmic antibiotic preparation (Drops or ointment) needs to be placed on the eye surface at the specified intervals daily. Depending on the cause, topical atropine is given to control pain associated with inflammation, and to dilate the pupil to prevent possible intraocular adhesions (bridging scars, which hamper the normal optical function). To control ocular inflammation and effective pain management, oral anti-inflammatory drug may be prescribed. Dogs with rapidly developing deep ulcers, or ruptured eyes may also require treatment with systemic antibiotics and/ or ocular surgery. A commonly employed surgical procedure involves creating a conjunctival flap, a small section of the most superficial layer of the white region of the eye. The flap is auto grafted into a new site overlying the ulcer, with the patient under general anaesthesia. The micro-capillaries in the flap promote the ulcer healing process. The eyelids may be partly sutured/stitched to serve as a natural protective barrier, usually for a few weeks. The sutures are removed carefully, and the eye resumes its normal geometric configuration and function. Novel clinical application of regenerative therapy, using subconjunctival injection of autologous platelet-rich plasma (17) is a promising biomedical innovation. Topical application of cross-linked thiolated carboxymethylated hyaluronic acid hydrogel, abbreviated xCMHA-S (0.75% solution in phosphate buffer saline, pH 7.4) has proved highly beneficial, promoting accelerated closure of corneal epithelial

defects and repair of stromal 'indolent' ulcers (18). In the instant case, timely referral to the ophthalmologist promoted conventional stromal ulcer healing without novel intraocular medicaments.

After complete healing of the corneal ulcer surgical resection of the hair and follicle was followed by cryosurgery of the surrounding tissue. This procedure was adopted to totally eliminate the chance of re-growth of the deleterious ectopic cilia. The patient recovered completely, as per the regular updates from the highly satisfied owner.

References

1. Gilger, B.C. (2007). Ed. Diseases and surgery of the cornea, 4th edn. Oxford Blackwell Publishing. pp. 690.
2. Startup, F.G. (1984). Corneal ulceration in the dog. *J. Small Anim. Pract.* 3: 367.
3. Slatter, D. (2001). Ed. *Eyelids*, 3rd edn. Saunders, Philadelphia, USA.
4. Marfurt, C.F., Murphy, C.J., and Flarczak, J.L. (2001). Morphology and neurochemistry of canine corneal innervations. *Invest. Ophthalmol. Vis. Sci.* 42: 2242.
5. Chidambaram, J. (2007). Recent advances in the diagnosis and management of bacterial keratitis. *Int. Ophthalmol. Clin.* 47: 1.
6. Stanley, R.G. (2007). Management of corneal ulcers in small animals. In: *Proc. World Small Animal Veterinary Association (WSAVA)*, 32nd World Congress, Sydney, Australia, August 19-23.
7. Barnett, K.C., Samson, J. and Heinrich, C. (2002). *Canine Ophthalmology: An Atlas and Text*, Saunders, Philadelphia, USA.
8. Packer, R.M.A., Hendricks, A. and Bum, C.C. (2015). Impact of facial conformation on canine health: corneal ulceration. *PLoS ONE* 10 (5): e0123827. <https://doi.org/10.1371/journal.pone.0123827>
9. O'Neill, D.G., Lee, M.M., Brodbelt, D.C., Church, D.G. and Sanchez, R.F. (2017). Corneal ulcerative diseases in dogs under primary veterinary care in England: epidemiology and clinical management. *Canine Genet. Epidemiol.* 4: 1.
10. Li, D.Q., Shang, T.Y., Kim, H.S., Solomon, A., Lokeswar, B.L. and Plugfelder, S.C. (2003). Regulated expression of collagenases MMP-1, -8, and -13 and stromelysins MMP-3, -10 and -11 by human corneal epithelial cells. *Invest. Ophthalmol. Vis. Sci.* 44: 2428.

11. Fini, M.E., Girard, M.T., Matsubara, M. and Bartlett, J.D. (1995). Unique regulation of the matrix metalloproteinase, gelatinase B. *Invest. Ophthalmol. Vis. Sci.* 36: 622.
12. Meller, D., Li, D.Q., and Tseng, S.C. (2000). Regulation of collagenase, stromelysin, and gelatinase B. in human conjunctival and conjunctivochalasis fibroblasts by interleukin-1 β and tumor necrosis factor- α . *Invest. Ophthalmol. Vis. Sci.* 41: 2922.
13. Matsubara, M., Girard, M.T., Kublin, C.L., Cintron, C. and Fini, M.E. (1991). Differential roles for two gelatinolytic enzymes of the matrix metalloproteinase family in the remodeling cornea. *Dev. Biol.* 147: 425.
14. Arican, M. and Ceylan, C. (1999). Metalloproteinases in canine experimental traumatic keratoconjunctivitis. *Zentralblatt Veterinarmed.* 46: 527.
15. Wong, T.T.L., Sethi, C., Daniels, J.T., Limb, G.A., Murphy, G. and Khaw, P.T. (2002). Matrix metalloproteinases in disease and repair processes in the anterior segment. *Surv. Ophthalmol.* 47: 239-256.
16. Birkedal-Hansen, H. (1988). From tadpole collagenase to a family of matrix metalloproteinases. *J. Oral Pathol.*, 17: 445-451.
17. Farghali, H.A., AbdElkader, N.A., AbuBakr, H.O., Ramadan, E.S., Khattab, M.S., Salem, N.Y. and Emam, I.A. (2021). Corneal ulcer in dogs and cats: Novel clinical application of regenerative therapy using subconjunctival injection of autologous platelet-rich plasma. *Front. Vet. Sci.* 8: 641265. Doi:10.3389/fvets.2021.641265
18. Williams, D.L., Wirostko, B.M., Gum, G. and Mann, B.K. (2017). Topical cross-linked HA-based hydrogel accelerates closure of corneal epithelial defects and repair of stromal ulceration in companion animals. *Invest. Ophthalmol. Vis. Sci.* 58: 4616-4622.

Successful Surgical Treatment of Interdigital Follicular Cyst in a Dog

Sabita Rakshit, Dale Clark, Kabita Roy and I C Datta

Milford Veterinary Clinic, 110 Canal Street, Milford, USA and College of Veterinary Science & Animal Husbandry (NDVSU), Jabalpur, Madhya Pradesh.

Abstract

Sammy Vincent, a 13-year old spayed female pug, weighing 11.5kg was presented for treatment of a persistent swelling in the two lateral digits of the left front paw. Anamnesis revealed waxing and waning pattern of the oedematous mass/ cyst over the past several months, not amenable to home treatment. On proper briefing, the owner was convinced about the best viable option: surgical excision of the entire biodegraded mass with curative intent. The in-house regional survey radiographs did not reveal any signs of osteolysis, or traumatic injury. Accordingly, as per the schedule, the patient was admitted in the operation theater, and the entire surgical procedure was conducted under adequate antibiotic cover. The recovery, with proper home care support, was uneventful, and the final recheck in the clinic did not reveal any evidence of recurrence 24 weeks post-surgery.

Introduction

Interdigital follicular cysts in dogs are large abnormal bumps, or nodules that develop between adjacent digits in the toe. Also named interdigital furuncles, or follicular pododermatitis these cutaneous lesions, often appear as shiny red nodules, engorged with blood and/or pus. The affected dog may limp with pain, especially while negotiating an uneven surface, and also lick or chew at the adnexal area. The pathoclinical syndrome is multi-factorial in origin. A critical appraisal of the published case reports revealed that the genetic profile, body conformation, and surface skin geometry are significantly involved. Thus, dog breeds with a short and coarse hair coat: English bull dog, Great Dane, Basset hound, Mastiff, Bull terrier and Boxer are at increased risk to develop inflamed, open follicles. Further, dogs with wider paws: Labrador retriever, English bulldog, German shepherd and Pekingese are more likely to bear the body weight on the haired skin patch between the pads. Dogs with obesity, arthritis, or other joint complications, or with abnormal behaviour like licking, or chewing are more susceptible (1). The paw lesion may exhibit pruritis, alopecia, ulceration, paw pad involvement, comedones and serosanguineous/ seropurulent exudates. The foot may be conspicuously oedematous, and the adjacent skin area may appear visibly moist because of constant licking episodes and associated infection (2). In perspective, the differential diagnosis includes systematic evaluation of (i) microbial infection (bacterial/ fungal), (ii) hypersensitivity, (iii) immune-mediated disorder, (iv) endocrine imbalance, (v) traumatic insult, (vi) genetic predisposition, (vii) metabolic disorders, and (viii) neoplastic



transformation (3-8). In pet practice, the most common aetiology factors are allergic pododermatitis, and superimposed bacterial/ mycotic infection, parasitic pododermatitis, especially Demodex mite infestation (9).

Case History

Dog patient, Sammy Vincent a 13-year spayed female pug weighing 11.5 kg was presented in the clinic on January 13, 2021 with a swelling between the two lateral digits of the left front paw (Fig. on top left). Anamnesis revealed that the swelling was noticed intermittently over the past few months, sometimes with a crusty scab on top of the paw, not bothering the patient. The owner was cleaning it with hydrogen peroxide solution and applying topical Neosporin. All the vitals were within normal limits, rectal temperature 100.8oF, heart rate 96 bpm, respiration rate not recorded because of panting, mucous membranes pink, and body condition score (BCS) 3.5/5.

Physical examination

An interdigital cyst in the left front paw between the lateral two digits. Further, a solitary skin tag in the neck area, and one growth in the right groin area, were noticed, possibly both lipomas. A fine needle aspirate was suggested, but the owner declined. Owner was briefed on the interdigital cyst removal. To prevent the bleeding cyst to become infected, Penicillin-G [300,000 units/ml] @ 24,000 units/kg, 1.0 ml was injected S/C. Cefpodoxime @ 5-10 mg/kg; 100mg tablets were dispensed, one-tab OD with food for 10 days.

The dog patient, fasted overnight, was brought to the clinic in the early hours of February 17, 2021 for the growth removal surgery. In-house regional radiographs (**Fig. 1**) did not reveal any signs of bone infection.



Fig 1. Survey radiographs of both front limbs.

Pre-surgery Preparation

The patient was sedated with acepromazine @ 0.025-0.2 mg/kg [10mg/ml] 0.1 ml + butorphanol tartrate (Torbugesic®, Zoetis US, Inc.) @ 0.2 mg/kg [10mg/ml] 0.5 ml, injected S/C. IV catheter was inserted into the right cephalic vein and lactate Ringer's solution drip was started @ 50ml/ hr. After 30 minutes, anaesthesia was induced with ketamine @ 2-10 mg/kg [50 mg/ml], and midazolam @ 0.1-0.3 mg/kg [5mg/ml] 0.5 ml each, injected I/V. After intubation, the dog was transferred to isoflurane gas anaesthesia, hooked to the auto-monitoring systems for EKG, blood pressure, O2 levels and pulse rate. Isoflurane gas is generally maintained at 2.5% flow rate. However, the flow rate is suitably adjusted, depending on the patient's reflexes.

Surgery

For excision of the cyst in between the digits

number 4 and 5 on left front paw, the incision was made starting from the base of the digit with #10 scalpel blade, proceeding towards the nail, around the cyst with as wide margins as possible, virtually 'U'-shape format with the widest possible margins. The cyst was removed completely with a pair of Metzenbaum scissors. Hemostasis of the bleeders was ensured with electrocautery. The subcutaneous layer was closed with 4-0 absorbable sutures, and skin with non-absorbable 3-0 sutures.



Fig 2. Surgery in progress



Fig 3. Suturing completed uneventfully.

Post-Surgery Medical Care

Antibiotic Penicillin-G [300,000 units/ml] @ 24,000 units/kg; 1.0 ml was injected S/C, and analgesic Carprofen [50mg/ml] (Rimadyl®) @ 2.2-4.4 mg/kg, 0.5ml was injected S/C. Take home antibiotics: Cefpodoxime @ 5-10 mg/kg, 100mg



Fig 4. Surgical wound on day 7.

tablets:one-tab OD with food for 10 days, and Rimadyl 25mg BID with food for 5 days were dispensed with advisory.

Discussion

Dermatologic tests: Examination of deep skin scrapings, or hair plucks, skin cytology, bacterial culture, and fungal culture may be advised (9). Direct impression cytology of the final needle aspirate (FNA) sample provides valuable information on the presence of infiltrating neutrophils, other inflammatory cells, and phagocytosed bacteria (2, 10). Fungal hyphae may also be noticed (2). Skin scrapings and hair plucks may be used in the diagnosis of parasitic pododermatitis. Need-based microbial culture and sensitivity tests are advised for antibiotic therapy. However, this contingency did not arise in the instant case. If the underlying causes are not diagnosed and treated in time, the pathobioentity may return and induce scarring with impaired locomotion. The habitat also needs to be checked. The front paws are commonly targeted in the skinfold between digit numbers 4 and 5 (as in the instant case), and the efficacy of curative surgery is on record (2).

References

1. The VIN Dermatology Consultants (2018). Interdigital cysts in dogs. Veterinary Partner®-VIN.
2. Miller, W. H., Griffin, C. E., Campbell, K. L. (2013). Muller and Kirk's Small Animal Dermatology, 7th edn. St. Louis, Missouri, Elsevier. pp. 201.
3. Hnilica, K. A. (2011). Small Animal Dermatology: A Color Atlas and Therapeutic Guide, 3rd edn. St. Louis, Missouri, Elsevier Saunders. pp. 60.
4. Miller, W. H., Griffin, C. E., Campbell, K. L. (2013). Muller and Kirk's Small Animal Dermatology, 7th edn. St. Louis, Missouri, Elsevier Saunders. pp. 106.
5. Duclos, D. D. (2013). Canine pododermatitis. Vet. Clin. Small Anim. Pract. 43: 57.
6. Saridomichelakis, M. N., Koutinas, A. F., Farmaki, R., Leontides, L. S. and Kasbalis, D. (2007). Relative sensitivity of hair pluckings and exudates microscopy for the diagnosis of canine demodicosis. Vet. Dermatol. 18: 138.
7. Gross, T. L., Ihrke, P. J., Walder E. J., and Affolter, V. K. (2005). Skin Diseases of the Dog and Cat-Clinical and Histopathological Diagnosis. 2nd edn. Hoboken, New Jersey, Wiley-Blackwell. pp. 9.
8. Breathnach, R. M., Fanning, S., Mulcahy, G., Bassett, H. F., Jones, B. R. and Daly, P. (2006). Evaluation of Th1-like, Th2-like and immunoregulatory cytokine mRNA expression in the skin of dogs with immunomodulatory-responsive lymphocytic-plasmacytic pododermatitis. Vet. Dermatol. 17: 313.
9. Bajwa, J. (2016). Canine pododermatitis. The Canadian Vet. J. 57 (9): 991.
10. Duclos, D. D., Hargis, A. M., and Hanley, P.W. (2008). Pathogenesis of canine interdigital palmar and plantar comedones and follicular cysts, and their response to laser surgery. Vet. Dermatol. 19: 134.

Surgical Management of Urinary Bladder Calculus in a Female Maltese Dog

Sabita Rakshit, Dale Clark, Brianna Petrylak, Kabita Roy and I C Datta

Milford Veterinary Clinic, 110 Canal Street, Milford, USA and College of Veterinary Science & Animal Husbandry (NDVSU), Jabalpur, Madhya Pradesh.

Abstract

A young spayed female Maltese dog was presented to the Milford Veterinary Clinic with the complaint of frequent haematuria and dysuria. The patient was seen in a different clinic for suspected bladder stone and was kept on prescription food. The owner wanted other options to resolve the problem. The patient's haemato-biochemical profile was virtually normal. Urinalysis and abdominal survey radiography confirmed the existence of a large bladder stone. Successful cystotomy was performed in the clinic on November 11, 2019, and the submission to the Minnesota Urolith Center revealed the chemical identity: magnesium ammonium phosphate (struvite) and calcium oxalate carbonate. The recovery was remarkably fast: virtual normalcy restored on day 1 post-surgery. The owner was advised therapeutic diet, Hill's S/D life-long to preempt recurrence of bladder stone and associated red urine in the dog.



Introduction

Uroliths, Calculi or 'bladder stones' originate from minerals, clumped into a solitary big mass, or a clustered mass of varying size, shape and chemical composition in different parts of the urinary system. In dogs, the most common forms are struvite (magnesium ammonium phosphate, MAP) and calcium oxalate.

The clinical signs include blood-stained, or discoloured pungent urine, polyuria, inconsistency, abnormal licking of the genitalia, lethargy, anorexia, and vomiting. Most bladder stones are identified through abdominal survey radiography and/ or ultrasonography. Need-based urine analysis, microbial culture and antibiotic sensitivity test may be recommended (1). Haematuria is the leading clinical sign of urolithiasis (2).

According to the Veterinary Information Network (USA), 85% of dogs with struvite uroliths are young females (average age 2.5 years), and Shih-tzu, Schnauzer, Yorkshire Terrier, Labrador Retriever and Dachshund are at increased risk. Special dog food, formulated to promote accelerated dissolution of the stones, e.g., Hill's S/D is recommended. Antibiotic support is also advised in concurrent

UTI. Lithotripsy, using high frequency sound waves to break up stones in situ is a less invasive, innovative option. Surgical removal of the bladder stone is the last resort in life-threatening urinary obstruction (3).

Calcium oxalate uroliths are more common in the male dogs, especially in the higher age group (average 9.3 years); some breeds, e.g., Norfolk terrier and Pomeranian are at increased risk. Change of the dietary regimen is not effective in dissolving calcium oxalate stones. These can be removed through surgical intervention, or the less invasive lithotripsy / urohydropropulsion technique. Minnesota Urolith Center is a reputed agency for obtaining definitive information on the diagnosis and treatment options on bladder stones in the dog patients in the USA (1).

Case History

Bianca Olinzock, 10-year-old female spayed Maltese dog (4.7 kg,) was presented to the Milford Veterinary Clinic on October 7, 2019 for the treatment of frequent blood-stained urination. The patient was kept on the Hill's prescription S/D diet by some other clinic, to dissolve the harmful uroliths (1). Abdominal survey radiograph revealed a solitary large bladder.

All the vitals were within normal limits, rectal temperature 102.4° F, heart rate 132 beats/ minute, respiration rate not recorded because of panting, capillary refill time (CRT) <2 seconds, visible mucous membranes pink, and body condition score (BCS) 3/5. The patient on assured biosafety, based on the haemato-biochemical profile (**Tables 1 and 2**) was put on broad-spectrum antibiotic, Penicillin G @ 24,000 U/kg, injected S/C in the clinic, followed

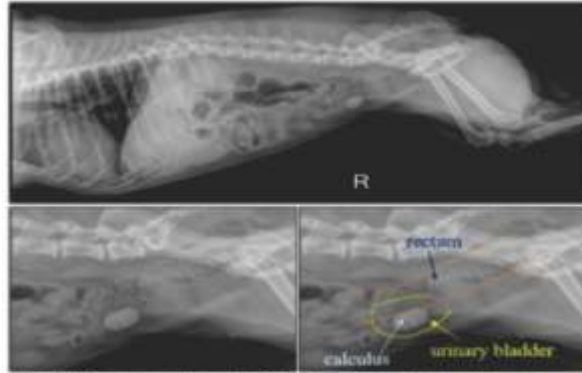


Fig. 1. RL survey radiograph: Note the solitary calculus (above), closeup (below left) and outlined (below right).

by home care oral medication by the well-briefed owner: Clavamox @ 10 mg/ kg; 62.5 mg 1 tablet BID for 10 days prior to cystotomy.

Table 1. Patient's haemogram on 8.10.2019

Parameters (Units)	Value	Reference Interval	Status
TEC (1x106/ μ L)	7.01	5.65-8.87	Normal
Hematocrit (%)	49.8	37.3-61.7	Normal
Hemoglobin (g/dL)	16.9	13.1-20.5	Normal
MCV (fL)	71.0	61.6-73.5	Normal
MCH (pg)	24.1	21.2-25.9	Normal
MCHC (%)	33.9	32.0-37.9	Normal
RDW (%)	15.9	13.8-21.7	Normal
Reticulocyte (1x103/ μ L)	135.3	10.0- 10.0	High
Reticulocyte Hemoglobin(pg)	24.8	22.3- 29.6	Normal
TLC (1x 103/ μ L)	12.86	5.05-16.76	Normal
Neutrophil (%)	76.6		
Lymphocyte (%)	10.4		
Monocyte (%)	6.1		
Eosinophil (%)	5.6		
Basophil (%)	1.3		
Neutrophil (1x103/ μ L)	9.85	2.95-11.64	Normal
Lymphocyte(1x103/ μ L)	1.34	1.05-5.10	Normal
Monocyte (1x103/ μ L)	0.78	0.16-1.12	Normal
Eosinophil (1x103/ μ L)	0.72	0.06-1.23	Normal
Basophil (1x103/ μ L)	0.17	0.00-0.10	High
Thrombocyte (1x103/ μ L)	481	148-484	Normal

Auto CBC Analyzer; Blood Chemistry Auto Analyzer

Surgery

I. Preparation of the patient: Premedication was done with butorphanol [10 mg/ml] @ 0.2 mg/kg, 0.09 ml and acepromazine [10 mg/ml] @ 0.025- 0.2 mg/kg, 0.04ml, injected S/C. Sterile lactate Ringer's solution was infused with the I/V catheter @ 200 ml/hr. Anaesthesia was induced I/V with ketamine [50 mg/ml] @ 2-10 mg/kg, and midazolam [5mg/ml] @

Table 2. Patient's blood chemistry profile on 8.10. 2019.

Parameters (Units)	Value	Reference Interval	Status
Glucose (mg/dL)	132	74-143	Normal
SDMA (μ g/dL)	9	0-14	Normal
Creatinine (mg/dL)	0.8	0.5-1.8	Normal
BUN (mg/dL)	14	7-27	Normal
BUN/ Creatinine ratio	18		
Phosphate (mg/dL)	3.3	2.5-6.8	Normal
Total calcium (mg/dL)	9.6	7.9-12.0	Normal
Total protein (g/dL)	7.4	5.2-8.2	Normal
Albumin (g/dL)	3.3	2.3-4.0	Normal
Globulin (g/dL)	4.1	2.5-4.5	Normal
A/G ratio	0.8		
ALT (U/L)	114	10.-125	Normal
ALP (U/L)	60	23-212	Normal
GGT (U/L)	0	0-11	Normal
Amylase (U/L)	415	500-1500	Low
Lipase (U/L)	338	200-1800	Normal
Total Bilirubin(mg/dL)	0.8	0.0-0.9	Normal
Cholesterol (mg/dL)	273	110-320	Normal
Na+ (mmol/L)	152	144-160	Normal
K+ (mmol/L)	4.7	3.5-5.8	Normal
Na/K	32		
Cl- (mmol/L)	119	109-122	Normal

Blood Chemistry Autoanalyzer

Table 3. Urinalysis* on 7.10. 2019.

Parameter		Inference
Color	reddish	Haematuria
Clarity	Cloudy	
Specific Gravity	1.025	High
pH value	8.0	High*
Total Protein (mg/dL)	500	mg/dL
Glucose (mg/dL)	Negative	mg/dL
Ketone bodies (mg/dL)	Negative	mg/Dl
Bilirubin (mg/dL)	1	mg/Dl
Urobilinogen (mg/dL)	4	mg/Dl
Erythrocytes (cell count/ μ L)	250	Haematuria
Leucocytes (cell count/ μ L)	100	Haematuria

Comments: 1. * Table top sample of urine

0.1-0.3 mg/kg, 0.25 ml each, and the patient was transferred to isoflurane gas, hooked to automonitoring of the vital body functions. The operation site on the caudal abdomen was clipped, surgically scrubbed and sanitized.

II. Laparotomy-Cystotomy: In the suitably prepared abdominal site, a midline incision was made with #10 sterile scalpel blade, caudally from

the umbilicus to the pubis. The urinary bladder was carefully exteriorized (**Fig. 2a**). Sterile lap sponges were planted around the bladder. Care was taken to prevent the contents of the bladder seeping into the open abdominal cavity. Two stay sutures were placed on the bladder for the technician to assist the surgeon conveniently. Urine was drained out completely with a syringe and needle (cystocentesis). With a stab incision into the empty bladder a large calculus / stone (**Fig. 2b**) was cautiously retrieved.

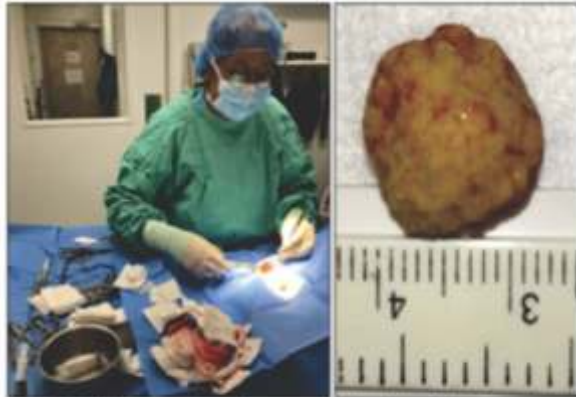


Fig. 2a. Bladder exteriorized

Fig. 2b. calculus

A urinary catheter was passed from the bladder to flush the lower chamber with normal saline solution, passing into the urethra to wash out any residual small uroliths, till clear fluid was seen. The mucosal lining of the bladder was perceptibly inflamed. The visible blood clots were removed manually, and the remaining flushed out. The bladder was closed tightly with two layers of 3-0 absorbable sutures, scrupulously avoiding contact with the mucosal lining. The first layer is a simple continuous pattern, and the second either Cushing or Lambert pattern, inverting to oversee the first layer. A leak test was carried out by slowly pushing normal saline solution into the bladder and squeezing gently to locate any seepage at the incision site. After removing all the counted lap sponges, the bladder was carefully replaced into the abdomen. The Linea alba was closed with 3-0 absorbable sutures in the simple continuous pattern. The subcutaneous tissues were closed with 3-0 absorbable sutures in the simple continuous pattern. The cutaneous layer was closed with non-absorbable 3-0 suture in the simple interrupted cruciate pattern. Complete removal of stones was reflected in the post-operative radiographs (**Fig. 3**). The owner was briefed on the harmless presence of blood in the urine for 2-3 days.

Post-operative Protocol

Penicillin G injection @ 24,000U /kg and pain medication, Meloxicam 5mg/ ml] @ 0.2 mg/kg 0.2

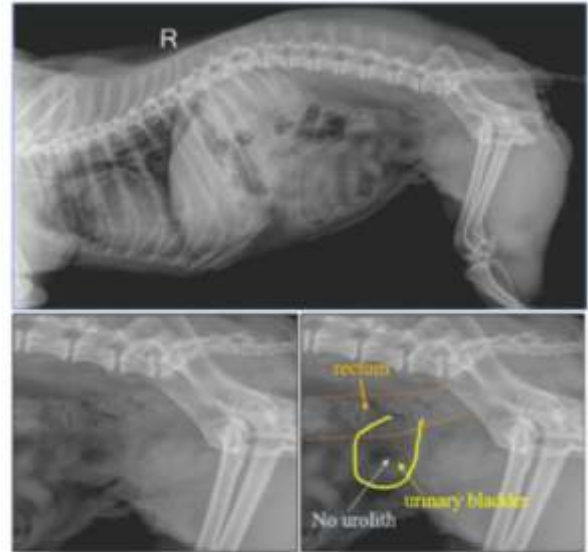


Fig. 3. post surgery radiograph: Note no calculus/ urolith (above), closeup (below left) and outlined (below right).

ml, injected S/C. Take home antibiotics: Clavamox® 62.5 mg tablet @ 10 mg/ kg dose, 1 tablet two times every day for 10 days, and Meloxicam oral with food once a day were dispensed. Home care advisory: constant use of e-collar and restricted movements for at least 10-14 days. On day 1 post-surgery the pet was doing fine. The sutures were removed on day 14.

Discussion

Chemical composition, size, location, numbers, and surface morphology of the uroliths are of much clinical concern in pet practice. Food, genotype and gender of the companion dog are importantly involved. Notably, magnesium ammonium phosphate (struvite) uroliths, more common in the younger females, result from unabated bacterial infection in the bladder, causing abnormal release of ammonia from degradation of urea. On the other hand, calcium oxalate uroliths, most common in the aged male dogs, are formed because of excessive urinary oxalate formation in renal dysfunction. Dalmatians with genetic deficiency of uricase enzyme in the liver and kidney are prone to hyperuricemia with stone formation. In portosystemic shunt, a congenital vascular anomaly, the liver cells are unable to clear uric acid from the blood circulation, resulting in significantly increased incidence of urolithiasis. Partial or complete urinary obstruction in multi-factorial urolithiasis in dogs mandates early definitive diagnosis for effective surgical and/or medical therapy (3-5). Haematuria, stranguria (difficult urination), and oliguria are the common signs of lower urinary tract disease, not specific to urocystolithiasis (6). These stones are difficult to localize through digital palpation. On presentation, the patient's signalment, case history, clinico-

haemato-biochemical profile, and urinalysis report are not conclusive, and only diagnostic imaging is definitive (2).

Abdominal survey radiography, in combination with ultrasonography is often used for the diagnosis and localization of uroliths in the urinary tract. Contrast radiography may be required to detect non-radiopaque uroliths (7). Ultrasound scan reveals a clear hyperechoic area with acoustic shadows in the bladder stones (8). Radiography remains the basic imaging tool, covering the entire urinary tract, and also the complete length of the urethra to identify and locate the radiopaque stones (6). Surgical excision of the deleterious mineral masses along with medical management is the best option (9).

In the instant case, cystotomy under broad-spectrum antibiotic cover, conforming to the standard protocol was effective, evidenced by the fast recovery within two hours post-surgery (photograph on top of the title). On day 1 post-surgery Bianca Olinzock appeared comfortable: eating, drinking, and performing other body functions normally. The skin sutures from the healed surgical wound were removed on day 14 post-surgery. Hill's S/D prescription food was recommended for life-long use for prevention of recurrence of uroliths and the associated haematuria. Minnesota Urolith Center report on the chemical analysis of the referred bladder stone, received on 21.11.2019 established magnesium ammonium phosphate (struvite), and calcium phosphate carbonate mineral composition. The relevance of client education on home food management with optimized protein content is highlighted in our earlier communication (10).

References

1. Wooten, S.J. (2020). Bladder stones in dogs: types, symptoms and treatment. Hill's Pet. <https://www.hillspet.com/dog-case-/healthcare/bladder-stones-in-dogs>
2. Forms, A. and Saini, N.S. (2015). Canine urolithiasis and concurrent urinary bladder

- abnormalities: symptoms, haematology and comparative radiographic and ultrasonographic diagnosis. *Vet. Med. Open J.* 4 (1): 18. Doi:17140/VMOJ-4-132.
3. Lulich, J.P., Berent, A.C., Adams, L.G., Westropp, J.L., Bartegs, J.W. and Osborne, C.A. (2016). American College of Veterinary Internal Medicine (ACVIM) Consensus recommendations on the treatment and prevention of uroliths in dogs and cats. *J. Vet. Int. Mede.* 30 (5): 1564.
4. Osborne, C.A., Clinton, C.W., Bamman, L.K., Moran, H.C., Coston, B.R. and Frost, A.P. (1986). Prevalence of canine uroliths. Minnesota Urolith Center. *Vet. Clin. North Am.: Small Anim. Pract.* 16: 27.
5. Houston, D.M., Moore, A.E., Favrin, M.G. and Hoff, B. (2004). A look at over 16,000 urolith submissions to the Canadian Veterinary Urolith Centre from February 1998 to April 2003. *Can. Vet. J.* 45: 225.
6. Tion, M.T., Dvorska, J. and Saganuwan, S.A. (2015). A review of urolithiasis in dogs and cats. *Bulg. J. Vet. Med.* 18: 1.
7. Johnston, G.R., Walter, P.A. and Feeney, D.A. (1986). Radiographic and ultrasonographic features of uroliths and other urinary tract filling defects. *Vet. Clin. North Am. Small Anim. Pract.* 16: 261.
8. Leveille, R. (1998). Ultrasonography of urinary bladder disorders. *Vet. Clin. North Am.: Small Anim. Pract.* 28: 799.
9. Mukherjee, P., Sanyal, A. and Das, D.K. (2020). Surgical management of urolithiasis in a female dog. *Raksha Tech. Rev.* 10 (1): 45.
10. Rakshit, S., Roy, K. and Datta, I.C. (2020). Diet-related recurrent haematuria in a puppy: diagnosis and successful treatment of the rare clinical syndrome. *Raksha Tech. Rev.* 10 (1): 40.

Feline Upper Lip Eosinophilic Ulcer 'Rodent Ulcer': Diagnosis and Treatment

Sabita Rakshit, Dale Clark, Kabita Roy and I C Datta

Milford Veterinary Clinic, 110 Canal Street, Milford, USA and College of Veterinary Science,
Nanaji Deshmukh Veterinary Science University, Jabalpur, Madhya Pradesh.

Abstract

A 10-year-old spayed female Domestic Short Hair (DSH) cat was presented to the Milford Veterinary Clinic with an abnormal swelling in her left upper lip. On physical examination, all the vitals were within the normal limits (WLN). No change in the behavioural profile was noticed by the owner. The haemato-biochemical panel revealed no noteworthy deviation. However, referral microscopic examination of the impression cytology of the final needle aspirate (FNA) from the lesion indicated eosinophilic ulcer, name empirically 'rodent ulcer'. The ongoing combination oral Prednisolone and Clavamox® therapeutic regimen elicited favourable response, evidenced by abatement of the lip lesion. Suitable dietary changes and improved habitat (flea control measures) were advised to the owner.

Introduction

'Rodent ulcer' or 'indolent ulcer', discrete lesions bunched in the upper lip of house cats usually near one of the canine teeth, has no association with rodents. The name originated from the old erroneous belief that the morbidity results from rat bite, or other intruding rodents. The name has stuck. These are typically small flat or conclave, shallow, circumscribed, with thickened edges reddish-brown lesions, located either directly below the nose, or slightly lateral. In most cases, these do not elicit increased pain perception, or affect the cat's behavioral profile and appetite (1).

Though the precise aetiology of rodent ulcer remains unclear, bacterial infection, flea / food allergy, parasitic infestation, autoimmune reaction, ring worm infection, traumatic insult, viral infection, mosquito bite hypersensitivity may be involved. Fleas may serve as the trigger antigen (2). When primary allergic disease is suspected, followed by bacterial (3, 4).

The diagnostic tests include FeLV / FIV, CBC, skin scrapping for mite detection, and fungal culture (2). The haemogram clearly establishes eosinophilia, the hallmark of hypersensitivity reaction. Impression cytology of the Final Needle Aspirate (FNA) is a simple, yet highly dependable diagnostic test for the initial assessment: demonstration of eosinophilic inflammation and bacteria, with or without simultaneous neutrophilic ingress (1). Follow-up histopathology of the biopsy samples helps to differentiate from malignancies like lymphoma, and mast cell tumor (5). When infection is firmly established, appropriate use of antibiotics is indicated, concurrent with the diagnostic panel aimed to detect the primary allergy-inducing agent.

In this context, the integrated protocol of impression cytology, clinical assessment of the lesion, and microbial culture and sensitivity testing as a more dependable method for planning the optimized antibiotic schedule of 4 to 6 weeks duration is emphasized (1). A pragmatic approach in the allergy work-up is flea control in the entire household premises (6) along with dietary elimination trial employing a new single protein diet, or a hydrolyzed protein diet. The anti-inflammatory agent, steroids have been used as the first treatment choice (4, 7).

Case History



Fig 1. Army Hanson, the patient with rodent ulcer

Amy Hanson (Fig. 1), a 10-year-old spayed female Domestic Shorthair (DSH) cat weighing 4.9 kg, was presented to the Milford Veterinary Clinic on 2.11.2020 with an abnormal swelling in her left upper lip, a chance detection by the owner only three days earlier. There was no change in the behaviour of the indoor-outdoor cat. No history of any vomiting, diarrhea, coughing or sneezing.

Haematobiochemical Profile

Table 1. Patient's hemogram on 2.11.2020.

Parameter (Units)	Result It	Range	Status
TEC (1x 106/ μ l)	8.62	6.54-12.2	Normal
Hemoglobin (g/dl)	12.6	9.8-16.2	Normal
Hematocrit (%)	38.6	30.3-52.3	Normal
MCV (fl)	44.8	35.9-53.1	Normal
MCH (g/dl)	14.6	11.8-17.3	Normal
MCHC (%)	32.6	28.1-35.8	Normal
RDW (%)	21	15-27	Normal
TLC (1x 103/ μ l)	7.76	2.87-17.0	Normal
Reticulocyte (%)	0.2		
Reticulocyte-Hb (pg)	15.5	13.2-20.8	
Neutrophil (%)	50.6		
Lymphocyte (%)	37.1		
Eosinophil (%)	8.4		
Monocyte (%)	3.4		
Basophil (%)	0.5		
Neutrophil(1x103/ μ l)	3.93	2.9-17	Normal
Lymphocyte (%)	2.88	0.92-6.88	Normal
Eosinophil (%)	0.65	0.17-1.57	Normal
Monocyte (%)	0.26	0.05-0.57	Normal
Basophil (%)	0.04	0.01-0.26	Normal
Thrombocyte(1x103/ μ l)	138	151-600	Low

Catalyst One CBC Analyzer, IDEXX, USA

On physical examination, the vitals were within the normal limits (WNL). The upper left lip was apparently swollen, puffy, a little discoloured and ulcerated.

The peripheral lymph nodes popliteal, pre-femoral were swollen. Differential diagnosis at this point: rodent ulcer, cyst, infection and tumor. Blood work, radiology of the face, and FNA (Fine Needle Aspirate) for cytology of upper lip inside lesion (2 cm x 1cm) were suggested. Radiology was declined on cost consideration. For FNA the patient needed to be sedated to facilitate patient handling, so the patient was gassed in the tank with Isoflurane, and maintained on it, till the FNA procedure was over. Bleeding was stopped by digital pressure followed by antibiotic ointment Animax® application on the FNA site.

Table 2. Blood chemistry panel on 2.11.2020.

Parameter (Units)	Result It	Range	Status
Glucose (mg/dl)	135	74-159	Normal
SDMA (μ g/dl)	5	0-14	Normal
Creatinine (mg/dl)	1.4	0.8-2-4	Normal
BUN (mg/dl)	24		
BUN/Creatinine ratio	17		
Inorganic Phosphate (mg/dl)	4.3	3.1-7.5	Normal
Total calcium (mg/dl)	9.1	7.8-11.3	Normal
Total protein (g/dl)	7.5	5.7-8.9	Normal
Albumin (g/dl)	3.1	2.3-3.4	Normal
Globulin (g/dl)	4.4	2.8-5.1	Normal
A/G ratio	0.7		
ALT (U/l)	31	12-130	Normal
ALKP (U/l)	30	14-111	Normal
GGT (U/l)	0	0-4	Normal
Amylase (U/l)	802	500-1500	Normal
Lipase (U/l)	1157	100-1400	Normal
Bilirubin (mg/dl)	0.1	0.0-0.9	Normal
Cholesterol (mg/dl)	113	65-225	Normal
Na+ (mmol/l)	159	150-165	Normal
K+ (mmol/l)	3.9	3.5-5.8	Normal
Cl- (mmol/l)	120	112-129	Normal

Catalyst One Chemistry Analyzer, IDEXX, U

Diagnostics

Cytology report (IDEXX Lab)

Microscopic Findings

Low to moderate number of inflammatory cells with a few erythrocytes inside pale protein droplets. The inflammatory cells: neutrophils (some with phagocytized bacterial cocci indicating infection) and eosinophils, Mixed population of neutrophils, lymphocytes, eosinophils, and monocytes is seen in the other slides. Marked cell lysis with eosinophilic and neutrophilic mixed cell inflammation, septic amorphous proteinaceous material is seen.

Microscopic Interpretation

An idiopathic eosinophilic granuloma "rodent ulcer" with secondary bacterial infection is indicated. Latent primary food allergy, if any, needs to be identified and eliminated with suitable dietary changes.

Treatment

Polyflex® [200mg/ml] 0.75 ml and Dexamethasone [2mg/ml] 0.4 ml were given subcutaneously. Take home medications were antibiotic Clavamox® tablets 62.5 mg, to be given 1 BID (twice a day) for 10 days and Prednisolone liquid [10mg/ml] at a tapering dose of 0.5 ml BID for 3 days, 0.5ml OD (once a day) for 3 days and 0.5 ml EOD (every other

day) for 3 days.

Discussion

Feline eosinophilic ulcer, also named 'rodent ulcer' or 'indolent ulcer' represents hyper-sensitivity tissue reaction to some triggering antigen, often appearing in the mucocutaneous junction in the oral cavity. The geometry is variable: bilateral (symmetrical/ asymmetrical), or unilateral, mostly located adjacent to the upper canine teeth (1). Notably, pruritis and pain perception are rare. Clinically, the lesions in the lip margins exhibit superficial crusting, erythema and depressions. Occasionally, with increasing severity, these lesions may enlarge, but remain typically circumscribed alongside the small concave centre of localized granulation tissue. Pale yellow-white cell debris may be seen. The lesion appears as firm, proliferative ulcerated masses. Sometimes purulent exudate may be expressed from these lesions (5). Actually, 'rodent ulcer' is a component of feline eosinophilic granuloma complex (EGC). Eosinophilic plaque in the skin, and eosinophilic granuloma in the inner aspect of the oral cavity are the other two components. The inflammatory triadic may co-exist in some cats (8).

In the present study, a 10-year-old spayed DSH cat, apparently bright and alert, had a 2cm x 1cm ulcerated lesion inside the left upper lip, mild (1/3) dental tartar with no clinical signs of any discomfort, or dehydration/ No cutaneous inflammation seen in any body part. Close monitoring of the surface coat did not reveal the presence of fleas and ticks. All the physical parameters were within the normal limits (WNL). The hemogram (2.11.2020) revealed only marginal thrombocytopenia. The parallel blood chemistry panel testified to the structural/ functional integrity of all vital organs. Referral impression cytology test demonstrated inflammatory cells, predominantly eosinophils and neutrophils (some with phagocytosed bacteria), corroborating the clinical diagnosis of feline 'rodent ulcer'. The therapeutic strategy is aimed to treat the lesion effectively, and prevent aggravation of the problem through proven medicines, followed by suitable dietary changes and fleas/ tick's free habitat (9-11). The on-going Prednisolone and Clavamox® combination oral therapy is highly beneficial; the remission has been effectively controlled.

References

1. Bajwa, J. (2019). Feline indolent ulcers and their significance. *The Canadian Veterinary Journal*. 60: 1009.
2. Jellison, Jennifer. (2021). Case study: eosinophilic ulcer (rodent ulcer) in a cat Case study Banfield Pet Hospitals.
3. Lee, M., Bosward, K. L. and Norris, J. M. (2010). Immuno-histological evaluation of feline herpes virus-1 infection in feline eosinophilic dermatoses or stomatitis. *Journal of Feline Medicine and surgery*. 12: 72-79.
4. Wildermuth, B. E., Griffin, C. E. and Rosenkranz, W. S. (2012). Response of feline eosinophilic plaques and lip ulcers to amoxicillin trihydrate- clavulanate potassium therapy: A randomized, double-blind placebo controlled prospective study. *Veterinary Dermatology*. 22: 521.
5. Miller, W. H., Griffin, C. E. and Campbell, K. L. (2013). *Muller and Kirk's Small Animal Dermatology*, Elsevier, St. Louis, Missouri, USA. pp. 716.
6. Colombini, S. and Hodgins, E. C., Foil C. S., Hosgood, G., and Foil, L. D. (2001). Induction of feline flea allergy dermatitis and the incidence and histopathological characteristics of concurrent indolent lip ulcers. *Veterinary Dermatology*. 12: 155.
7. Nelson, R. W. and Couto, G. C. (2003). *Small Animal Internal Medicine*. p.407.
8. Hunter, T. and Ward, E. (2021). Eosinophilic granuloma complex in cats. *VCA Animal Hospital* Accessed on 11.2.2021 <https://vcahospitals.com/know-your-pet/feline-eosinophilic-granuloma-complex-in-cats>
9. Ployngam, T., Tobias, A. H., Smith S. A., Torres, S. M. F. and Ross, S. J. (2006). Hemodynamic effects of methyl prednisolone acetate administration in cats. *American Journal of Veterinary Research*. 67: 583.
10. Wildermuth, H. T., Griffin, C. E. and Rosenkranz, W. S. (2006). *Clinical Techniques in Small Animal Practice*. 21: 150.
11. Vercelli, A., Raviri, G. and Cornegliani, L. (2006). The use of oral cyclosporine to treat feline dermatoses: A retrospective analysis of 23 cases. *Veterinary Dermatology*. 17: 201.

Nutritional Deficiencies in Guinea Pig

Amishi, Amita Dubey, Madhu Swamy and Yamini Verma

Dept. of Veterinary Pathology, College of V S & A H, NDVSU, Jabalpur, Madhya Pradesh

Abstract

The guinea pig or domestic guinea pig (*Cavia porcellus*), is a species of rodent belonging to the genus *Cavia* in the family Caviidae. The natural diet of the guinea pig is grass; their molars are especially adapted to grinding plant matter and continuously grow throughout their lives. Guinea pigs do not synthesise their own vitamin C, like humans, but unlike most other mammals, and must receive this essential nutrient from food. They will suffer from potentially lethal scurvy if they do not consume enough vitamin C. Poor diets supplementation in guinea pigs can cause muscular dystrophy, pregnancy related issues, metastatic calcification, teeth problems and different vitamins deficiencies specially vitamin C. So, a healthy diet with proper supplementation of essential amino acids, minerals and vitamins is required to prevent nutritional deficiency in guinea pigs.

Introduction

The domestic guinea pig (*Cavia porcellus*) has been bred for at least 400 years in captivity and is likely to have originated in Peru, Argentina or Brazil. This herbivorous species eats large amounts of vegetation in its natural habitat. The guinea pig is simple-stomached, like the rat, mouse, and rabbit; but in comparison to these animals, stomach of the guinea pig is lined with glandular epithelium (1). The intestine enables the production of primarily gram-positive bacterial flora, which may lead to the nutritional requirements of the host, probably by direct absorption of bacterial metabolites or digestion and absorption of intestinal bacteria and other materials. There is a large semi-circular cecum in the guinea pig with several lateral pouches. This organ is identical to that of the rabbit and may have similar digestive roles, such as microbial production of vitamin B and essential amino acids and coprophagic recycling of intestinal contents (2). From a nutritional point of view, the guinea pig is better known for its dietary need for vitamin C. In studies of collagen biosynthesis,

wound healing, and bone growth, this function has made the guinea pig exceptionally helpful. Young guinea pigs tend to have a relatively high nutritional demand for arginine, folic acid, and chosen minerals, but this may not be accurate as more information becomes available about individual nutrient requirements.

Major Nutrition Deficiencies in Guinea Pigs

Protein, Amino Acids and Essential Fatty Acids

Protein and fatty acid deficiencies are usually observed in case of restricted or neglected feeding, or may be due to malocclusion. Reproductive disability, both miscarriage and the mortality of low-weight (under 50 g) neonates, and hair loss are the normal effects of these deficiencies. Arginine, methionine, and Tryptophan are the limiting amino acids for guinea pigs. Guinea pigs can produce niacin from tryptophan, and diets that are low in tryptophan can produce cataract (4). Classical signs of EFA deficiency are mainly ulcers around the neck and ears, loss of hairs on the ventral aspect, dermatitis, lack of normal growth. Some of the additional signs may also found in EFA deficiency like priapism, spleen underdevelopment, and enlargement of kidneys, liver, heart and adrenals (5).

Minerals

Calcium, Phosphorous and Magnesium

The requirements of calcium, phosphorus, potassium, and magnesium for guinea pig, like those of the rat, seem to represent the interactions between these elements. Guinea pigs, which lower calcium concentration, may develop rachitic lesions in ribs and long bones. Younger animals usually have more bone defects than older animals. Enamel hypoplasia is usually seen with calcium deficiency in guinea pigs (6). Guinea pigs use cation exchange and phosphate anions to remove excess hydrogen ions rather than to remove protons by

Nutrient	Requirement per Kg diet
Protein	180g
Essential fatty acids	1.33-4g
Arginine	12g
Lysine	8.4g
Methionine	6g
Calcium	8g
Phosphorus	4g
Magnesium	1g
Vitamin A (Retinol) & B-carotene	6.6mg & 28mg
Vitamin D	0.025mg
Vitamin E	26.7mg
Vitamin C	200mg

Nutrient Requirements of Guinea Pig According to NRC, 1995 (3)

excretion of ammonium ions. Calcium turnover in guinea pigs is rapid in comparison to other rodents. The magnesium requirement of guinea pigs depends on calcium, phosphorus, and potassium dietary concentrations. Excess of calcium and phosphorous can increase the requirement of magnesium. Magnesium-deficient guinea pigs not only decreased muscle concentration of extracellular and intracellular magnesium (20 per cent and 80 per cent of control levels, respectively) but also decreased muscle potassium and increased intracellular sodium and water concentrations (7). Poor growth, hair loss, reduced movement, poor muscular coordination and stiffness of the hind limbs, increased serum phosphorus, and anaemia are clinical signs of magnesium deficiency in young guinea pigs (8). Necropsy findings in magnesium deficiency are enlarged pale kidneys, white foci and streaks in liver, soft tissue calcification, and darkened, eroded, and soft incisors (9).

Vitamins

1. Vitamin A

Guinea pigs apparently have a high vitamin A requirement. Onset of symptoms of deficiency differs greatly with age, liver vitamin A concentration, and stress factors. Young guinea pigs can show symptoms of deficiency within 2 weeks, while older pigs may take almost 10 weeks to consume vitamin A or provitamin A-free diets. Poor growth, weight loss, followed by eyelid incrustation and severe dermatitis are the definitive signs of vitamin A deficiency (10). Gross pathology reveals organic debris accumulation in the bile ducts and gallbladder, corneal clouding, and xerophthalmia. Histologically, squamous metaplasia and keratinization is observed in epithelia of different organs (11). The primary effect of vitamin A deficiency on guinea pig incisors is on odontogenic epithelium with incomplete cell differentiation, loss of organisation and development of defective dentin by atrophic odontoblast (12).

2. Vitamin E

Vitamin E or selenium deficiency is associated with a variety of symptoms in guinea pigs and other animals. In guinea pigs, the primary distinctive symptoms are hindlimb failure due to myasthenia or muscular dystrophy, diminished reproductive performance, paralysis and death (13). Coagulative muscle necrosis, testicular degeneration, degenerative alterations in seminiferous tubules, spermatozoa and spermatid depletion, and elevated serum creatine phosphokinase are the underlying or related symptoms.

3. Vitamin C

Vitamin C deficiency causes scurvy in guinea pigs. Guinea pigs cannot synthesize vitamin C therefore need dietary supplementation. Vitamin C is essential for hydroxyproline and hydroxylysine synthesis in collagen. Collagen is an essential component for maintaining integrity of blood vessels, wound healing and bone formation. Deficiency of vitamin C usually results in fragile blood vessels, which lead to haemorrhages from the gums and other internal organs. Its deficiency may also cause abnormal growth of bone and cartilage, mostly affects the joints, leading to painful swelling of joints. It can also affect the anchoring of teeth in their bony sockets, contributing to malocclusion of the teeth and trouble feeding. Deficiency of vitamin C results in reduced absorption of vitamin B12; improved absorption of alanine and leucine; increased activity of brush boundary sucrase, alkaline phosphatase, and leucine aminopeptidase; and increased sialic acid and total lipid intestinal membrane concentrations (14). Within 2 weeks of obtaining a poor diet, symptoms may be observed. Owing to the combination of rapid growth and an increased use of vitamin C in guinea pigs under 4 months of age, young animals are more susceptible.

Signs and Pathology

Major clinical signs of vitamin C deficiency are lameness, bunny-hop gait, decreased appetite or inability to eat if teeth are involved. Joint pain and swelling in legs and ribs, bleeding of the gums or skin, diarrhoea, rough hair coat and weight loss is also observed. Lesions associated with the overt classical ascorbic acid deficiency are easily identified and consist of subperiosteum, skeletal muscles, joints and intestinal haemorrhage. Microscopically, dentin formation is altered and epiphyseal growth centres of long bones with osteoid formation are substantially reduced (1). Histological analysis of the long bones of affected guinea pigs reveals a pronounced reduction or complete absence of osteoid trabeculae in metaphysis (15). Ascorbic acid deficiency results in an accumulation of cholesterol in the liver and a concomitant reduction in bile acid formation (16).

Prevention

Daily requirement for vitamin C is 25–30 mg/animal/day for lifelong, however, should be given @ 50–100 mg daily in case of deficiency. While guinea pig diets are fortified with vitamin C, their contents are diminished by storage, lightness, heat and moisture. Within 6 weeks of storage, as much as 50 percent of vitamin C activity can be lost; it is thus advised to feed the product within 90 days of milling at the rate of minimum 6mg/kg/day.

Conclusion

Guinea pigs are herbivorous animals having several similarities with rat, mice and rabbits. Yet they exhibit certain specific nutritional disturbances unlike other species. Guinea pigs usually become deficient in different amino acids, minerals and vitamins if they are not provided in sufficient amount in the diet, but vitamin C deficiency is the most commonly encountered problem in guinea pigs, especially the young ones. They cannot synthesize or store vitamin C. Vitamin C deficiency contribute to several clinical signs associated with blood vascular system, bone and cartilage formation, collagen synthesis and problems related to gums and teeth. These deficiencies may seriously affect the growth of the affected animal as well different body systems like reproduction, musculoskeletal and cardiovascular systems and may cause mortality in guinea pigs. Daily dietary supplementation of essential nutrients in desirable amount can overcome these deficiencies.

References

1. Navia, J. M. and Hunt, C. E. (1976). Nutrition, nutritional diseases and nutrition research application. In: Wagner, J. E. and Manning, P. J. (ed.). *Biology of the Guinea Pig*, New York: Academic Press. pp. 235.
2. Hunt, C. E. and Harrington, D. D. (1974). Nutrition and nutritional diseases of the rabbit. In: *The Biology of the Laboratory Rabbit*, New York: Academic Press.
3. National Research Council (NRC) (1995). *Nutrient Requirements of the Laboratory Rat: Nutrient Requirements of Laboratory Animals*, 4th ed. National Academy Press. Washington, DC.
4. Reid, M. E. and Von Sallman, L. (1960). Nutritional studies with the guinea pig. VI. Tryptophan (with ample dietary niacin). *Journal of Nutrition*. 70: 329.
5. Reid, M. E. and Martin, M. G. (1959). Nutritional studies with the guinea pig. V. Effects of deficiency of fat or unsaturated fatty acids. *Journal of Nutrition*. 67:611.
6. Howe, P. R., Wesson, L. G., Boyle, P. E. and Wolbach, S. B. (1940). Low calcium rickets in the guinea pig. In: *Proceedings of the Society for Experimental Biology and Medicine*. 45:298.
7. Grace, N. D. and O'Dell, B. L. (1970). Interrelationship of dietary magnesium and potassium in the guinea pig. *Journal of Nutrition*. 100:37.
8. Morris, E. R., and O'Dell, B. L. (1963). Relationship of excess calcium and phosphorus to magnesium requirement and toxicity in guinea pigs. *Journal of Nutrition*. 81:175.
9. Morris, E. R., and O'Dell, B. L. (1961). Magnesium deficiency in the guinea pig. Mineral composition of tissues and distribution of acid-soluble phosphorus. *Journal of Nutrition*. 75:77.
10. Bentley, L. S. and Morgan, A. F. (1945). Vitamin A and carotene in the nutrition of the guinea pig. *Journal of Nutrition*. 30:159.
11. Howell, J. M., Thompson, J. N. and Pitt, G. A. J. (1967). Changes in the tissues of guinea pigs fed on a diet free from vitamin A but containing methyl retinoate. *British Journal of Nutrition*. 21:37.
12. Wolbach, S. B. (1954). Effects of vitamin A deficiency and hypervitaminosis A in animals. In: Sebrell, W. H. and Harris, R. S. (ed.). *The Vitamins*, Vol. 1, New York: Academic Press. Pp.106.
13. Hill, K. E., Montine, T. J., Motley, A. K., Li, X., May, J. M. and Burk, R. F. (2003). Combined deficiency of vitamins E and C causes paralysis and death in guinea pigs. *American Journal of Clinical Nutrition*. 77 (6): 1484.
14. Dulloo, R. M., Majumdar, S., Chakravarti, R. N. and Mahmood, A. (1982). Intestinal brush border membrane structure and function effect of chronic vitamin C deficiency in guinea-pigs. *Biochemical Medicine*. 27:325.
15. Vilter R.W. (1967). Effects of ascorbic acid deficiency in man. In: Sebrell and Hams (ed.). *The Vitamins*, Vol. 1, 2nd edn., New York: Academic Press. Pp. 457.
16. Horing, D. and Weiser, H. (1976). Ascorbic acid and cholesterol: effect of graded oral intakes on cholesterol conversion to bile acids in guinea pigs. *Experientia*. 32(6): 87.

Anti-microbial Resistance Crisis in COVID-19 Pandemic

Bhoomika, Sonam Bhatt, Vivek Kumar Singh, Anjay and Purushottam Kaushik

Dept. of Veterinary Public Health and Epidemiology, Dept. of Veterinary Medicine, Dept. of Veterinary Clinical Complex, BVC, BASU, Patna, Bihar.

Abstract

The modern medicine era begins with the discovery of systemic sulphonamides and penicillin, that has increased life expectancy to mitigate the hazard of bacterial infections. Although soon after discovery bacteria became resistant to commonly used antibiotics. Antimicrobial Resistance (AMR) becomes a threat to public health and economy. While global efforts to curb further development and spread of AMR are being established, the emergence of COVID-19 pandemic might have disrupted these efforts and further augmented the problem, especially in low- and middle-income countries. In spite of that factors like strict lockdown, hand hygiene practice, use of face mask, social distancing contributes to decreasing AMR. The contribution of pandemic in AMR is global and possible causes has been discussed here.

Introduction

Since the first case of 2019 novel coronavirus (COVID-19) pneumonia, caused by SARS Cov-2 (Severe Acute Respiratory Syndrome Coronavirus-2) was recorded in wet market of Wuhan, China, the diseases havenow affected more than 198,234,951 people in 220 countries with more than 4,227,359 fatalities globally (1).

To combat this pandemic, health authorities of different countries developed and implemented numerous control and preventive measures, such as wearing of mask, social distancing, hand hygiene practices, active identification and quarantine of close contacts, isolation of sick patients, lockdown strategies and rapid set of shelter hospital etc.

These extensive management measures in response to COVID-19 came up with additional benefits in terms of reducing incidence of other infections. According to many studies, incidences of several infectious respiratory infections such as seasonal influenza, pneumococcal disease and tuberculosis during COVID-19 pandemic has been markedly reduced as compared with the same period in previous years. On the other hand, we cannot ignore the increased incidence of antimicrobial resistance (AMR), which may be attributed due to injudicious use of antimicrobial agents during the COVID-19 pandemic (2).

Possible Reasons for the AMR Upsurge

Although the initial phase of pandemic, most of the emphasis has been paid on COVID-19 diagnosis and disease management, and the impact on AMR has been largely overlooked. The strict implementation of antimicrobial stewardship (AS) and infection prevention and control measures are

imperative the development and spread of AMR. Hence, disruption of AS measures is imperative by the ongoing pandemic may speed the expansion of AMR globally. Some possible reasons for increase incidences of AMR are as follows:

A. Prophylaxis, Nosocomial Infection and Self-medication

COVID-19 is a disease caused by the SARS-CoV-2, therefore as like other viral infections, as such, antibacterial will not directly affect the virus. The superimposed bacterial infections are responsible for bacterial pneumonia. Mechanical ventilator is responsible for most of the cases of bacterial pneumonia and required antibiotic intervention. Additionally, antibiotics are used as prophylaxis to prevent bacterial respiratory diseases among the patients affected by viral infections. The use of antibiotics as a prophylaxis is a part of COVID-19 management. Such cautious use of antibiotics as prophylaxis against nosocomial bacterial infections is often thought to be necessary, as secondary bacterial infections grave the conditions in the patients suffering from viral infections. Therefore, physicians may feel obliged to prescribe antibiotics as prophylaxis to prevent secondary bacterial and fungal super infections.

Another common problem noticed in the developing countries is the self-medication. The low testing capacity, inaccessibility to healthcare, poverty and broad availability of over-the-counter antibiotics could contribute to self-medication in symptomatic but undiagnosed individuals. The ongoing pandemic exacerbate such practice, which will inevitably contribute to AMR. Together, an increase in secondary bacterial infections, antibiotic prophylaxis, and self-medication driven by the pandemic likely contribute to AMR.

B. Lack of Healthcare Facilities and Compromised Antibiotic Stewardship (AS)

Despite the standards guidelines released by the national and international organizations such as WHO and NIH, healthcare staffs are incapable to follow AS when they are under pressure to treat acutely ill patients who are under the risk of dying. In addition, a rapid and abrupt increase in COVID-19 related hospitalizations led to shortage of healthcare essentials such as the personal protective equipment (PPE) and ventilators, forcing healthcare employees to either reuse or share these resources.

There are also reports of lack of infection and control measures, including gaps in screening of AMR, increased patient-to-nurse ratio, and compromised hospital-waste management.

C. Tele Medication

Face to face treatment and prescription is increasingly being replaced by telemedicine both in developed and developing countries. Such type of consultation increased the antibiotic prescription. According to The National Health Service (NHS) reported that despite of 50% decrease in a face-to-face appointment during lockdown, the antibiotic prescription has increased by over 6%, which could contribute the misuse of antibiotics and selection of resistant bacteria (3).

D. Improper Medical Wastes Management

The dissemination of the resistant bacteria in the environment occurs throughout various sources, including healthcare, agriculture, and the environment. Many countries have experienced a massive increase in health care wastes, during the initial stage of COVID-19 pandemic. The compromised waste management, and inadequate handling of medical wastes due to shortage of workforce and restrictions in the movement. Such inadequate waste management can be source of antibiotics and resistant bacteria in the environment through contaminated water, excretion and food. Therefore, strict implementation of proper waste disposal in the hospital settings along with pronged one health approach to protecting humans, animals, and the environment, is obligatory to address the issue of AMR.

E. Discontinuity in AMR Research and Implementation of Health Programs

The ongoing pandemic has also affected the scientific research and innovations. Many national and international collaborative studies, AMR surveillance programs and clinical trials has been temporarily suspended during lockdown period. Similarly, pandemic also subsided the implementation of various global health programs,

including routine childhood vaccination program, tuberculosis and HIV control programs. The mass vaccination campaigns against, cholera, measles, meningitis, polio, tetanus, typhoid and yellow fever have been temporarily suspended. Such interrupted scientific communication regarding research and innovation and health services increases immunosuppression and predispose patients afflicted with bacterial infections, which leads to increase hospitalization and negatively impact on AMR.

Conclusion

The AMR has emerged as an unforeseen and unavoidable consequence of the COVID-19 pandemic. However, the exact role of COVID-19 pandemic in AMR is not known and can only be speculated at this point. Among the multifactorial suppose cause, particularly a high rate of antimicrobial agents' consumption in COVID-19 patients with relatively low rates of co-or-secondary infections. The appropriate prescription and judicious use of antimicrobials according to the principles of antimicrobial stewardship, along with the proper diagnosis and implementation of proper control measures, may prevent the occurrence of drug resistant bacteria during pandemic.

References

1. World Health Organization. (2021). Corona virus dashboard. <https://covid19.who.int/>.
2. Subramanya, S. H., Czyz, D. M., Acharya, K. P. and Humphreys, H. (2021). The potential impact of the COVID-19 pandemic on antimicrobial resistance and antibiotic stewardship. *Virus Disease*, 1.
3. Armitage, R. and Nellums, L. B. (2020). Antibiotic prescribing in general practice during COVID-19. *Lancet Infect Dis*.

Functional Role of Common Ingredients Used in Preparation of Value Added Meat Products

Z Naveen, S A K Azad, S Bindhu and B Eswara Rao

Dept. of Livestock Products Technology, Sri Venkateswara Veterinary University, Tirupati, Andhra Pradesh

Value addition to meat by-products entails a degree of novelty which makes a by-product that is considered as waste, to be used as raw material subjected to further processing into edible food items preferred by consumers or inedible products with commercial viability (e.g. plastics, pharmaceuticals, energy) (1). In case of edible items, meat by-products constitute an excellent source of nutrients like essential amino acids, minerals and vitamins (2). Value addition can be attained in terms of sustenance, shelf stability, enhanced technological functions (emulsifiers, flavouring compounds, water bonding agents), superior sensory quality (color, texture, flavor) or even more convenience.

The objectives of meat processing are varying with time. Initially, the basic purpose of meat processing focussed on improving shelf life, flavour and nutritive value. Later the emphasis was shifted to modify or upgrade the less desirable meat cuts to desirable products. Advances in the concept of processing have focussed to provide convenience and variety products. Thus, that in processing of meat, low value meat cuts is processed to improve shelf life, flavour, taste, nutritive value and convenience. Currently, the consumer demands for healthier meat and meat products with decreased levels of fat, cholesterol, reduced contents of sodium chloride and nitrite, enhanced composition of fatty acid profile and incorporated with health enhancing ingredients are rapidly increasing globally. The consumer acceptance of functional foods is diverged broadly based upon their economic, social, geographical, political, cultural, ethnic backgrounds (3).

Value added meat products are further processed meat products with increasing convenience to consumer through decreasing preparation time, minimizing preparation steps, allowing use of specific parts, taking risks out of kitchen and increasing value of product. The three basic requirements to be considered as a functional food include 1) derived from a natural occurring ingredients; 2) consume as a part of daily diet; and 3) involve in regulating specific process for human including delaying aging process, preventing the risk of disease and improving immunological ability (3).

Meat and meat products are important sources for protein, fat, essential amino acids, minerals and vitamin and other nutrients (4). The commonly used ingredients in preparing meat products of Indian cuisine are of natural origin and have a lot of functional roles, which not only enhances the taste, but also

make the meat product a functional food. The functional roles of commonly used ingredients in the preparation of different meat products are discussed.

Chilli (*Capsicum annum L*)

Antioxidant Property

Chillies have vitamin C and Vitamin A containing beta-carotenoids which are powerful antioxidants. These antioxidants destroy free radicals damaging nerve cells and blood vessel in diabetes. Antioxidants present in the chilli wipe out the radical bodies that could build up cholesterol causing major heart diseases such as atherosclerosis, and also effective against osteoarthritis and rheumatoid arthritis. Flavonoids, β -carotene, α -carotene, lutein, zeaxanthin, and cryptoxanthin antioxidant substances in capsicum help to protect the body from injurious effects of free radicals generated during stress and other disease conditions.

Detoxicant Activity

Capsaicin and Flavonoids act as detoxifiers as they remove waste products from our body. It also acts as gastrointestinal detoxicant, helping in digestion of food.

Acts as Pain Killer

Chillies stimulate the release of endorphins that are natural pain killers.

Acts as Antibiotic

Capsaicin bring fresh blood to the site of the infection. The fresh blood fights infection. The white blood cells and leukocytes present in the fresh blood fights viruses.

Acts as Brain Stimulant

Capsaicin stimulates brain to excrete endorphin and gives a sense of pleasure when ingested. This is the reason people get addicted to chilli.

Acts against Cancer

It has been noted that vitamin C, beta-carotene and folic acid found in chilli reduces the risk of colon cancer. Chillies such as red pepper have carotenoid lycopene, which prevents cancer disease.

Prevention of Heart Attack

Chillies have vitamin B6 and folic acid. The vitamin B reduces high homocysteine level. High homocysteine levels have been shown to cause damage to blood vessels and are associated with a greatly increased

risk of heart attack and stroke. It also converts homocysteine into other molecules which is beneficial to lower cholesterol level.

Counter Smoking Effect

Cigarette smoke contains benzopyrene which destroys the vitamin A in the body. The vitamin A present in chilli reduces inflammation of lungs and emphysema caused due to cigarette smoking.

Acts as Gastroprotective

Capsaicin, the pungent ingredient of chilli, is gastro-protective. Epidemiological and clinical data suggests that chilli ingestion may have a beneficial effect on human peptic ulcer disease. Acute intra-gastric administration and chronic ingestion of chilli powder in doses comparable with that consumed in humans (up to 200 mg in single doses or 200 mg daily for four weeks) likewise protected the gastric mucosa. Both the mucosa and gastric juice had higher mucus contents when capsaicin or chilli is administered. Increased gastric mucus production may therefore be mechanism by which capsaicin and chilli exert their gastro protective effect.

Black Pepper (*Piper nigrum*)

Anti-inflammatory Effect

Piperine, the active phenolic compound in black pepper, has anti-inflammatory, anti-arthritic and analgesic effect. Piperine inhibited the expression of pro-inflammatory interleukin 6, and MMP13, a gene involved in the promotion of arthritis and metastasis. It reduces the production of a pro-inflammatory prostaglandin thus preventing the inflammation all over the body, even at a very low dose.

Inhibits the Protein Glycation

Protein glycation is a process in which the sugar molecules bind to protein molecules without enzymatic control. The result is the accumulation of end products that speed ageing and the degeneration of cellular substances, caused by diabetes. Black pepper, ginger, cumin, cinnamon, and green tea where the only extracts tested which showed significant ability to inhibit these end products. (British Journal of Nutrition, November 6, 2008).

Acts as an Antioxidant

Black pepper had the highest level of antioxidant activity and also the highest phenolic content which further contribute for antioxidant property. The data indicates that supplementation of black pepper or the active principle of black pepper, piperine, can reduce high-fat diet induced oxidative stress to the cells. (redox report 2004).

Action against Colon Cancer

Black pepper acts on the colon cancer by preventing the proliferative activity of the cancer cells (*Clinical Laboratory Science, Summer, 2008.*)

Digestive Activity

Hydrochloric acid is necessary for digesting proteins and other food components. Most digestive difficulties occur due to lack of hydrochloric acid. Black pepper stimulates the taste buds and alerts the stomach to increase hydrochloric acid secretion, improving digestion.

Black pepper can act as a diuretic, carminative reducing bloat in the intestinal tract wherein it also promotes digestive health through its antioxidant effects.

Bio Availability of Nutrients

Piperine increases absorption and this compound operates through several other pathways to increase the bioavailability of nutrients from food and supplements. It stimulates amino-acid transporters in the intestinal lining and inhibits the removal of substances from cells. Each of these actions allows nutrients to enter and remain within their target cells for longer periods of time so they are utilized completely.

Turmeric (*Curcuma langa*)

Anti-inflammatory Activity

Curcumin helps in reducing inflammation, treats upset stomach, scabies, diabetes and many viral infections. Curcumin is thought to be the primary pharmacological agent in turmeric. In numerous studies, curcumin's anti-inflammatory effects have been shown to be comparable to the potent drugs hydrocortisone and phenylbutazone. Curcumin produces no toxicity whereas its benefits are the results of not only antioxidant activity, but also because of inhibition of a major cellular inflammatory agent called NF kappa-B.

Antioxidant Activity

Curcumin also exerts very powerful antioxidant effects. As an antioxidant, curcumin is able to neutralize free radicals. Free radicals are responsible for the painful joint inflammation and eventual damage to the joints. Turmeric's combination of antioxidant and anti-inflammatory effects can reduce joint pains and chronic rheumatoid arthritis. Other antioxidant activities include wound healing and also reduce the liver damage caused by drinking of alcohol. And it was found to be very effective against the Alzheimer's disease.

As an Anti-carcinogenic Agent

Curcumin's antioxidant actions enable it to protect the colon cells from free radicals that can damage cellular DNA. Curcumin also helps the body to destroy mutated cancer cells, so they cannot spread through the body. A primary way in which curcumin acts as an anticancer agent is by enhancing the liver function. Additionally, other suggested mechanisms by which it may protect against cancer development include inhibiting the synthesis of a protein thought to be instrumental in tumour formation. Also by preventing the development of additional blood supply necessary

for cancer cell growth. Prevents the formation of the inflammatory chemical cyclooxygenase-2 (COX-2), Polyphenol curcumin is another compound that reduces the rate of growth in cancer cells.

Improves Liver Function

Turmeric helps in enhancing the liver function. In a recent study conducted on rat, to evaluate the effects of turmeric on the liver's ability to detoxify xenobiotic (toxic) chemicals, it was observed that levels of two very important liver detoxification enzymes (UDP glucuronyl transferase and glutathione-S-transferase) were significantly elevated in rats fed turmeric as compared to controls.

Prevents Heart Disease

Curcumin may be able to prevent the oxidation of cholesterol in the body. Since oxidized cholesterol is what damages blood vessels and builds up in the plaques that can lead to heart attack or stroke. Hence, preventing the oxidation of new cholesterol may help to reduce the progression of atherosclerosis and diabetic heart disease. B6 in turmeric prevents homocysteine proliferation which damages the coronary blood vessels.

Reducing the Cholesterol Levels

Curcumin is also a messaging molecule that communicates with genes in liver cells, directing them to increase the production of mRNA (messenger proteins) that directs the creation of receptors for LDL (bad) cholesterol. With more LDL-receptors, liver cells are able to clear more LDL-cholesterol from the body.

Ginger (*Zingiber officinale*)

Ginger is successful in eliminating gastrointestinal distress and symptoms of motion sickness, especially seasickness. Journal, *Obstetrics and Gynecology*, has confirmed that ginger is effective in relieving the severity of nausea and vomiting during pregnancy.

Anti-inflammatory action

Ginger contains very potent anti-inflammatory compounds called gingerols which are effective against osteoarthritis or rheumatoid arthritis. In this research, ginger was shown to suppress the pro-inflammatory compounds (cytokines and chemokines) produced by synoviocytes (cells comprising the synovial lining of the joints), chondrocytes (cells comprising joint cartilage) and leukocytes (immune cells).

Anti-oxidant Effect

Of more than 50 antioxidants isolated from the rhizomes of ginger. The isolated antioxidants are divided into two groups: gingerol related compounds and diarylheptanoids. Free radical protection afforded by one of its active phenolic constituents, 6-gingerol. In this in vitro (test tube) study, 6-gingerol was shown to significantly inhibit the production of nitric oxide, a highly reactive nitrogen molecule that quickly forms a very damaging free radical called peroxynitrite.

Gingerol also reduces the effect of oxidation due to the radiation exposure.

Against Colon and Ovarian cancer

Gingerols, the main active components in ginger and the ones responsible for its distinctive flavor, may also inhibit the growth of human colorectal cancer cells. Research presented at the Frontiers in Cancer Prevention Research suggested that, the active phytonutrients in ginger kill ovarian cancer cells by inducing apoptosis.

Immune Booster

Ginger is not just only be warming agent on a cold day, but can help to promote healthy sweating, which is often helpful during cold and flu. A good sweat may do a lot more than simply assist detoxification.

Investigators have named a skin protein in sweat as dermicidin. Dermicidin is produced in the body's sweat glands, secreted into the sweat, and transported to the skin's surface where it provides protection against invading microorganisms, including bacteria such as *E. coli* and *Staphylococcus aureus* (a common cause of skin infections) and fungi, including *Candida albicans*.

Garlic (*Allium sativum*)

Prevents Heart Diseases

Studies do suggest that garlic may help to prevent heart disease. It may slow down atherosclerosis and lowers the blood pressure a little, between 5% and 8%. Garlic also seems to act as a blood-thinner, which may help to prevent heart attacks and strokes.

Against the Parasites

It kills roundworms, *Ascaris lumbricoides*; the most common type of intestinal parasite. But it hasn't been tested in humans yet. Several studies report that a garlic gel, applied to the skin, may treat ringworm, jock itch, and athlete's foot.

Possess only partial antioxidant property.

Fennel (*Foeniculum vulgare*)

It is known as *saunf* or *mouti saunf* which is used as a flavouring agent. Fennel is a major source of vitamin c, polyphenols and selenium that act as antioxidants which are active against Parkinson's disease and Alzheimer's disease. Further it is effective in inflammation of visceral organs.

It also contains minerals such as copper, iron, calcium, magnesium, manganese, zinc and selenium required for building up immunity. Anethole is the active anticancer component present in Fennel which resists activities like adhesive and invasive activities of the cancer cells.

Cinnamon (*Cinnamomum verum* - J. S. Pregl)

Cinnamon is high in polyphenols, proantho-cyanidins, essential oils, and is a great source of manganese, fibre, iron, and calcium.

Regulates the Blood Sugar

Cinnamon contains certain proanthocyanidins that have insulin-like properties. These compounds appear to mimic the action of insulin in the body and may help to regulate blood sugar levels. It also contains certain polyphenols that help to activate insulin and transport glucose. Thus Cinnamon helps people with Type 2 diabetes in order to control blood sugar levels.

Antimicrobial Action

Essential oils in cinnamon are potent antibacterial and antifungal agents. They contain active components called cinnamaldehyde, cinnamyl acetate, and cinnamyl alcohol, and a wide range of other volatile substances which are very effective against bacteria and fungi. The addition of cinnamon essential oil inhibited the growth of the foodborne pathogenic *Bacillus cereus* for at least 60 days (August 2003 issue of the *International Journal of Food Microbiology*).

Prevents Heart Diseases

Cinnamon health benefits include potent anti-inflammatory properties that are helpful in the prevention of heart disease. Cinnamon also improves which ensures oxygen supply to the cells. The cinnamaldehyde in cinnamon helps to prevent unwanted clumping of blood platelets during blood circulation, by inhibiting the release of an inflammatory fatty acid called arachidonic acid from platelet membranes and reducing the formation of an inflammatory messaging molecule called thromboxane A₂. The ability to lower the release of arachidonic acid from cell membranes also puts it in the category of an "anti-inflammatory" food.

"A growing consensus among cardiologists pinpoints abnormal inflammation in artery walls as a root cause of atherosclerosis and coronary heart disease."

Andrew Weil, M.D.

Cancer Prevention

In a study published by researchers at the U.S. Department of Agriculture in Maryland, cinnamon reduced the growth of leukaemia and lymphoma cancer cells. Cinnamon reduces risk of colon cancer by helping to remove excess bile in digestive tract and prevents the damage to colon cells.

Reduces Blood Sugar

Cinnamon slows the rate at which the stomach empties after meals, reducing the rise in blood sugar after eating. Adding cinnamon to the rice pudding lowered the gastric emptying rate from 37% to 34.5% and significantly lessened the rise in blood sugar levels after eating. Insulin-enhancing complexes present in cinnamon are catechin / epicatechin oligomers that increase the body's insulin-dependent ability to use glucose roughly 20-fold (*Journal of Agricultural and Food Chemistry, Anderson et. al.*)

Antioxidant Activity

Cinnamon is powerful antioxidant, when compared to six other antioxidant spices (anise, ginger, liquorice, mint, nutmeg and vanilla) and the chemical food preservatives (BHA (butylated hydroxyl anisole), BHT (butylated hydroxyl toluene), and propyl gallate). Cinnamon prevented oxidation more effectively.

Clove (*Syzygium aromaticum*)

Clove contains significant amounts of an active component called eugenol, clove is an excellent source of manganese, a very good source of vitamin K and dietary fiber, and a good source of iron, magnesium, and calcium, also high in omega 3 fatty acid.

Eugenol is very effective in prevention of toxicity from environmental pollutants like carbon tetrachloride,

Anaesthetic and Anti-bacterial

Eugenol and other component of clove beta-caryophyllenecombine to make clove a mild anaesthetic as well as an anti-bacterial agent. Of these reasons and peculiar odour of the clove oil it is being used in the mouth washes and throat clearing agents.

Anti-inflammatory Substance

Eugenol, the primary component of clove's volatile oils contributes to clove's anti-inflammatory properties. Clove also contains a variety of flavonoids, including kaempferol and rhamnetin, which are effective in overcoming the joint inflammation.

Cumin (*Cuminum cyminum*)

Bitter cumin is used extensively in traditional medicine to treat a range of diseases from vitiligo to hyperglycemia. It is considered to be antiparasitic and antimicrobial and science has backed up claims of its use to reduce fever or as a painkiller. It also contains high levels of phenolic compounds from plants, especially polyphenolic antioxidants, which prevent oxidation of liposome membrane preventing the cell damage and offer complete protection against DNA damage.

References

1. Pearl, G. G. (2004). Inedible. In Jensen, W., Devine, C. and Dikemann, M. (Eds.), *Encyclopedia of Meat sciences*. pp. 112. London, UK: Elsevier Science Ltd.
2. Honikel, K. O. (2011). Composition and calories. In L. M. L. Nollet and F. Toldrá (Eds.), *Handbook of analysis of edible animal by-products*. pp. 105. Boca Raton, FL, USA: CRC Press.
3. Jimenez-Colmenero, F., Carballo, J. and Cofrades, S. (2001). Healthier meat and meat products: Their role as functional foods. *Meat Science*. 59: 5.
4. Biesalski, H. K. (2005). Meat as a component of a healthy diet - Are there any risks or benefits if meat is avoided in the diet? *Meat Science*. 70: 509.

Fluoride Toxicity: A Major Global Problem in Domestic Animals

Aamrapali Bhimte, Archana Jain, Jyotsana Shakkarpude, Shweta Rajoriya and Ranjit Aich
Dept. of Veterinary Physiology and Biochemistry, College of Veterinary Science and Animal Husbandry,
NDVSU, Mhow, Madhya Pradesh.

Introduction

Environmental pollution is a major global problem which poses real threats to human and animal health besides causing imbalance in the ecosystem. Natural geological sources and increased industrialization have contributed greatly to the increasing incidence of fluoride-induced human and animal health issues. In animals and human beings, it exerts adverse effects mainly through the attenuation of antioxidant defense mechanism and chelation of enzymatic cofactors.

The variability and presence of fluoride depends upon the location. It was found that fluoride is present in the soil within the range of 10-1000 parts per million (ppm). However, in water it ranges from 0.5 to 2000 ppm. This incident depends upon the sources of water. According to World Health Organization (WHO), fluoride exposure to animals above the 1.5 ppm, set at chronic fluoride toxicity. Through water exposure, this type of toxicity is going to endemic in most of the countries across the world.

Fluoride toxicity in animals has become an important toxicologic problem in many parts of the world due to the expansion of certain types of industrial operations that emit excessive fluorides into the atmosphere in areas of animal habitat. Fluorosis is a chronic disease caused by the body weight produces clinical signs after continued ingestion of small but toxic amounts of ingested fluoride in the diet or drinking water over a long duration. Animals normally ingest low levels of fluoride with no adverse effects. Small amounts of fluoride may even have beneficial effects, but when excessive amounts are ingested for prolonged periods of time, it will cause the adverse effects.

Etiological Factors

Fluoride is an electronegative element, distributed ubiquitously as fluoride in nature. Water is the major medium of fluoride intake by humans and animals. Animals normally ingest small amounts of various fluorides in their diets with no harmful effects, but excessive ingestion can be damaging. The domestic animals any consume naturally occurring high-fluoride components of the diet or from the ingestion of forage that has been contaminated by industrial fluoride emissions. In case of pregnant cows, the fluorine content of leading the cumulative effect, while acute poisoning bones of new born calves depend on the dam's intake occurs due to inhalation of fluorine containing gases by of fluorine

in the last 3-4 months of pregnancy. Smoke, vapours, dust which eliminated from industrial area. Copper, glass, enamel, iron, steel and super phosphate these are the common causes which are present in drinking water and soil.

Toxicity in Animals

Domestic animals, buffaloes, cows and goats are affected by fluorosis. Many sources may contribute to the total fluoride intake of animals. Various factors influence biologic responses of animals to excessive ingested fluorides. Signs and lesions of fluoride toxicity in animals have been characterized. The lesions observed in fluoride toxicity in various species of animals are similar.

Fluoride from industrial air bone particulates containing fluoride are settled over leafy surface of plants which in turn consumed by grazing cattle. Since, fluoride not readily transported through plant tissues, leafy portions tend to concentrate fluoride, and thus the herbivores are expected to consume more fluoride than grainivores in industrial areas.

In case of acute intoxication there is ruminal stasis, constipation or gastroenteritis occurs due to irritation of stomach with the formation of hydrofluoric acid. Nervous signs are characteristic and include muscle tremor, weakness, pupillary dilatation, hyperesthesia, and constant chewing. Tetany and collapse follow and death usually occurs within few hours. One of these, dental fluorosis, characterized by enamel mottling appears clinically at the level of 1.5 ppm fluoride in drinking water.

Chronic fluoride intoxication of man and animals is a health problem worldwide and is endemic in areas where fluoride is quite high in drinking water and aggravates in the form of fluorosis.

Geological Distribution of Fluorosis in Madhya Pradesh

In Madhya Pradesh severe fluorosis has been

noticed in bovines due to contamination of herbage due aluminum smelter, brick work and super phosphate manufacturing unit.

Fluorosis is now going to be a severe problem throughout the globe due to toxic effects of fluoride (F) on both plants and animals.

Hot Spots in Different Areas of Madhya Pradesh

S. No.	Districts	Fluoride Range (ppm)	
		Max.	Min.
1	Shivpuri	0.21	3.89
2	Gwalior	0.21	6.20
3	Khargone	0.10	1.58
4	Betul	0.28	0.68
5	Seoni	0.21	14.20
6	Mandla	0.89	3.30
7	Bhopal	0.19	2.69
8	Raisen	0.00	1.25
9	Sehore	0.26	1.25
10	Vidisha	0.36	4.43
11	Jabalpur	0.00	5.00
12	Dhar	0.78	4.07
13	Jhabua	0.00	13.86
14	Shajapur	0.12	0.46

Control and Prevention

Supplementation with calcium carbonate, aluminum salts, magnesium metasilicate, or boron will reduce absorption or enhance excretion. It is recommended that livestock consume supplements and mineral mixes containing <1% fluoride content. If it is impractical to limit fluoride exposure, raising species with a relatively short production life, such as poultry, pigs, or sheep, should be considered. Reducing fluoride exposure of young or pregnant animals may limit the development of chronic fluorosis.

Feedback Form

Dear Reader,

This is our twenty ninth issue of “**Raksha Technical Review**” and your valuable feedback will motivate the editorial team to make the next issue better. To help us continuously improve and bring you the technical magazine that you enjoy reading, we invite your feedback in the format below. Please fill up this form and send back to us with your passport size photograph or soft copies through e-mail.

Looking forward to your early response.

D. Prabhakar
Manager - CRM & CSR
Indian Immunologicals Limited
Road No. 44, Jubilee Hills,
Hyderabad - 500033. India.
Email: rtr@indimmune.com
Toll Free Number: 1800 212 999977

The rating scale is 1 to 5 (1 is poor and 5 is excellent)

1. Appearance

1	2	3	4	5
---	---	---	---	---

2. Article

1	2	3	4	5
---	---	---	---	---

3. Information and content

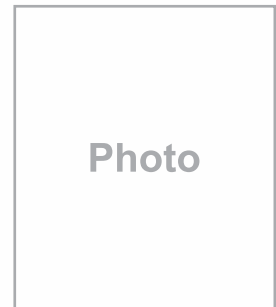
1	2	3	4	5
---	---	---	---	---

4. Language and vocabulary

1	2	3	4	5
---	---	---	---	---

5. Quality of photographs

1	2	3	4	5
---	---	---	---	---



- **Appearance:** Color combination, design and layout
- **Article:** Reputation of the author, relevance of the topic of the column and value addition to you.
- **Information and content:** Order of articles, quality of articles and relevance of the information
- **Language and vocabulary:** Spelling, grammatical correctness and appropriateness of vocabulary
- **Quality of the photographs:** Colour, contrast, placement and relevance

Suggestions, if any:

.....

.....

.....

.....

Name:

Signature:

Designation:

Location:

Address:

Mob. No.:

E-mail ID:

Guidelines to Authors

Raksha Technical Review recommends the following Guide lines to authors while submitting their manuscripts. These Guide Lines are essentially adopted from standard international journals to keep the quality of the material published.

Introduction

Raksha Technical Review is a comprehensive and pre-eminent journal for the Veterinary profession which would like to act as an interface between academics, those in Research and Development and Veterinary profession.

Types of paper: Raksha Technical Review publishes primary research papers, review articles, short communications, letters and commentary. Contact details for submission: Papers should be submitted to the Editor or Publisher using the following email address:

rtr@indimmune.com

Page charges: We do not levy any page charges for publishing the articles.

PLEASE NOTE

Ethics in Publishing: Send only original articles. The work described in your article must have been carried out by you in accordance with the guidelines in vogue in your institute and by following all statutory requirements. Manuscripts submitted as review or commentary must be your own informed opinion on the subject.

Conflict of interest: All authors must disclose any financial and personal relationships with other people or organizations that could inappropriately influence (bias) their work. Examples of potential conflicts of interest include employment, consultancies, stock ownership, honoraria, paid expert testimony, patent applications / registrations, and grants or other funding.

Submission declaration and verification: Submission of an article implies that the work described has not been published previously (except in the form of an abstract or as part of a published lecture or academic thesis) , that it is not under consideration for publication elsewhere, that its publication is approved by all authors and tacitly or explicitly by the responsible authorities where the work was carried out, and that, if accepted, it will not be published elsewhere in the same form, in English or in any other language.

Contributors: Each author is required to declare his or her individual contribution to the article: all authors must have materially participated in the research and /or article preparation, so roles for all authors should be described. The statement that all authors have approved the final article should be true and included in the disclosure.

Authorship: All authors should have made substantial contribution to all of the following: (1) the conception and design of the study, or acquisition of data, or analysis and interpretation of data, (2) drafting the article or revising it critically for important intellectual content, (3) final approval of the version to be submitted.

Copyright: The articles published in Raksha Technical Review will be copy righted to M/S Indian Immunologicals Ltd., and the should not be reproduced without written permission from the publishers

Language and language services : Please write your text in good English (American or British usage is accepted, but not a mixture of both)

Use of word-processing software: It is important that the file be saved in the native format of the word processor used. The text should be in single-column format. Keep the layout of the text as simple as possible. Most formatting codes will be removed and replaced on processing the article. In particular, do not use the word processor's options to justify text or to hyphenate words. However, do use bold face, italics, subscripts, superscripts etc. When preparing tables, if you are using a table grid, use only one grid for each

Individual table and not a grid for each row. If no grid is used, use tabs, not spaces, to align columns. The electronic text should be prepared

in a way very similar to that of conventional manuscripts.

Essential title page information

- Title concise and informative. Titles are often used in information-retrieval systems. Avoid abbreviations and formulae if possible
- Author names and affiliations. Where the family name may be ambiguous (e.g., a double name), please indicate this clearly. Present the authors' affiliation addresses (where the actual work was done) below the names. Indicate all affiliations with a lower-case superscript letter immediately after the author's name and in front of the appropriate address. Provide the full postal address of each affiliation.
- Corresponding author. Clearly indicate who will handle correspondence at all stages of refereeing e-mail address and the complete postal address. Contact details must be kept up to date by the corresponding author.
- Present/permanent address. If an author has moved since the work described in the article was done, or was visiting at the time, a "Present address" (or 'Permanent address") may be indicated as a footnote to that author's name. The address at which the author actually did the work must be retained as the main, affiliation address. Superscript Arabic numerals are used for such footnotes.

Abbreviations: Define abbreviations that are not standard in this field in a footnote to be placed on the first page of the article. Such abbreviations that are unavoidable in the abstract must be defined at their first mention there , as well as in the footnote. Ensure consistency of abbreviations throughout the article.

Footnotes: Footnotes should be used sparingly. Number them consecutively throughout the article, using superscript Arabic numbers.

Tables: Number tables consecutively in accordance with their appearance in the text. Place footnotes to tables below the table body.

References: Citation in text

Please ensure that every reference cited in the text is also present in the reference list (and vice versa).

Reference style

Text: Indicate references by number(s) in brackets in line with the text. The actual authors can be referred to, but the reference number(s) must always be given.

List: Number the references (numbers in brackets) in the list in the order in which they appear in the text.

Example: 1) Crowther, J.R. and Abu Elzein, E.M.E. (1979). Detection and quantification of Foot and Mouth Disease virus by enzyme labeled immunosorbent assay . J.Gen.Virol.42:747

Submission checklist: The following list will be useful during the final checking of an article prior to sending it to the journal for review. Please consult this Guide for Authors for further details of any item.

Ensure that the following items are present: One Author designated as **corresponding Author:**

- Email address
- Full postal address
- Telephone and fax numbers
- All necessary files are uploaded
- All figures and photographs with captions
- All tables (including title, description, footnotes)

Further considerations

- Manuscript has been "spell checked" and "grammar-checked"
- References in the correct format for this journal
- All references mentioned in the Reference list are cited in the text, and vice versa
- Permission has been obtained for use of copyrighted material from other sources (including the Web)

Disclaimer: Indian Immunologicals Ltd. shall not be responsible for the opinion expressed by any of the authors contributing to Raksha Technical Review. Indian Immunologicals Ltd., does not necessarily endorse all the views expressed by authors.

Products

Animal Health Formulations

NIMOVET
(Nimesulide Inj., Bolus., Gel)

OXFENVET
(Oxfendazole - Broad Spectrum Dewormer)

CLOSITEL
(Closantel Oral Solution)

IVECTIN
(Ivermectin - Inj, Bolus and Tab)

XYLAXIN
(Xylazine HCl - Pre Anesthetic, Sedative, Muscle relaxant and analgesic)

TIKKIL
(Cypermethrin - the Acaricide in Powder Form and Liquid form)

BOVOPLEX ORAL
(Health Tonic)

INIMOX FORTE
(Amoxicillin Sodium + Sulbactam Sodium Inj)

BOVICEF
(Ceftiofur Inj., Antibiotic against Metritis)

SEFTRIVET Forte
(Cefoperazone+ Sulbactam Inj)

GARBHAMIN
(Chelated Minerals & Coated Vitamins)

PET FORTE
(A Calcium Supplement)

HEX-D
(Herbal Digestive Powder)

PROWOMB
(Herbal Uterine Syrup)

TIKKIL RAZ
(Amitraz)

ZUSPRAY
(A Herbal Spray for Open wounds)

ELP
(Toldimfos sodium)

GLUFLU
(Flunixin Meglumine Inj)

MORBAXIN
(Morboflaxacin Inj)

TIKKIL POWER
(Flumethrin Pour-On Ectoparasiticide)

GYROFLOX
(Enrofloxacin - Oral, inj.)

GYROFLOX BH DS
(Combi. of Enrofloxacin and Bromhexine hydrochloride)

MOXEVICT
(Moxidectin Inj)

MECTADOR
(Doramectin Inj)

PROZOFF
(Buparvaquone Inj)

Vetalexin
(Cephalexin Monohydrate-Oral)

Bovoplex CC
(Bcomplex Inj with liver extract and Choline Chloride)

Calgonate Inj
(Calcium Borogluconate Inj)

Tikkil Shampoo
(Cypermethrin with conditioner shampoo)

FOLYSON Inj
(Human Chorionic Gonadotrophin Inj)

IVECTIN-T
(Ivermectin +Triclabendazole Oral)

MIPHOCAL
(Calcium Magnesium Borogluconate Inj)

VETALBEN R
(Albendazole and Rafoxanide Suspension)

Imidectin spot-on
(Imidacloprid+ Moxidectin spot-on for endo and ecto parasites)

Corforce
(Pimobendan 5mg for management of CHF in dogs)



Biologicals

RAKSHA OVAC
(Foot and Mouth Disease Vaccine)

RAKSHA OVAC ULTRA
(NSP Free Foot and Mouth Disease Vaccine)

RAKSHA TRIOVAC
(FMD, HS & BQ Combined Vaccine)

RAKSHA BIOVAC
(FMD & HS Combined Vaccine)

RAKSHA HS
(Adjuvanted Vaccine of Pasteurella Multocida)

RAKSHA HS BQ
(Combined Vaccine for HS & BQ)

RAKSHAVAC T
(Theileriosis Vaccine)

BRUVAX PLUS
(Brucellosis Vaccine S19)

RAKSHARAB
(Cell Culture Antirabies Vaccine)

STARVAC R
(Cell Culture Antirabies Vaccine)

STARVAC 7
(Multicomponent Vaccine)

MEGAVAC 6
(Multicomponent Vaccine)

MEGAVAC P
(Canine Parvovirus Vaccine)

MEGAVAC CC
(Canine Coronavirus Vaccine)

MEGAVAC 7
(Multicomponent Vaccine)

RAKSHA ET
(Clostridium Perfringens Type D Vaccine)

Raksha PPR
(Peste Des Petitis Ruminants Vaccine)

RAKSHA SP
(Sheep Pox Vaccine)

RAKSHA BLU
(Pentavalent Bluetongue Vaccine)

BRUVAX RB 51
(Brucellosis Vaccine RB 51)

RAKSHA ET+TT
(Combined Vaccine for Enterotoxemia & Tetanus)

RAKSHA
(Gel vaccine against FMD)

CYSVAX
(Porcine Cysticercosis Vaccine)

RAKSHA Class
(Classical Swine Fever Vaccine)

Nutraceuticals

CalSagar Plus
(Granular Calcium feed supplement with Vitamin D3 and Herbs)

Gouvit Chelated
(Chelated Minerals and Vitamins for higher productivity)

R Vita
(Raksha Vitamin and Mineral Premix)

GouMix
(Area specific mineral mixture)

GouMix TM Chelated
(Chelated trace minerals)

Gousac
(Rumen specific probiotic live yeast culture)

Gousac Power
(Rumen specific lyophilized probiotic live yeast culture)

Kshir Sagar Chelated
(Glycinated trace minerals with Chromium Propionate and Herbs)

Kshir Sagar
(Calcium enriched high energy milk booster for improved milk production)

Vetfen 600
(Medicated Feed Pellets containing Fenbendazole)

Trisomix
(Nutritional supplement for Mastitis management)

4P
(Multi nutrient feed supplement for transition period)

Goudhara Shakti
(Bypass fat with calcium and herbs)

ParvoGuard
(Chicken egg yolk protein)

Flexicruz
(Joint Support supplement)

Nephro K9
(Supplement for management of Chronic Renal Failure in cats and dogs)

Shinikoat
(Omega Fatty Acid supplement)

Human Health

Biologicals

ABHAYRAB
(Purified Vero Cell Rabies Vaccine)

ABHAY-TOX
(Tetanus Toxoid Vaccine)

ELOVAC-B
(Recombinant Hepatitis B Vaccine)

VAXTAR-5
(Pentavalent (DTwP-HepB-Hib) Vaccine)

Formulations

VIVAFIT
(Antioxidant and Omega 3 Fatty Acids capsule)

VIVAGUT
(Pre & Probiotic Capsules)

VIVACAD
(Calcium, Calcitriol & Vitamins capsule)

VIVAFLORA
(Pre and Probiotics capsule for Female Urogenital Health)

HERVIBE
(Vitamins, Minerals, Antioxidants, Trace Elements, Evening Primrose Oil, L-Carnitine and Green Tea extract for women health)

MOMSVIBE
(Vitamins, Minerals and Amino Acid Tablet for Pre-Conception, Pregnancy & Lactation)

OVACORE
(DHEA, Coenzyme Q 10 & Melatonin Capsule for Pre-Conception)

Gonadotropins

INCEPTOVA HCG
(Human Chorionic Gonadotropin for injection)

INCEPTOVA HMG
(Human Menopausal Gonadotropin for injection)

INCEPTOVA FSH
(Follicle Stimulating Hormone for injection)

INCEPTOVA rFSH
(Recombinant Follicle Stimulating Hormone for injection)

PROGESHIELD
(Hydroxyprogesterone Injection)

Vision

“Shaping Global Healthcare by Spearheading the One Health Initiative”.

Mission

“To Innovate, Produce and Market Quality Healthcare Products and Services to Improve and Extend lives”.



INDIAN IMMUNOLOGICALS LIMITED

Road No. 44, Jubilee Hills, Hyderabad, Telangana - 500033. India.

Tel: +91 8466924444, Tel: +91 7997915555, Tel: 040 - 67682200

www.indimmune.com

Toll Free Number: 1800 212 999977