



Control and Management of Diseases of Domestic Animals in the North Eastern Hilly Region of India with Special Reference to Mithun (*Bos frontalis*)

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Assistance

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**ICAR – National Research Centre on Mithun
Medziphema, Nagaland**

In collaboration with

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Hyderabad, Telengana 500030**

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MESSAGE

It gives me immense pleasure to introduce this comprehensive guide on the "Prevention, Control, and Management of Different Bovine Diseases with Special Reference to Mithun (*Bos frontalis*)." The book is a vital contribution to the field of veterinary science and livestock management, especially in the context of Mithun, a unique bovine species of great cultural, economic, and ecological significance in the North eastern states of India. Mithun, known as the "Cattle of the Mountain," holds a special place in the agrarian societies of the region. Despite its importance, there has been a notable lack of focused literature on the health and management of this species. This book aims to fill that gap by providing valuable insights into the various diseases affecting bovines, particularly Mithun, and offering practical strategies for their prevention, control, and management.

The book meticulously covers a range of topics, from common infectious and parasitic diseases to emerging zoonotic threats and nutritional deficiencies. Each chapter is crafted with the expertise of seasoned veterinarians, researchers, and field practitioners who have first-hand experience dealing with these challenges.

I congratulate the authors, editors, and all contributors for their diligent efforts in bringing this important work to fruition. I hope this book will inspire further research and innovation in the field of veterinary science and play a pivotal role in advancing the health and welfare of our bovine population.

I want to extend my appreciation to, team of ICAR-National Research Centre on Mithun, Nagaland & EAAS Centre, MANAGE, Hyderabad for the tremendous effort in compiling this e-book. I also thank the authors, editors, and designers who have contributed to this e-book creation.

A handwritten signature in blue ink that reads "Shekara".

Dr. P. Chandra Shekara
(Director General, MANAGE)



FOREWORD

The North Eastern Hilly Region of India, encompassing the states of Arunachal Pradesh, Assam, Manipur, Meghalaya, Mizoram, Nagaland, Sikkim, and Tripura, is characterized by its unique topography, climate, and biodiversity. This region, often referred to as the "Paradise Unexplored," is home to an incredible variety of flora and fauna, many of which are endemic to the area. The indigenous livestock species, particularly Mithun (*Bos frontalis*), hold a special place in the cultural and economic fabric of these states. Mithun, is not only a symbol of prosperity and wealth but also plays a crucial role in the socio-economic and cultural life of the local tribes. The domestication of Mithun dates back centuries and is deeply intertwined with the traditions and rituals of the local communities. These semi-domesticated animals are primarily reared under free-range conditions in the forests, where they graze on natural vegetation. Given their unique management system and the environmental conditions of the North Eastern Hilly Region, the control and management of diseases in Mithun pose significant challenges. The lack of infrastructure, veterinary services, and awareness among farmers further complicates the situation. This book aims to provide a comprehensive overview of the control and management of diseases affecting domestic animals in the North Eastern Hilly Region of India, with a particular focus on Mithun. The content is divided into several key sections, each addressing different aspects of disease management, including epidemiology, diagnostic techniques, preventive measures, and treatment protocols. Additionally, the use of ethno-veterinary practices, which involve the use of medicinal plants and traditional remedies, has also been covered. Integrating these traditional practices with modern veterinary medicine can enhance the overall effectiveness of disease control programs. The control and management of diseases in domestic animals, particularly Mithun, in the North Eastern Hilly Region of India, require a holistic and integrated approach. Addressing the challenges of geographical isolation, limited veterinary services, and lack of awareness among farmers is essential for the success of disease management programs.

The development and dissemination of these advanced practices represent a significant step towards safeguarding the health of Mithuns and ensuring the continued prosperity of the tribal communities that depend on them. Through collaborative efforts and a commitment to adopting these practices, the resilience and well-being of Mithun herds can be maintained, securing a thriving future for both the animals and the people.

A handwritten signature in blue ink that reads "Girish Patil S".

Girish Patil S
(Director, NRC on Mithun)

PREFACE

In the verdant hills and valleys of Northeastern India, where tradition dances hand in hand with nature's bounty, lies a livestock of immense significance: the majestic Mithun (*Bos frontalis*). Within the tapestry of tribal life in Arunachal Pradesh, Nagaland, Manipur, Mizoram, and certain areas of Assam, the Mithun stands as a symbol of prosperity, deeply ingrained in the socioeconomic and cultural fabric of these regions. This majestic animal is not only a cultural icon but also an essential component of the livelihoods of these tribal communities. Mithun husbandry, alongside traditional livestock, plays a pivotal role in sustaining the economic well-being of these regions. Northeastern India, with its distinct agro-geo-climatic circumstances, is regarded as a biodiversity haven. Factors such as high relative humidity, specific soil pH ranges, fluctuating temperatures, and substantial rainfall create an environment conducive to the propagation of various parasites. The region's unique topography, coupled with a lack of awareness about disease predisposing factors, exacerbates the incidence of illnesses among Mithuns. The combination of these environmental conditions results in a higher prevalence of parasitic diseases, which can severely impact Mithun health and productivity.

The ICAR - National Research Centre (NRC) on Mithun, located in Medziphema, has undertaken proactive steps to study the various diseases affecting Mithuns in the Northeastern Himalayan region. This includes extensive documentation of microbes incidences and their respective treatments, with a particular focus on controlling of viral, bacterial, helminths, protozoans, and ticks that infest Mithuns. Such advancements are crucial for sustaining Mithun husbandry in this geographical region. The dissemination of this acquired knowledge through educational programs and practical guidelines empowers farmers to implement preventive and control measures. Collaboration with local veterinarians ensures that farmers receive the necessary support and expertise to manage parasite infestations.

Proactive measures taken by the ICAR - NRC on Mithun through research, capacity building, large scale awareness programs and organizing health camps, combined with the commitment of tribal communities to preserve and promote Mithun husbandry, hold the promise of a sustainable future. Through continued research, education, and the application of effective control strategies, the health and productivity of Mithuns can be safeguarded, ensuring that these majestic creatures remain a cornerstone of prosperity and cultural heritage in Northeastern India.

Editors

Chapter 1

Overview of the Mithun Sector in Northeast India: Diseases, Prevention, and Challenges

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INTRODUCTION TO MITHUN (*Bos frontalis*)

Mithun, also known as “Gayal,” is a semi-domesticated bovine species that holds a unique and forested regions of Arunachal Pradesh, Nagaland, Manipur, and Mizoram, mithun **are well** adapted to the temperate and subtropical climates of these areas. Revered for their role in traditional ceremonies, social exchanges, and as a source of high-quality meat, mithuns are integral to the livelihoods of tribal communities. Their management involves a blend of traditional practices and modern veterinary care, highlighting the dynamic interaction between cultural heritage and contemporary animal husbandry.

1.2 DISTRIBUTION AND ECONOMIC IMPORTANCE

1.2.1 Geographical Distribution

Mithun are primarily found in the tribal belts of Arunachal Pradesh, Nagaland, Manipur, and Mizoram. The geographic distribution of mithun aligns with their adaptation to the forested, hilly terrains where they thrive on the rich foliage and natural vegetation. Each state has developed unique management practices reflecting local environmental conditions and cultural traditions.

- **Arunachal Pradesh:** The largest population of mithun is found here, where they are commonly managed under traditional free-range systems. The diverse flora of the region provides ample forage, supporting mithun husbandry.
- **Nagaland:** Mithun are a vital part of the socio-cultural fabric, used extensively in social ceremonies and community exchanges. The Naga tribes have developed specific rites and rituals involving mithun, underscoring their cultural importance.

- **Manipur:** In Manipur, mithun are less common but still significant in the high-altitude areas. The state's diverse topography requires adaptive management practices to cater to the varying climate zones.
- **Mizoram:** The Mizos have a deep-rooted tradition of mithun rearing, often integrating them into agroforestry systems, which provide a sustainable source of forage and enhance biodiversity.

1.2.2 ECONOMIC SIGNIFICANCE

Mithun contribute significantly to the rural economies of Northeast India through their roles in agriculture, ceremonial functions, and as a source of high-value meat. Their economic impact can be understood in several dimensions:

- **Ceremonial and Social Value:** Mithun are indispensable in traditional ceremonies, such as weddings, festivals, and tribal rites. They serve as a symbol of wealth and social status, and their exchange is a means of establishing and reinforcing social bonds within communities.
- **Meat Production:** Mithun meat, known for its rich flavor and nutritional value, commands a premium price in local markets. It contributes to food security and nutritional needs in tribal diets, providing a crucial source of protein and other nutrients.
- **Agricultural Utility:** In addition to their direct economic value, mithun are used in barter trade and serve as a form of capital or wealth reserve. They also contribute indirectly to agriculture through manure production, which enhances soil fertility and supports crop cultivation.
- **Cultural Heritage:** Mithun are embedded in the cultural narratives of the tribal communities, with their management practices reflecting a deep understanding of local ecosystems and environmental stewardship.

1.3 CHARACTERISTICS AND HUSBANDRY

1.3.1 Physical Characteristics

Mithuns are distinguished by their robust build, which combines strength with adaptability to the challenging terrains of Northeast India. Key physical characteristics include:

- **Size and Weight:** Mithun bulls typically weigh between 500 to 800 kg, while cows weigh between 400 to 600 kg. Their size and muscular build are indicative of their evolutionary adaptation to the forested and hilly landscapes.
- **Coat and Color:** The coat color of mithuns ranges from dark brown to black, often with lighter markings on the underbelly and legs. Their thick coat provides protection against the elements, particularly in colder, high-altitude regions.
- **Head and Horns:** Mithun have a broad forehead and distinctively curved horns, which are used in social interactions and defense. The horn structure varies between individuals and can serve as a distinguishing feature in herd management.
- **Feet and Gait:** Short, sturdy legs and strong hooves enable mithuns to navigate the rugged terrain of their natural habitat. Their gait is well-adapted for foraging and moving through dense forests and steep slopes.

1.3.2 HUSBANDRY PRACTICES

Mithun husbandry in Northeast India combines traditional knowledge with contemporary animal management practices, reflecting a holistic approach to livestock care:

- **Traditional Free-Ranging System:** The predominant system of mithun rearing involves free-ranging in forested areas. This method leverages the animals' natural grazing habits, allowing them to forage on a diverse array of vegetation. Free-ranging reduces the need for intensive feeding and housing, but it also poses challenges in monitoring health and controlling diseases.
- **Semi-Intensive Farming:** Some farmers have adopted semi-intensive systems, where mithuns are provided with shelter and controlled grazing areas. This approach offers a balance between traditional and modern practices, facilitating better health monitoring and more efficient use of forage resources. Semi-intensive farming often includes rotational grazing and supplementary feeding during lean seasons.
- **Shelter and Housing:** In areas where semi-intensive farming is practiced, mithuns are provided with simple shelters constructed from locally available materials. These shelters offer protection from extreme weather conditions and predators, contributing to improved animal welfare.
- **Feeding and Nutrition:** Mithuns primarily feed on natural vegetation, including bamboo, grasses, shrubs, and tree leaves. Supplementary feeding may include

agricultural by-products, salt licks, and mineral supplements, particularly during dry seasons when forage quality declines.

- **Breeding Practices:** Traditional breeding practices are often employed, with minimal intervention. However, there is growing interest in selective breeding to enhance desirable traits such as disease resistance and productivity. Community-based breeding programs are emerging as a means to improve genetic diversity and herd quality.

1.4 COMMON DISEASES IN MITHUN

The health and productivity of mithun are influenced by a range of diseases, which can be broadly categorized into infectious diseases, parasitic infestations, and nutritional deficiencies. Effective disease management is essential for sustaining mithun populations and maximizing their economic value.

1.4.1 INFECTIOUS DISEASES

- **Foot and Mouth Disease (FMD):** FMD is a highly contagious viral disease affecting cloven-hoofed animals, including mithuns. It is characterized by fever, blisters in the mouth and feet, excessive salivation, and lameness. FMD outbreaks can cause significant economic losses due to reduced productivity, mortality, and trade restrictions. Control measures include vaccination, movement control, and biosecurity practices.
- **Haemorrhagic Septicaemia (HS):** Caused by *Pasteurella multocida*, HS is prevalent during the rainy season. Symptoms include high fever, swelling in the neck and submandibular regions, respiratory distress, and rapid progression to death if untreated. Vaccination and prompt antibiotic treatment are essential for controlling HS outbreaks.
- **Black Quarter (BQ):** BQ, also known as Blackleg, is a bacterial disease caused by *Clostridium chauvoei*. It leads to severe muscle necrosis, toxemia, and rapid death. Symptoms include swelling, crepitation in affected muscles, and a high fever. Vaccination and proper carcass disposal are key preventive measures.
- **Brucellosis:** A zoonotic disease caused by *Brucella abortus*, brucellosis affects reproductive health, causing abortions, retained placenta, and infertility. Control involves vaccination, culling infected animals, and strict hygiene practices to prevent transmission.

- **Tuberculosis (TB):** Caused by *Mycobacterium bovis*, TB is a chronic disease that affects the respiratory system, leading to coughing, weight loss, and emaciation. Control measures include testing, segregation of infected animals, and improved management practices to reduce exposure.

1.4.2 PARASITIC INFESTATIONS

- **Internal Parasites:** Mithuns often suffer from gastrointestinal parasites, including liver flukes (*Fasciola spp.*) and roundworms (*Strongyles spp.*). These parasites cause malnutrition, anemia, weight loss, and reduced productivity. Control strategies involve regular deworming, pasture management, and rotational grazing to break the parasite life cycle.
- **External Parasites:** Ticks, lice, and mites are common external parasites affecting mithuns. They cause skin irritation, blood loss, and secondary infections, leading to reduced weight gain and overall health. Control measures include the use of acaricides, insecticides, and improved housing sanitation.
- **Trypanosomiasis:** Also known as Surra, this disease is caused by *Trypanosoma evansi* and transmitted by biting flies. It affects the blood, causing fever, anemia, and weight loss. Control involves vector control, chemoprophylaxis, and treatment with trypanocidal drugs.

1.4.3 NUTRITIONAL DEFICIENCIES

- **Mineral Deficiency:** Deficiencies in essential minerals such as copper, zinc, and selenium can lead to poor growth, reproductive issues, compromised immune function, and specific conditions like enzootic ataxia and swayback. Providing mineral licks and supplements is crucial for preventing these deficiencies.
- **Vitamin Deficiency:** Lack of vitamins, especially A and E, can result in night blindness, reduced fertility, and increased susceptibility to infections. Ensuring a diet rich in natural forage and supplements during periods of scarcity is essential for maintaining health.
- **Protein-Energy Malnutrition:** Inadequate intake of protein and energy due to poor-quality forage or insufficient supplementary feeding can lead to stunted growth, weight loss, and decreased productivity. Balanced nutrition, including high-quality forage and

supplemental feeds, is critical during the dry season and periods of increased metabolic demand.

1.5 PREVENTION AND CONTROL STRATEGIES

Effective prevention and control of diseases in mithuns require a multifaceted approach, incorporating vaccination, deworming, nutritional management, and biosecurity measures. These strategies are essential for maintaining herd health, productivity, and economic viability.

1.5.1 VACCINATION PROGRAMS

Implementing systematic vaccination programs is crucial in controlling infectious diseases and preventing outbreaks:

- **FMD Vaccination:** Vaccination against FMD is administered biannually in regions with a high risk of outbreaks. Regular booster doses are necessary to maintain immunity in the herd.
- **HS and BQ Vaccination:** Annual vaccination before the onset of the monsoon season is recommended to prevent Haemorrhagic Septicaemia and Black Quarter. Vaccines are administered based on local epidemiological data and risk assessment.
- **Brucellosis Vaccination:** Vaccination of female mithuns before they reach breeding age helps control brucellosis. Infected animals should be culled to prevent the spread of the disease within the herd.
- **TB Testing and Control:** Regular testing for tuberculosis, followed by segregation or culling of infected animals, is essential for preventing the spread of TB. Improved housing and management practices can also reduce exposure.

1.5.2 DEWORMING PROTOCOLS

Regular deworming is essential to manage parasitic infections and improve overall health:

- **Internal Parasites:** Administering anthelmintics every three to four months helps control gastrointestinal parasites. Rotation of deworming agents is recommended to prevent resistance.

- **External Parasites:** Use of acaricides and insecticides, along with regular grooming and improved housing sanitation, helps control external parasites. Integrated pest management practices can enhance the effectiveness of control measures.

1.5.3 NUTRITIONAL MANAGEMENT

Providing a balanced diet with adequate minerals and vitamins is crucial for preventing nutritional deficiencies and supporting overall health:

- **Mineral and Vitamin Supplements:** Mineral licks and vitamin supplements should be readily available, especially during periods of forage scarcity. Supplementation with specific minerals and vitamins can prevent deficiencies and support reproductive health.
- **High-Quality Forage:** Ensuring access to high-quality forage and supplemental feeds during lean seasons helps maintain body condition and productivity. Integrating agroforestry systems can enhance forage availability and nutritional quality.
- **Protein and Energy Supplementation:** Providing protein and energy-rich supplements during critical periods, such as lactation and growth phases, supports metabolic demands and prevents malnutrition.

1.5.4 BIOSECURITY MEASURES

Enhancing biosecurity on farms is essential to prevent the introduction and spread of diseases:

- **Isolation of New Animals:** Quarantine new arrivals for a period of 2-4 weeks to monitor for signs of disease and prevent the introduction of pathogens into the herd.
- **Sanitation and Hygiene:** Regular cleaning and disinfection of shelters, feeding areas, and equipment help reduce the risk of disease transmission. Proper disposal of manure and carcasses is also crucial.
- **Movement Control:** Regulating the movement of animals, people, and equipment can minimize the spread of infectious diseases. Implementing strict protocols for entry and exit of farms can enhance biosecurity.
- **Community-Based Biosecurity:** Engaging the community in biosecurity practices and raising awareness about disease prevention can strengthen collective efforts to control diseases.

1.6 CHALLENGES IN MITHUN MANAGEMENT

Despite the cultural and economic significance of mithun, the sector faces several challenges that impede effective management and development. Addressing these challenges requires a comprehensive approach that integrates traditional practices with modern techniques and policy support.

1.6.1 DISEASE SURVEILLANCE AND DIAGNOSIS

- **Lack of Diagnostic Facilities:** Limited access to veterinary laboratories and diagnostic facilities hampers accurate disease diagnosis and timely intervention. Expanding diagnostic infrastructure and training veterinary personnel are essential for improving disease surveillance.
- **Inadequate Field Monitoring:** Insufficient veterinary infrastructure and personnel in remote areas result in inadequate field monitoring and delayed disease reporting. Strengthening veterinary networks and deploying mobile units can enhance field surveillance.
- **Emerging Diseases:** The emergence of new diseases and changing epidemiological patterns pose challenges to disease management. Continuous monitoring and research are needed to understand and address emerging threats.

1.6.2 VETERINARY CARE AND SERVICES

- **Limited Veterinary Access:** Remote locations and difficult terrains restrict the availability of veterinary services. Developing community-based veterinary programs and improving transportation infrastructure can enhance access to veterinary care.
- **Training Deficiency:** Lack of training programs for local farmers and para-veterinary workers on modern husbandry practices and disease management limits the effectiveness of interventions. Capacity-building initiatives are crucial for empowering local communities with knowledge and skills.
- **Resource Constraints:** Limited financial and logistical resources constrain the ability to provide comprehensive veterinary care. Policy support and investment in veterinary infrastructure are needed to address these constraints.

1.6.3 TRADITIONAL PRACTICES VS. MODERN TECHNIQUES

- **Resistance to Change:** Farmers often rely on traditional practices and may resist adopting new techniques due to cultural beliefs or lack of awareness. Promoting awareness and demonstrating the benefits of modern practices can encourage adoption.
- **Integration Challenges:** Integrating modern veterinary practices with traditional systems can be challenging but is essential for sustainable improvements in mithun health and productivity. Collaborative approaches that respect cultural traditions while introducing innovations are needed.
- **Knowledge Transfer:** Effective knowledge transfer mechanisms, including extension services, training programs, and community engagement, are essential for integrating modern techniques with traditional practices.

1.6.4 ENVIRONMENTAL AND ECOLOGICAL FACTORS

- **Deforestation:** Loss of forest cover due to agricultural expansion, logging, and infrastructure development affects the natural habitat of mithuns, reducing forage availability and increasing the risk of disease transmission. Conservation efforts and sustainable land management practices are needed to preserve mithun habitats.
- **Climate Change:** Variations in climate patterns, including changes in rainfall, temperature, and extreme weather events, impact forage availability and increase the incidence of certain diseases. Adaptation strategies, such as improved forage management and disease surveillance, are essential for mitigating the effects of climate change.
- **Biodiversity Loss:** The decline in biodiversity due to habitat fragmentation and environmental degradation can affect the availability of diverse forage species and disrupt ecological balance. Promoting agroforestry and conservation practices can enhance biodiversity and support mithun rearing.

1.6.5 ECONOMIC CONSTRAINTS

- **Limited Financial Resources:** Farmers often lack the financial resources to invest in improved husbandry practices, vaccines, and veterinary care. Access to credit, subsidies, and financial support can help farmers adopt better management practices.
- **Market Access:** Restricted market access, poor infrastructure for transporting livestock products, and limited market information hinder economic benefits from mithun

rearing. Developing market linkages and improving infrastructure can enhance economic opportunities for farmers.

- **Value Addition:** Limited opportunities for value addition, such as processing and branding mithun products, constrain the potential economic benefits. Promoting value-added products and developing market strategies can enhance the economic value of mithun rearing.

1.7 FUTURE PROSPECTS AND RECOMMENDATIONS

To address the challenges in mithun management and enhance the sector's contributions to rural economies and cultural heritage, several future prospects and recommendations can be considered:

1.7.1 ENHANCING DISEASE MANAGEMENT

- **Strengthening Veterinary Infrastructure:** Investment in veterinary clinics, diagnostic laboratories, and mobile veterinary units is needed to improve disease diagnosis and treatment. Expanding veterinary networks and providing training for veterinary personnel can enhance disease management capabilities.
- **Capacity Building:** Training programs for farmers, local veterinary staff, and para-veterinary workers on advanced husbandry practices, disease management, and biosecurity measures can empower communities with the knowledge and skills needed for effective mithun management.
- **Community-Based Health Programs:** Developing community-based health programs that involve local stakeholders in disease monitoring, vaccination campaigns, and health education can enhance disease control efforts and build community resilience.

1.8 CONCLUSION

The mithun sector in Northeast India represents a unique intersection of cultural heritage, economic significance, and environmental sustainability. Addressing the challenges in disease management, husbandry practices, and economic development requires a comprehensive approach that integrates traditional knowledge with modern techniques. By enhancing veterinary infrastructure, promoting sustainable practices, and engaging local communities, the

mithun sector can contribute to the livelihoods and cultural heritage of tribal communities while supporting broader goals of rural development and environmental conservation.

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Chapter 2

Foot and mouth disease: Status, Control and challenges

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Foot-and-mouth disease is one of the viral diseases of livestock affecting all the cloven-hoofed animals. It is caused by the *Aphthovirus* of the *Picornaviridae* family. The disease is usually not fatal in adult animals, but there is often high mortality in young animals due to myocarditis. The virus is quasi species in nature and undergoes rapid mutation. There are seven antigenically and genetically distinct serotypes *viz.* A, O, C, SAT1, SAT2, SAT3, and Asia1; and they do not show cross-protection in vaccinated animals. In India, FMDV O, A, and Asia1 serotypes are reported and antigenically matched strains of all the three serotypes are mandatorily included in the commercial vaccine formulation. The genome is a linear positive-sense single-stranded RNA measuring about 8.5 kb in size encoding four structural proteins (VP1-4) and varying numbers of non-structural proteins (NSPs).

MODE OF TRANSMISSION

The FMD virus (FMDV) is shed in all secretions and excretion from infected animals. Transmission can be either direct or indirect contacts between susceptible and infected animals. The FMDV can be introduced to healthy animals through any of the following points-

- Through contact with clinically affected animals.
- Through contaminated inanimate vectors such as vehicles, fodders, utensils, equipments etc.,
- Through air. Infected animals can secrete large amount of aerosol virus through exhaled air, which can infect other animals via the respiratory or oral routes. The virus can travel up to 60 km overland and 300 km by sea.
- All secretions and excretions from the infected animal such as saliva, faeces and urine.
- The virus may be present in milk and semen for up to 4 days before clinical signs appear.
- The disease has been transmitted to calves via infected milk.
- Through animal handlers, visitors and physicians.
- Recovered animals can remain as a carrier following infection. Carrier may transfer the virus from one animal to another. Carrier cattle may harbor the virus in the esophageal-pharyngeal fluid for 6-24 months.

CLINICAL SIGNS

In Mithun, the clinical signs and symptoms are more or less the same as in cattle and other livestock, however, there are reports that shows higher morbidity and mortality in case of Mithun reared in free range system. The severity of clinical signs will depend on the strain of virus, the exposure dose, the age and species of animal and the host immunity. Morbidity can reach 100% in susceptible populations. Mortality is generally low in adult animals (1–5%), but higher in young calves, lambs and piglets (20% or higher). The incubation period is 2–14 days.

Clinical signs can range from mild or in-apparent to severe: they are more severe in cattle and intensively reared pigs than in sheep and goats. Common signs observed in acute infection are

- ❖ High fever up to 104-106°F (41°C) and anorexia.
- ❖ Profuse salivation (saliva hanging in long ropy strings up to the ground).
- ❖ Vesicles in feet and wounds in the interdigital space of legs followed by lameness.
- ❖ Oral ulcers and lesions.
- ❖ Smacking of lips.
- ❖ Vesicles in the mammary gland

As the disease advances, rupturing of blisters can result in extreme lameness and reluctance to move or eat. Usually, blisters heal within 7 days (sometimes longer), but complications, such as secondary bacterial infection of open blisters can occur if proper care is not taken. In young animals, death can occur before development of blisters due to a multifocal myocarditis. Recovered animals may have complications like weight loss, growth retardation and a drop in milk production.

FMD control programme in India: India is enzootic for foot-and-mouth disease (FMD) and economic losses due to the disease are more than ₹200 billion/annum. Currently, India is in stage three of the progressive control pathway for control of FMD and aims to achieve FMD-free status with vaccination by 2030. The success of the FMD control program not only depends on the biannual vaccination of 500 million susceptible livestock such as cattle, sheep, goat, pig, buffalo, yak, mithun, and other wildlife, but also depends on effective sero-surveillance.

The flagship FMD control programme in India started in the year 2003 covering 54 districts. The programme then gradually expanded and further, NADCP was launch in the year 2019.

Currently, there are 26 collaborating centers, 6 Regional Centers and ICAR-NIFMD laboratories under the FMD control programme engaging in seromonitoring and surveillance of FMD in the country. The programme covers many other important activities for effective control of the disease including disease diagnosis, development of diagnostic kits, vaccine matching, cataloguing of virus strains, vaccine quality control, disease epidemiology etc.

Due to the non-stop and active vaccination campaign coupled with serosurveillance/seromonitoring under the programme, the disease occurrence and severity in India has been gradually reduced.

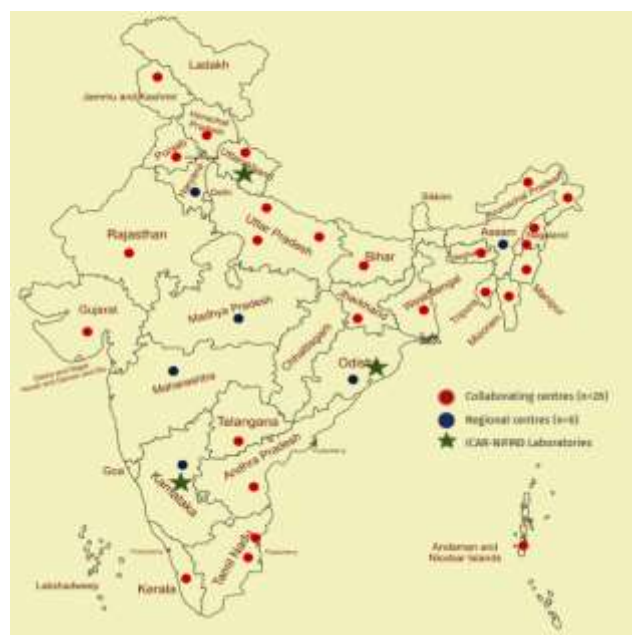


FIG: Map showing FMD control network (DFMD annual report 2022)



FIG: Activities under FMD Control programme.

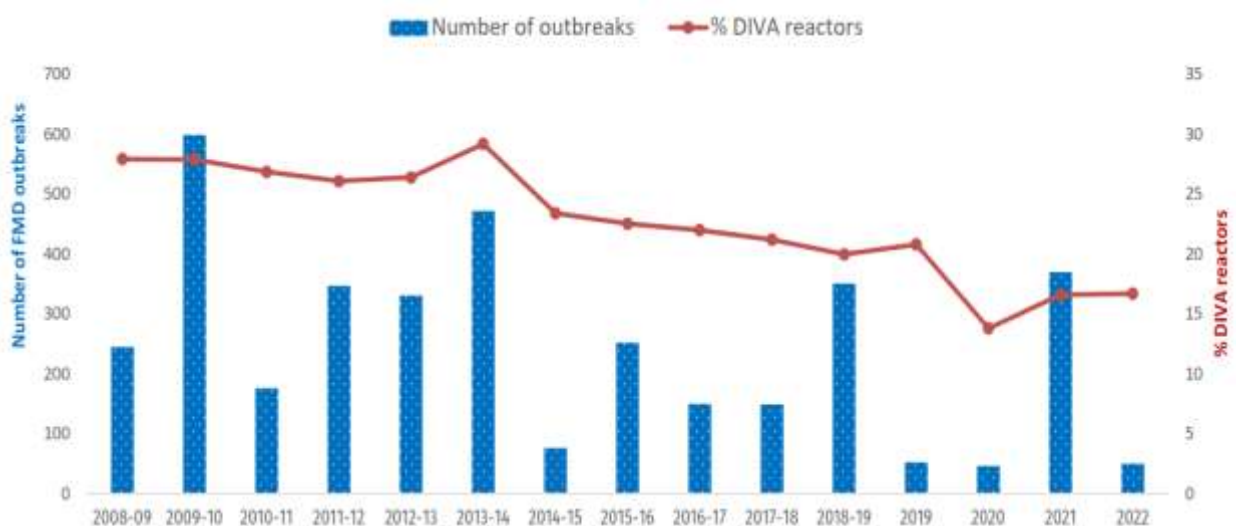


FIG: Trend of FMD outbreak and DIVA reactivity (DFMD Annual report 2022)

PREVENTIVE MEASURES

In FMD endemic countries like India, vaccination is the foremost action to prevent the occurrence of FMD. Bi-annual vaccination with inactivated trivalent vaccine is being followed. Routine vaccination of animals with proper dose is important to maintain herd immunity. Young animals should be vaccinated at 3 months followed by booster dose at 30 days after first vaccination. Then it should be repeated every 6 months interval. Following points must be followed for effective control of FMD-

- Vaccination of the entire animal in farm/village.
- Routine monitoring of health and veterinary care.
- In case of FMD outbreak, ring vaccination may be followed to prevent further spread of the disease in and around the area.
- Healthy and preferably routinely vaccinated animals should be brought into the village from outside sources that too only 15-21 days following vaccination.
- Avoid purchase of animals from disease prevailing areas.
- New animals should not be purchased until six months following outbreak.
- Strict quarantine measures for newly purchased animals.
- A foot bath or truck bath may be made at the entrance of the village/farm.
- Avoid community grazing during outbreak.



Fig: FMD Vaccine and vaccination of Mithun

CONTROL MEASURES

In case of outbreak of FMD, care must be taken to control and prevent the virus circulation to the healthy herds. Implementation of good sanitary measures and hygienic practices are important to check the dissemination of the virus to the surroundings and nearby places.

Following measures are to be considered to prevent the spread of disease-

- Proper biosecurity measures should be in place at farm level-
 - Control of people's movement in and around livestock and equipment
 - Controlled introduction of new animals into existing herds
 - Proper cleaning and disinfection of livestock pens, buildings, vehicles and equipment
 - Appropriate disposal of manure and dead carcasses
 - Foot bath should be made at the entrance of the farm.

- Isolation and confinement of affected animals immediately after detection of clinical symptoms and restriction of animal movements.
 - Infected animals should not be allowed to graze in common grazing pasture.
 - Affected animals should not be allowed to drink water from ponds/streams/ rivers etc.
 - Diseased animals should not be allowed to roam about with other animals of the village.
- Movement of animal handlers and attendants who attended diseased animals should be restricted to the other animal population / farms. If it is not practicable, people should scrub themselves and their belongings with soap and caustic soda.
- In case of outbreaks, healthy animals should be attended first and then the affected ones. After attending the sick animals, persons should wash himself and his clothes with 4% sodium carbonate solution. Utensils used for collecting milk should be cleaned with 4% sodium carbonate solution.
- Calves should not be allowed to suckle affected mothers and they should not be fed with milk from affected animals.
- Mouth of the affected animals may be washed with antiseptic mouth wash. Potassium permanganate (KMnO₄) solution may be applied 3-4 times a day.
- Feet of the affected animals may be washed with 2% copper sulphate solution. Antiseptic lotion and fly repellents are to be used to avoid infection and maggot formation on the wound.
- Proper disinfection of the farm premises.
- Surveillance and monitoring in compliance with the State/National norms.
- Reporting of any suspected disease to the local authority/veterinary department.

Challenges: Although the reduction of FMD outbreaks and virus circulation due to the stringent FMD control in India is evident in the last two decades, the goal of being a FMD free status is still at the horizon. Many factors are involved for this matter which may include the vast geographical area of India, in which many parts of the area are under developed and lack proper communication as well. The huge number of livestock in the country and stray animals could also be a contributing factor in recurrent virus circulation. The occurrence of multiple serotypes and variants of the virus and a wide host range make controlling a huge task in order to stop the virus circulation. Lack of awareness by the general public on the importance of

vaccination and control of disease is also an important factor. Besides, the following points may directly or indirectly contributed to the problems-

- Emergence and re-emergence of genetic/antigenic variants of the FMD virus continuously occurring in the nature
- Porous international borders
- Lack of zoo-sanitary and bio-security measures at outbreak sites
- Climate change and animal movement
- Under reporting and poor emergency response to the disease
- Inadequate quantity and quality vaccine for FMD control in the country
- Unrestricted animal movement and presence of stray livestock in several parts of the country

CONCLUSION:

FMD is one of the most important livestock diseases that cause a huge economic lost to the country. Although a strong network of control programme is in place, further strengthening of the surveillance system and more vaccine coverage is required to effectively control the virus circulation and the outbreaks. Better and more efficient vaccine, robust monitoring and reporting system will enhance the control strategy. Awareness and active participation of all the stakeholders is required to achieve the goal of FMD free country.

Chapter 3

Bacterial diseases of domestic animals of the northeastern hilly region of India with special reference to Mithun (*Bos frontalis*)

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Mithun (*Bos frontalis*), a semi-domesticated bovine species native to the hill regions of Northeast India and neighboring countries, is geographically restricted to a small, hilly region, making various aspects of its health and diseases less understood. Given the anatomical, physiological, and behavioral similarities between Mithun and cattle and buffalo, it is presumed that Mithun may be similarly susceptible to common diseases affecting these other bovids.

Over the past two decades, periodic studies at institutes and field surveys have shown that Mithun, like cattle and buffalo, are vulnerable to a wide range of diseases, including viral, bacterial, fungal, and parasitic infections. Many diseases have been observed in clinical settings, while others have been identified through seroprevalence studies conducted at institute farms and through field surveys in Mithun-inhabited states.

Due to the ever-evolving changes in the climate and rise in the global warming, there is an increase in the number of incidences of the common diseases occurring in the animals of the region. An increase in the number of vectors such as ticks, mosquitoes, flies, etc. in this part of the country has resulted in the changing disease patterns in the livestock population of this region. Further, the porous borders of the north-eastern states with the neighbouring countries such as Tibet, China, Myanmar, Bangladesh, Nepal, and Bhutan bolsters the incursion of transboundary diseases into the country via unauthorized livestock transport, meat, and meat-derived products, etc. Despite its socio-cultural importance, there is a significant gap in the understanding and management of infectious diseases affecting mithun, which poses a serious threat to its health, productivity, and conservation.

COMMON BACTERIAL DISEASES OF MITHUN

The infectious diseases of mithun can be categorized into bacterial, viral, and parasitic infections. Among bacterial infections, bovine tuberculosis (*Mycobacterium bovis*), brucellosis (*Brucella abortus*), leptospirosis (*Leptospira* serovars), and hemorrhagic septicemia (*Pasteurella multocida*) are prevalent. These diseases not only affect the health of mithun but also pose zoonotic risks to humans. Clinically, these bacterial infections manifest as chronic

wasting, reproductive failures, and systemic illnesses. Diagnosis relies on a combination of clinical signs, culture techniques, serological tests, and molecular methods such as PCR. Control measures primarily focus on vaccination, biosecurity, and movement control, although challenges in vaccine coverage and disease surveillance persist.

Following is a brief on the common bacterial diseases recorded in Mithun viz., Tuberculosis, Paratuberculosis or Johne's Disease, Brucellosis, Black Quarter, Colibacillosis, Bronchopneumonia, and Naval ill.

Tuberculosis: Tuberculosis is a chronic and highly infectious disease and is caused by *Mycobacterium tuberculosis* and is characterized by the formation of tubercles in various organs. All mammals including Mithuns are susceptible to the disease.

Tuberculosis is one of the most common diseases in organized farms and is associated with the health condition of the workers in the farm premises as it is of zoonotic importance. Transmission of tuberculosis occurs through the aerosol route and ingestion of feed, water, and fomites contaminated with the excretions and secretions of the clinically affected animals. The animals remain undetected in the herd except for severe coughing, exercise intolerance, emaciation, cachexia, and death in the terminal stage of the disease.

Tuberculosis has been recorded in Mithuns by single intradermal (SID), and Stormont tests. The affected animals were associated with enlargement of lymph nodes, severe loss in body condition, exercise intolerance, and deep coughing. The lesions associated were tuberculous granuloma in bronchial and mediastinal lymph nodes, lungs, liver, and spleen. The nodules were of size ranging from pea to bean and were occasionally calcified and fibrosed.

Johne's Disease: Johne's disease is caused by *Mycobacterium paratuberculosis* and is one of the most common diseases of the ruminants in organized farms and is highly important from an economic point of view. It occurs in almost all species of animals including Mithuns. It is characterized by lympho-granulomatous enteritis affecting the mucosa of the ileocaecal valve, ileum, jejunum, and colon. It's a chronic condition associated with scouring, lymphadenitis, and micro-abscessation in the gut-associated lymph nodes. The clinical signs are scouring, malabsorption, chronic diarrhea, and emaciation leading to wasting and death. The disease has zoonotic importance and its occurrence in animals is closely associated with the incidences of Crohn's disease in human beings. The transmission occurs by oro-fecal route and the feces of affected animals are the most common source of contamination of feed, water, milk, etc.

A study on Mithuns at the institute farm were tested for JD by tests like rectal pinch smear, Johnin test, and Indirect ELISA and were found to be equally susceptible to that of cattle.

Brucellosis: Brucellosis caused by *Brucella abortus*, is an important disease of cattle, buffalo, sheep goats, dogs, horses, and Mithuns. It affects animals all over the world. It is caused by gram-negative *Brucella* spp. and is characterized by frequent abortion, stillbirths, retained placenta, and occasionally orchitis. It is highly infectious and transmission occurs through ingestion, inhalation, breeding, entry through conjunctiva, and cut wounds. The organism has a special predilection for the pregnant uterus, mammary gland, testis, lymph nodes, joint capsules, and bursa. In the cases of abortion, necrotizing placentitis with thickened placenta and purulent exudates are seen. The diagnosis is confirmed by staining and microscopic examination of fetal discharge, preputial washings, blood films, placenta, etc. Serological tests Standard Tube Agglutination Test (STAT), Rose Bengal Plate Test (RBPT), and ELISA-based test are most commonly used for screening of the herd.

A serological survey of Mithun using RBPT, STAT, and Avidin-Biotin ELISA has revealed that Mithun too suffers from brucellosis. The prevalence of brucellosis was recorded as 33% in the Mithun herd. *Brucella abortus* was identified from cases of abortion, stillbirths, and retained placenta in Mithun.

Black Quarter: Black quarter or black leg is acute to per acute disease condition in domestic and wild animals and is caused by spore-forming anaerobic *Clostridium* spp. the organisms are commonly found in the soil as well as the gastrointestinal tract of animals. The disease is characterized by inflammation of the muscles followed by discoloration, edema, cracking, and drying of the skin. Blackleg disease has been reported in free-ranging Mithuns of Arunachal Pradesh.

Colibacillosis: Colibacillosis in neonatal calves of Mithun is caused by *E. coli* and is characterized by neonatal diarrhea with or without hemorrhage, pneumonia, and in severe cases arthritis and fibrinous pericarditis. In severe cases, there is endotoxic shock followed by death. The organism is ubiquitous and is a normal inhabitant of the gastrointestinal tract and infection occurs through the fecal-oral route. Poor hygiene, lack of colostrum, and overcrowding are the predisposing factors associated with colibacillosis in calves.

Various strains of enterotoxigenic and non-enterotoxigenic strains of *E. coli* have been isolated and identified from cases of neonatal diarrhea, white scours, and arthritis in Mithun calves. *E. coli* serotype 025 has been isolated and identified from Mithun calves with arthritis and fibrinous pericarditis.

Bronchopneumonia: Bronchopneumonia is one of the most common conditions causing mortality in young animals. It is a multi-etiological disorder comprising various bacteria, viruses, and parasites as the underlying cause. It is characterized by tracheobronchitis and

gradual consolidation of the lungs and is associated with loss of appetite, depression, cough, asphyxia, dullness, and high fever. An outbreak of bronchopneumonia in Mithuns was associated with tracheobronchitis, consolidation of lungs, respiratory distress, dyspnoea, and death. *Staphylococci* spp. and *Pseudomonas* spp. were been found to be associated with the cases of bronchopneumonia in Mithun. Diagnosis is based on the clinical signs, isolation, and identification of the organism from the clinical cases.

Anthrax: Anthrax is caused by *Bacillus anthracis*, a sporulating aerobic bacterium leading to acute or peracute disease in multiple species. The infection occurs through ingestion of spore-contaminated soil, water, and infected carcass/bones. The clinical signs observed are dullness, grinding of teeth, stiff-legged gate, and bloody discharge from nostrils, anus, vulva etc. In anthrax-suspected cases, the carcass should not be opened. In an accidentally opened anthrax-infected carcass, the spleen may be found enlarged several times and the body cavities are filled with copious amounts of serosanguinous fluid and the heart and major blood vessels are filled with unclotted tarry-colored blood.

People of the Monpa tribe of the west Kemang district of Arunachal Pradesh have reported the incidence of anthrax-like diseases in Mithun based on signs and symptoms.

CONCLUSION

Overall, the health management of mithun is constrained by limited veterinary infrastructure, lack of targeted research, and traditional husbandry practices. Enhancing disease surveillance, promoting community awareness, and improving access to veterinary care are essential steps toward mitigating the impact of infectious diseases in mithun. Furthermore, collaborative research efforts are needed to develop effective vaccines, diagnostic tools, and therapeutic interventions tailored to the unique biology and ecology of mithun. This comprehensive understanding of infectious diseases in mithun will contribute to their sustainable management, ensuring the well-being of both the animals and the tribal communities dependent on them.

Chapter 4

Metabolic diseases of domestic animals of the northeastern hilly region of India with special reference to Mithun (*Bos frontalis*)

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The common metabolic disorders encountered in cattle are mostly not encountered in Mithuns. The chapter outlines several key metabolic disorders in cattle, each with distinct causes, symptoms, and prevention/treatment methods.

However, the knowledge of these disorders with respect to cattle is important under field conditions for field veterinarians:

1. Ketosis

Cause: Energy deficiency due to inadequate carbohydrate intake.

Symptoms: Reduced milk yield, loss of appetite, weight loss, lethargy, and sometimes neurological signs like staggering.

Prevention/Treatment: Ensure a balanced diet with adequate energy, administer propylene glycol, and monitor blood ketone levels.

2. Milk Fever (Hypocalcemia)

Cause: Low calcium levels in the blood, often occurring around calving.

Symptoms: Muscle tremors, weakness, inability to stand, and in severe cases, coma.

Prevention/Treatment: Provide oral or intravenous calcium supplements, and manage dietary calcium and phosphorus levels before calving.

3. Grass Tetany (Hypomagnesemia)

Cause: Low magnesium levels, typically in cattle grazing on lush pastures.

Symptoms: Nervousness, muscle twitching, staggering, and convulsions.

Prevention/Treatment: Supplement with magnesium, provide mineral licks, and avoid sudden changes in diet.

4. Displaced Abomasum

Cause: The abomasum (fourth stomach compartment) shifts from its normal position, often due to gas buildup or dietary changes.

Symptoms: Reduced appetite, decreased milk production, and discomfort.

Prevention/Treatment: Ensure a balanced diet with adequate fiber, and surgical intervention may be required in severe cases.

5. Fatty Liver Disease

Cause: Excessive fat mobilization, usually in over-conditioned cows during the transition period.

Symptoms: Reduced feed intake, decreased milk production, and increased susceptibility to other diseases.

Prevention/Treatment: Manage body condition score, provide balanced nutrition, and avoid excessive weight loss around calving.

6. Acidosis

Cause: Rapid fermentation of carbohydrates in the rumen, leading to a drop in pH.

Symptoms: Reduced feed intake, diarrhea, bloat, and in severe cases, death.

Prevention/Treatment: Gradually introduce high-energy diets, provide adequate fiber, and use buffers like sodium bicarbonate.

7. Laminitis (Founder)

Cause: Inflammation of the laminae in the hoof, often linked to acidosis or other metabolic disturbances.

Symptoms: Lameness, reluctance to move, and swollen hooves.

Prevention/Treatment: Manage diet to avoid acidosis, provide proper hoof care, and use anti-inflammatory medications.

8. Nutritional Myopathy (White Muscle Disease)

Cause: Deficiency of selenium and/or vitamin E.

Symptoms: Muscle weakness, stiffness, and difficulty standing.

Prevention/Treatment: Supplement diets with selenium and vitamin E, and provide injectable supplements if necessary.

9. Rumen Alkalosis

Cause: High intake of protein-rich feeds leading to increased ammonia production.

Symptoms: Reduced feed intake, lethargy, and decreased rumen motility.

Prevention/Treatment: Balance dietary protein intake and provide adequate roughage.

10. Hypokalemia

Cause: Low potassium levels, often due to prolonged anorexia or high grain diets.

Symptoms: Muscle weakness, recumbency, and abnormal heart rhythms.

Prevention/Treatment: Supplement with potassium chloride, and ensure a balanced diet.

Management Strategies for Metabolic Diseases

Balanced Nutrition: It should be ensured that cattle receive a balanced diet with appropriate levels of energy, protein, vitamins, and minerals.

Monitoring: Regularly monitoring of body condition scores, blood parameters, and milk production to detect early signs of metabolic diseases.

Transition Period Management: Careful management of nutrition and health during the transition period around calving to minimize metabolic stress.

Education: Training of farm staff to recognize symptoms of metabolic diseases and implement preventative measures.

Veterinary Care: Establishment of a good relationship with a veterinarian for regular health checks and emergency interventions. By implementing these strategies and maintaining a proactive approach to herd health management, farmers can minimize the incidence and impact of metabolic diseases in cattle. Routine incorporation of vitamin A supplement along with other minerals in the ration of Mithun is suggested for prevention of hypovitaminosis-A besides inclusion of sufficient green fodder in the normal ration of Mithun. Use of hematinic mixtures are suggested as supportive therapy for anemia besides correction of primary cause of anemia. Such nutritional supplementation along with the regular diet of fast-growing animals can help in the prevention of metabolic diseases.

CONCLUSION

In summary, metabolic disorders commonly observed in cattle, though rare in Mithuns, present critical challenges that require thorough understanding and management strategies. Effective management strategies involve ensuring balanced nutrition, regular monitoring of health parameters, careful transition period management, education of farm staff, and maintaining veterinary care. These proactive measures can significantly reduce the incidence and impact of metabolic diseases in livestock.

Chapter 5

Parasitic diseases of mithun and future strategies for control measures

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INTRODUCTION

Mithun (*Bos frontalis*), commonly referred to as ‘the cattle of the hills,’ is believed to have descended from the Gaur (*Bos gaurus*). This unique bovine species primarily inhabits the northeastern region of India and certain other parts of Southeast Asia. Mithun holds significant cultural, social, and economic importance for the tribal communities of Northeast India. Its versatile utility has been widely acknowledged. Mithun meat ranks among the preferred choices of meat for the hill-dwelling people of northeastern India. Notably, our institute has recently undertaken a project to harness the draught capabilities of this animal. Research findings thus far have indicated that the sure-footed nature of mithun makes it exceptionally suitable for use as a draught and pack animal, particularly on the steep slopes of hills. Mithun also holds cultural significance, often being exchanged as a bridal gift or in trade within tribal communities. In this region’s society, owning mithun signifies prosperity and social standing.

Key environmental parameters for Northeast India include an average relative humidity of 80%, soil pH ranging from 6.5 to 8.0, and temperatures between 15°C and 37°C. These conditions foster the growth and propagation of parasitic life cycles and the transmission of microorganisms, including bacterial and viral agents. However, a comprehensive method for field diagnosis of subclinical disease infections in mithun has not yet been established, and the causative agents of these diseases remain insufficiently documented.

The epidemiology of parasitic diseases is influenced by a multitude of factors shaped by environmental conditions and interactions between hosts and parasites. The northeastern region serves as a hotspot for parasites due to its climate, which provides an ideal environment for the proliferation of parasitic populations. To ensure sustained economic development, veterinarians bear a crucial responsibility in managing parasitic burdens, recognizing the vital role livestock plays in the lives of the region’s indigenous communities. Therefore, it becomes imperative to implement robust strategies for controlling worm burdens in mithun, a step that

can contribute to the socioeconomic advancement of this area. This article systematically examines existing epidemiological data related to helminth infections in mithuns. The present article sheds light on the most frequently encountered diseases in mithun within the northeastern region of India, which lead to substantial economic losses. The insights gained from this review can form the foundation for the implementation of targeted control measures aimed at averting parasitic infections in mithuns within the context of northeastern India.

NEMATODIASIS

Parasitic gastroenteritis represents a significant concern within the mithun population. Substantial evidence indicates a direct or indirect correlation between parasitic gastroenteritis and both mortality and morbidity in mithun. Various parasite genera have been identified as culprits behind this condition, including *Haemonchus contortus*, *Oesophagostomum*, *Trichostrongylus* sp., *Mecistocirrus*, *Cooperia*, and *Trichuris* sp. Among these, *Toxocara vitulorum*, *Strongyloides papillosus*, *Bunostomum phlebotomum*, and *Haemonchus contortus* have emerged as the primary triggers of anemia and mortality in mithun calves.

The development, survival, and transmission of parasitic eggs and infective larvae are profoundly influenced by climatic and environmental factors, encompassing temperature, humidity, and precipitation. These elements frequently give rise to seasonal variations in infective larvae availability, consequently impacting the prevalence of infections and worm burdens within the host. These epidemiological dynamics may remain consistent for mithun under controlled farm conditions, but their severity might be diminished in mithun living in free-range conditions. Grazing animals, by nature, tend to harbor a varying yet substantial worm population, with parasite development notably active during the rainy season. In regions characterized by distinct rainy and dry seasons, a significant proportion of third-stage larvae (L3) undergoes arrested development towards the close of the rainy season. Subsequently, fecal egg counts diminish, maintaining a consistently low level during the dry season. As the rainy season commences, pastures experience a surge in larval challenge, resulting in heightened egg output – a pattern commonly observed.

However, areas surrounding water sources like drinking spots and permanent ponds might maintain the required humidity levels to support optimal larval development and year-round transmission. In terms of transmission dynamics, it's anticipated that higher stocking rates could lead to elevated rates of parasitic infection within animal herds. Certain nematode genera larvae exhibit the ability to postpone maturation into adult stages, a phenomenon termed

hypobiosis. Typically, resumption of parasitic development coincides with the advent of the rainy season, which provides the most conducive environment for larval development and transmission. This resumption can be attributed to acquired immunity of parasites or prior exposure to adverse climatic conditions during their free-living larval stages. While arrested parasite development has been linked to declining temperatures in temperate zones, the triggering stimulus in tropical regions differs significantly. In the waning phase of the rainy season, arrested larvae accumulate within the abomasal and intestinal mucosa of the host, maintaining this state throughout the dry season and subsequently resuming and completing development at the onset of the subsequent rainy season. It's noteworthy that mithun's preference for browsing on shrub and tree leaves contributes to a comparatively lower degree of parasitic infection within the alimentary tract.

The phenomenon known as periparturient rise in faecal egg count, widely observed in other ruminants, significantly shapes the epidemiology of gastrointestinal nematodes in ruminants. This occurrence primarily stems from prolactin hormone levels, inducing a temporary relaxation of immunity in pregnant animals.

MICROFILARIA

Microfilaria is a commonly observed condition in mithun and is primarily transmitted by the *Culicoides* mosquito. It is not influenced by the gender of the host animals. Its prevalence has been recorded during the monsoon season, and it tends to be more prevalent in older animals. However, it does not produce any clinical symptoms. Instances of this condition have been documented in both Arunachal and Nagaland through post-mortem examinations. Through gross morphology and PCR amplification of marker genes, it has been identified as *Setaria digitata*.

S. digitata is characterized as a lengthy, milky-white worm with tapering and spirally-coiled hind ends. It is most commonly found within the peritoneal cavity of mithun in the regions of Arunachal Pradesh and Nagaland. The larvae of this filarial nematode are referred to as microfilaria, and they reside in the blood of the host. The life cycle of this parasite is indirect, involving the culicine mosquito as an intermediate host. Although the adult worms are generally considered non-pathogenic, they might be accountable for causing mild fibrinous peritonitis. On the other hand, the larval forms are deemed to be pathogenic.

TREMATODES

The infestation of flukes stands as a prominent contributor to clinical manifestations in ruminants. Among these flukes, *Fasciola 37iarrhea* and amphistomiasis have emerged as pivotal factors driving mortality within ruminant populations. However, the occurrence of fluke infestations in mithun is notably limited. In mithun, infestations with adult amphistomum species such as *Paramphistomumepiclitum*, *Gastrothylax crumenifer*, *Cotylophoroncotylophorum*, and *Fischoederius 37iarrhea*³⁷ have been observed to induce subclinical manifestations. Additionally, instances of mortality in mithun calves have been reported due to heavy infestations with immature amphistomiasis.

The causal agent behind fasciolosis in mithun has been identified as *Fasciola 37iarrhea*. The geographical distribution of trematode species affecting ruminants hinges on the presence of suitable aquatic Lymnaid snails, which serve as intermediate hosts for the parasites. However, the establishment and propagation of fluke infections are contingent not only on the presence of snails but also on favorable climatic and ecological conditions conducive to both the parasite and its intermediate host. In the majority of tropical developing countries, temperatures generally favor the development of both flukes and their intermediate hosts. Yet, variations in precipitation and humidity lead to fluctuations in snail development and the free-living stages of parasites. The rainy season prompts a significant increase in snail reproduction, culminating in a peak snail population by the season's end. Conversely, this trend slows or halts during dry or cold periods, resulting in diminished snail populations during the dry season. This, in turn, results in marked fluctuations in herbage infestation and the survival of metacercariae.

In humid tropical environments, the infective stage can endure for up to 10 months, while the longevity of metacercariae ranges from a few weeks to 3-4 months in relatively hot and dry climates. The ultimate determinant of parasitic disease epidemiology lies in the rate of egg production by adult flukes, which subsequently influences the extent of pasture contamination. Furthermore, the grazing behaviors and management practices of animals can significantly impact the epidemiology of liver fluke infections.

Cestode

Cestode infestation poses a significant and prevalent challenge for mithun, particularly in the northeastern part of the region. Among the culprits are the species *Moniezia expansa* and *M. benedeni*, which not only disrupt the digestive processes but also substantially diminish the animals' overall productivity. The prevalence of tapeworms is profoundly influenced by a

complex interplay of factors, encompassing both the epidemiology of the parasites and various environmental elements.

The occurrence of these tapeworms is intricately tied to the presence of suitable intermediate hosts for the cestodes, as well as favorable climatic conditions. This intricate interrelationship between these factors becomes a determining force shaping the epidemiology and prevalence of these parasites within the animal population. The infective stage of these tapeworms, known as cysticercoids, resides within oribatid mites. It is this stage that initiates the infection following ingestion by the animals alongside their regular grass diet.

Moreover, the incidence of hydatidosis also affects significantly within the mithun population, attributable to the sharing of both sylvatic and non-sylvatic life cycles. This underscores the complexity of the transmission dynamics and the susceptibility of mithun to these parasitic infections. In light of these factors, understanding the relationships between epidemiological patterns, intermediate hosts, climatic conditions, and host behavior becomes paramount in developing effective strategies to mitigate the impact of cestode infestations and related health issues within the mithun population.

PROTOZOAL INFECTION

The most prevalent protozoal species reported in mithuns from northeast India are *Eimeria bovis*, *E. zuernii*, *E. ovoidalis*, *E. bukidonensis*, *E. auburnensis*, *E. ellipsoidalis*, *E. subspherica*, and *E. albamensis*. The prevalence of Eimerian species is generally highest in the pre-monsoon season compared to other seasons. Infestation with *Eimeria* species has been observed across all age groups of mithun. Notably, the pathogenicity has consistently been noted as higher in young mithun.

The severity of eimeriasis in ruminants may exhibit correlations with various managerial factors. Incidence rates tend to be higher in animal herds reared under semi-intensive systems characterized by unhygienic conditions. Conversely, the incidence of eimeriasis is notably lower in animals raised within a free-range system. The utilization of sulfadimidine and amprolium has demonstrated moderate effectiveness in treating Coccidiosis in mithun calves. Additionally, a few other tissue protozoa, such as *Balantidium coli* and *Cryptosporidium bovis*, have also been frequently observed in mithun calves.

Cryptosporidiosis, a tissue protozoan infection, predominantly impacts younger mithuns. It has been identified as a significant contributor to calf 39diarrhea, with the highest incidence recorded in mithun calves aged less than 1-2 months.

Toxoplasmosis is a tissue protozoan that affects a wide range of warm-blooded animals. However, clinical cases of toxoplasmosis have not yet been reported in mithun. Nevertheless, reports of serological prevalence have emerged from mithun populations in Nagaland. The prevalence of *Toxoplasma gondii* infection in mithun has been observed to increase with the advancing age of the animals. Highest prevalence is noted in adult mithun, while the lowest rates occur in individuals aged 6 months to 1 year.

Mithuns residing at lower altitudes exhibit greater vulnerability to *T. gondii* infection compared to those inhabiting higher altitudes. Native wildlife such as wild foxes and jungle cats significantly contribute to the epidemiology and transmission of toxoplasmosis, acting as intermediaries that sustain the sylvatic transmission cycle of the protozoan. Given its zoonotic significance as a tissue protozoan of animal origin, comprehensive studies encompassing various aspects of toxoplasmosis in animals are imperative.

Sporadic occurrences of the blood protozoan disease babesiosis in mithuns have also been documented. Additionally, clinical infestations of *Trypanosoma theileri* have been diagnosed through blood smear examinations in mithun. Clinical manifestations of trypanosomiasis in mithuns include intermittent fever, dullness, emaciation, anemia, mucopurulent eye discharge, increased respiration, and elevated pulse rates. Furthermore, biochemical analyses have indicated lower hematocrit values in mithuns infected with *Trypanosoma theileri*.

ECTOPARASITE INFESTATION

Ectoparasitic infestations are a prevalent concern among mithun. Infestations involving leeches and ticks, such as *Boophilus microplus*, are particularly commonplace in mithun populations. It's noteworthy that *Rhipicephalus microplus*, an ixodid tick species like *Ixodes ovis* and *I. acutitarsus*, has been identified as the sole tick found in mithuns residing in Nagaland. Nevertheless, instances of unidentified ticks have also been documented in mithuns from Mizoram, yet further confirmation is required for accurate species identification. A comprehensive study encompassing 176 mithuns highlighted that 58.52% of the examined population were afflicted by *R. microplus* infestations. Moreover, the intensity of these infestations tends to be more pronounced in older animals. Through seasonal analysis, it was

noted that the prevalence of *R. microplus* was most pronounced during the summer months. This correlation echoes findings from cattle studies in Assam, a neighboring state of Nagaland, wherein none of the observed cattle were exempt from *R. microplus* infestations. Furthermore, reports have emerged regarding infestations involving *Haemaphysalis daivasi* and *H. neumani* ticks in mithun populations. The variation in tick distribution patterns can be attributed to a lack of well-defined determinants governing the tick population's dispersion. However, among the factors, rainfall has been indicated as a pivotal limiting factor influencing the distribution of ticks. Lice like *Linognathus vituli* was also recorded from mithun.

A frequently encountered issue in mithun rearing is the infestation of leeches within both the body and nasal cavity. This problem, often perceived as a significant annoyance by the farmers of this region, is widespread among cattle and mithuns raised under grazing management practices. These leeches affix themselves to various parts of the animal's anatomy, including the body surface, nasal cavity, and even reproductive organs. Leech infestations, while occasional, present a considerable challenge due to their bloodsucking behavior. These parasites secrete anticoagulant substances that induce continuous bleeding, persisting even after the leech has detached from the animal's body. Consequently, the volume of blood loss stemming from leech infestations can be substantial, especially considering the amount of blood consumed by a single leech during its feeding process. Despite the prevalence of leech infestations and their substantial impact on animal health, no comprehensive and systematic investigation has been conducted to date on aspects such as taxonomy, pathology, economic ramifications, and effective control measures for leech infestations across various animal species.

STRATEGY FOR PARASITIC CONTROL IN MITHUN:

ANTHELMINTICS

There is a wide range of commercial anthelmintic formulations available in the market, each marketed under different brand names. However, the number of effective anthelmintics for treating worm infestations is limited. The situation arises from the existence of different chemicals within a group that share a similar mode of action. It's essential to be aware of the potential side effects and the likelihood of resistance developing within anthelmintics that share similar modes of action. Consequently, to prevent the emergence of anthelmintic resistance, it's crucial to understand the specific mode of action of a particular anthelmintic before its use.

Several critical factors need to be considered before administering an anthelmintic. These include the health condition of the animal, the type of parasitic infestation, the appropriate dosage, and the preferred route of drug administration. The evaluation of the efficacy of various chemical anthelmintics in mithun is currently in progress. The use of Fenbendazole at a dosage of 10mg/kg body weight has proven effective against moneiziasis in mithun. Similarly, Piperazine is the preferred choice for combating *T. vitulorum* infections in mithun. Piperazine offers an advantage over Levamisole and Ivermectin due to its ability to expedite recovery in the treatment of Ascariasis.

The efficacy of Albendazole at a dosage of 15mg/kg body weight as a broad-spectrum anthelmintic for mithun is well-documented . Subcutaneous administration of Ivermectin at a rate of 1ml/50kg body weight has been found to be highly effective against both endo and ectoparasites. Furthermore, the regular practice of applying Cypermethrin as a spray to control *R. microplus* infestations is observed in the institute’s mithun farm. A recent study has revealed that Ivermectin is the most effective anthelmintic against *Strongyloidpapillosus* infections in mithun.

Table 1. Recommended anthelmintic for deworming program in mithun:

Sr. No.	Anthelmintic	Used against	Dose (mg or ml / kg body wt.)	Route
1	Tetramisole	Round worm	7.5mg	S/C ly
2	Levamisole	Round worm	7.5 mg	S/C ly
3	Pyrantel	Tapeworm	10mg	Orally
4	Morantel	Broad spectrum	10-20 mg	Orally
5	Benzimidazole group	Broad spectrum	7-15 mg	Orally
6	Niclosamide	Fluke infestation	90 mg	S/C ly
7	Oxyclozanide	Fasciola, Amphistomum	10mg	Orally
8	Triclabendazole	Fasciola	9 mg	Orally
9	Ivermectin	Broad spectrum	0.02	S/C ly
10	Doramectin	Broad spectrum	0.02	S/C ly

GRAZING MANAGEMENT BASED ON EPIDEMIOLOGICAL KNOWLEDGE

In worm control program, the role of grazing management based on epidemiological knowledge is to provide clean pasture on which animals may graze safely.

1. Alteration of host species: To the extent that two or more host species in any given environment don't share common parasite species, alteration between species can be

successful means of enhancing worm control. Grazing of mithun along with sheep may be another option in which not sharing of common species of parasites by the mithun as well as sheep.

2. Rotational grazing: This comprises the withdrawal of the susceptible host from the pasture/forest until the free living parasitic stages have died due to aging and environmental exposure. Then the animals are again introduced in the grazing area. It is impractical under intensive farming condition and in the temperate climate since the long survival time of the infective larvae on pasture that requires longer period of resting. Nevertheless, this may be practical method for parasitic control in tropical countries where extensive farming is practiced and grazing fields are abundant.
3. Clean pasture approach: Under this practice collection and removal of deposits from pasture is done by pasture sweeping or vacuum cleaning twice in a week. This is a costly but effective measure that is practiced in some animal farms in USA. However, similar type of practice is not feasible for Mithuns and other animals reared under Indian condition.
4. Forecasting: On the basis of data on environmental factors like number of rainy days, amount of rainfall, temperature and prevalence of helminth infection in a year, it is possible to estimate rate of parasitic infection in pasture and predict risk of future worm infection. Adoption of such forecasting system may assist farmers to implement timely and appropriate measures against specific parasitic infestation

CONCLUSION

It is imperative for both the owner and the veterinarian to collaboratively design a deworming program tailored to suit the specific needs of the herd. This program should be meticulously aligned with recommended management practices. The implementation of strict biosecurity measures is paramount across animals, animal products, vehicles, personnel, and equipment. Additionally, comprehensive disinfection protocols should be observed for all areas in contact with the animals and any infected materials. Several disinfectants, such as sodium hydroxide, sodium carbonate, citric acid, or other appropriate antibiotic solutions, can be employed to combat infectious diseases.

In addition to this, controlling the dissemination of parasites via rodents and other vectors is essential. Measures should be taken to prevent these vectors from mechanically spreading the parasites. In the decision-making process concerning preventative herd health, the guidance of

a veterinarian should always be sought. Moreover, strict adherence to product instructions and precautionary measures is crucial.

Creating a successful deworming strategy and enforcing it through strict management practices, proper disinfection protocols, and collaboration with a veterinarian contributes to the overall health and well-being of the herd.

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Chapter 6

Parasitic zoonosis of bovines in the northeastern hilly region of India

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INTRODUCTION

The north-eastern region of India is exceptionally rich in natural resources and boasts a diverse cultural tapestry among its tribal population. This area is characterized by its lush landscapes, abundant flora and fauna, and a variety of natural resources that support the local economy. The region's cultural diversity is significant, with numerous tribes, each having its own unique customs, traditions, and languages. Nearly 70% of the region's livelihood activities revolve around agriculture and livestock, which form the backbone of the local economy. Livestock rearing plays a crucial role in meeting economic demands and providing essential animal protein, which is vital for the nutritional needs of the population. Furthermore, approximately 90% of the population in this region consumes non-vegetarian diets, relying heavily on meat and meat products for their daily sustenance.

Due to their cultural and dietary habits, the people of this region are more susceptible to various zoonotic diseases. These diseases are transmitted from animals to humans, and the consumption of a wide variety of meat and meat products increases the risk of infection. Parasitic zoonoses, in particular, are among the most significant causes of reduced productivity in domestic livestock and pose a severe threat to public health. These diseases not only affect the health of the livestock but also have a direct impact on the livelihoods of the people who depend on them. Unfortunately, the economic losses incurred by livestock owners, as well as the morbidity and mortality of livestock due to parasitic infections, have not been adequately understood or addressed.

The distribution, prevalence, and severity of these diseases are influenced by complex interactions between the biology of parasites, hosts, and vectors, along with prevailing environmental changes such as climate variability, humidity, and temperature fluctuations.

Additionally, socio-cultural practices, including traditional methods of animal husbandry and local dietary customs, play a significant role in the epidemiology of these diseases. This aspect needs to be carefully considered when assessing or developing regional control programs for the north-eastern region of India. Effective control and prevention strategies must be tailored to the specific environmental and cultural context of the region to mitigate the impact of zoonotic diseases on public health and the local economy.

Commonly recorded Zoonosis available in north eastern hilly region of India are as follows:

1. Taeniasis & Cysticercosis:

Taeniasis is a significant zoonotic disease caused by two parasites, *Taenia saginata* and *Taenia solium*. It affects millions of people globally, particularly in regions where hygiene standards are not adequately maintained and beef consumption is common. In such areas, up to 75% of the population may harbor these parasites, which thrive in environments where beef is a staple part of the diet. The prevalence of taeniasis is notably high in African countries, the Middle East, and South Asian countries like Iraq and Iran. *T. solium*, in particular, is endemic in Latin America, Africa, Asia, certain parts of Europe, and the former Soviet Union.

In India, especially in the northeastern region, taeniasis remains a significant health concern. A study by Borkataki et al. (2012) reported that in Assam, the prevalence of Porcine Cysticercosis in three districts was higher in crossbred pigs (12.53%) compared to local breeds (7.49%). This highlights the endemic nature of *T. solium* in this region. Similarly, *T. saginata* is highly endemic in Africa, Eastern Mediterranean countries, and parts of the former Soviet Union (Gloria et al., 2013).

The transmission of taeniasis occurs through the ingestion of infective cysticerci found in undercooked pork (*T. solium*) or beef (*T. saginata*). Contaminated food, water, and vegetables carrying the eggs of these parasites also play a role in transmission. *T. saginata* often causes taeniasis, with symptoms including the discharge of gravid proglottids from the anus, irritation, anal itching, abdominal pain, nausea, reduced appetite, and headaches. The cysticerci of *T. saginata* are transmitted to humans through the consumption of raw or inadequately cooked beef. Additionally, infected farm workers and the practice of open defecation can contaminate fields, leading to further spread of the disease. Transmission can also occur mechanically through birds, earthworms, and beetles that have ingested raw sewage sludge.

Cysticercosis, a condition caused by the tissue infection following exposure to *T. solium* eggs, is another severe consequence of taeniasis. This infection is primarily acquired through the intake of contaminated and unhygienic food or water containing viable *T. solium* eggs. In pigs, cysts are typically found in striated muscles such as the heart, tongue, and skeletal muscles, including the forearm. Cysts may also be present in the liver, lungs, spleen, eyes, conjunctiva, and nervous system of pigs, where they remain viable for extended periods, leading to meat condemnation and significant economic losses for pig producers.

The most common route of infection is through the consumption of raw vegetables grown in fields irrigated with untreated sewage water. Additional potential sources of fecal-oral contamination include the admixture of sewage water with drinking water in pipelines and the mechanical transmission by houseflies and cockroaches. In humans, cysts can form in the brain and muscles, persisting for years. Symptoms include generalized muscle pain, painful nodules in the muscles, and seizures when cysts are located in the brain. Cysticerci cysts can develop in any voluntary muscle, causing myositis with symptoms such as fever, headache, and eosinophilia. These cysts may block the outflow of cerebrospinal fluid, leading to increased intracranial pressure and related symptoms. Neurocysticercosis can also involve the spinal cord, often presenting as back pain and radiculopathy.

Diagnosing taeniasis traditionally involves demonstrating tapeworm eggs or proglottids in stool samples. However, the diagnosis of neurocysticercosis is primarily clinical, based on a presentation of compatible symptoms and imaging studies. CT scans can reveal both calcified and uncalcified cysts, distinguishing between active and inactive cysts, while MRI is more sensitive in detecting intraventricular cysts.

Preventative measures for taeniasis and cysticercosis include mass chemotherapy of infected individuals using Niclosamide at 10 mg/kg body weight and Praziquantel at 10 mg/kg body weight. Improving sanitation, educating people about the risks, thoroughly cooking or freezing pork, inspecting meat, and treating or vaccinating pigs are also critical strategies. Implementing these measures can significantly reduce the prevalence and impact of these zoonotic diseases, thereby improving public health and reducing economic losses for livestock producers.

2. Toxoplasmosis

Toxoplasmosis is caused by the parasite *Toxoplasma gondii*. Humans can acquire the infection through several routes, including the ingestion of tissue cysts present in raw or undercooked meat such as beef, lamb, or pork, and the ingestion of oocysts from contaminated soil, water, milk, or vegetables. Toxoplasmosis is a ubiquitous disease, present in every country around the world, with seropositivity rates ranging from less than 10% to over 90%. Globally, over 6 billion people have been infected with *T. gondii*.

Human infection typically occurs through the ingestion of tissue cysts in raw or poorly cooked meat from lamb and pork, or through the ingestion of sporocysts derived from cat feces that contaminate soil or inadequately washed vegetables. Congenital infections are most commonly caused when a mother acquires the infection during pregnancy, passing the parasite to the fetus. The incidence of toxoplasmosis in both animals and humans depends on various factors, including environmental conditions, climate, animal fauna, cultural habits, and feeding practices, which can vary significantly across different geographical areas.

Toxoplasmosis can also be acquired through the ingestion of food contaminated with mature oocysts or by consuming undercooked or raw infected meat. Mechanical transmission is possible as well; cockroaches and flies can spread oocysts mechanically. Laboratory and autopsy accidents, as well as improper handling, can also result in infection. Additionally, blood transfusion is a possible, albeit rare, route of transmission.

In acute toxoplasmosis, clinical symptoms are generally mild and may include influenza-like symptoms such as muscle aches, pain, and swollen lymph nodes. However, the disease can develop into more severe forms in young children and immunocompromised individuals, potentially causing encephalitis or necrotizing retinochoroiditis. Infants infected via placental transmission may be born with severe complications, including nasal malformations. Swollen lymph nodes are commonly found in the neck or under the chin, followed by the axillae (armpits) and the groin. Enlarged lymph nodes may subside within one to two months in about 60% of cases, although a quarter of those affected may take two to four months to return to normal.

Most infants who are infected while in the womb show no symptoms at birth but may develop symptoms later in life. These symptoms can include skin lesions such as roseola, erythema

multiforme-like eruptions, prurigo-like nodules, urticaria, and maculopapular lesions. Newborns may present with punctate macules, ecchymoses, or “blueberry muffin” lesions, indicative of congenital infection.

In livestock, the prevalence of toxoplasmosis can vary. For instance, in a study conducted in the northeastern hilly region of India, out of 195 Mithun animals examined, only 8 (4.10%) were suspected to be infected, and just one (0.51%) was confirmed positive. This highlights the regional variability in the prevalence of the disease.

Toxoplasmosis is a significant public health concern due to its widespread distribution and the potential for severe health outcomes, especially in immunocompromised individuals and during congenital transmission. Preventative measures are crucial in managing the spread of this disease. These measures include ensuring that meat is cooked thoroughly to kill tissue cysts, practicing good hygiene to prevent contamination of food and water with oocysts, and educating the public about the risks of consuming raw or undercooked meat and the importance of washing vegetables and hands properly.

Additionally, controlling the cat population and their access to livestock areas can help reduce the environmental contamination with *T. gondii* oocysts. Implementing these strategies can help mitigate the impact of toxoplasmosis on public health and prevent the spread of this zoonotic parasite. Understanding the environmental, cultural, and socioeconomic factors that contribute to the transmission of toxoplasmosis is essential for developing effective control programs and reducing the burden of this disease globally.

DIAGNOSIS

Laboratory diagnosis of toxoplasmosis primarily involves the detection of Toxoplasma-specific antibodies, which is the standard method to determine an infection with *Toxoplasma gondii*. Various commercially available test kits are used to detect *T. gondii*-specific antibodies, including IgG, IgM, IgA, and IgE. Several serological tests are commonly employed for the diagnosis of toxoplasmosis. These include the Indirect Fluorescent Antibody Test (IFAT), Latex Agglutination Test (LAT), Direct Agglutination Test (DAT), Sabin-Feldman Dye Test, Complement Fixation Test, Modified Agglutination Test, and Enzyme-Linked Immunosorbent Assay (ELISA). Among these, ELISA is widely used due to its sensitivity and specificity. In the case of congenitally infected babies, the detection of Toxoplasma-specific IgA antibodies

has proven to be more sensitive than the detection of IgM antibodies (Joao et al., 2011). Additionally, molecular methods such as the polymerase chain reaction (PCR) are employed to detect *T. gondii* in human blood samples, cerebrospinal fluid (CSF), and amniotic fluid, providing a reliable diagnostic approach (Webster et al., 2010). Imaging methods and immunohistochemical studies also play a crucial role in the diagnosis of toxoplasmosis. Imaging techniques, such as computed tomography (CT) and magnetic resonance imaging (MRI), can help identify characteristic lesions associated with the infection. Furthermore, the biopsy of enlarged lymph nodes, followed by histopathological examination and culture, offers a confirmative diagnosis. For rapid diagnosis, microscopic examination of impression smears stained with the Giemsa technique can be utilized to identify *T. gondii*. This method allows for the quick visualization of the parasite and is useful in clinical settings where immediate diagnosis is necessary.

In summary, the diagnosis of toxoplasmosis involves a combination of serological, molecular, imaging, and histopathological techniques. The choice of diagnostic method depends on the clinical context, the patient's immune status, and the need for rapid and accurate detection of the infection. Comprehensive diagnostic approaches ensure effective detection and management of toxoplasmosis, aiding in timely intervention and treatment.

3. Hydatid cyst

Hydatid cyst disease is a significant zoonotic concern, particularly in developing countries like India. Within India, the northeastern region is gaining attention due to its unique socio-economic, cultural, and dietary habits, which contribute to the transmission and prevalence of this disease. Hydatid cyst disease, or cystic echinococcosis (CE), is caused by the metacestode stage of the tapeworm *Echinococcus granulosus*. It is a neglected zoonotic disease that leads to substantial economic losses in livestock and poses serious health risks to humans, resulting in high morbidity and mortality rates.

The geographical distribution and endemicity of CE vary significantly between countries and regions, influenced by various biotic and abiotic factors. These factors include environmental conditions, local agricultural practices, and the presence of definitive and intermediate hosts. High concentrations of *E. granulosus* eggs in specific rural areas can be an important epidemiological factor in the transmission of CE. The eggs are shed in the feces of infected canids and can contaminate the environment, leading to the infection of intermediate hosts.

The life cycle of *E. granulosus* involves definitive hosts, such as dogs and other canids, which harbor the adult tapeworm in their intestines. The intermediate hosts, typically sheep and other herbivores, become infected when they ingest eggs from contaminated environments. The eggs hatch in the intestines of these intermediate hosts, releasing larvae that penetrate the intestinal wall and spread to various organs, where they develop into hydatid cysts. These cysts can be found in the liver, lungs, spleen, kidneys, eyes, and even the brain. Humans become incidentally infected through the ingestion of food or water contaminated with *E. granulosus* eggs, or by direct contact with infected definitive hosts.

Consumption of meat containing hydatid cysts with viable protoscoleces can also lead to human infection. This often occurs in areas where traditional practices include the slaughter of livestock without proper veterinary inspection, leading to the accidental ingestion of infected meat. The presence of hydatid cysts in livestock not only affects animal health but also poses a risk to public health, as humans can develop severe and potentially fatal complications from the disease.

Numerous studies have reported the prevalence of hydatid cysts in various regions, but molecular characterization of the parasites is still limited. This lack of detailed genetic information hinders the understanding of the parasite's epidemiology and the development of effective control strategies. Molecular characterization can provide insights into the genetic diversity of *E. granulosus*, its transmission dynamics, and the potential for zoonotic transmission. Such information is crucial for designing targeted interventions to control and prevent CE.

Efforts to control hydatid cyst disease in endemic regions should focus on breaking the transmission cycle of *E. granulosus*. This includes measures such as regular deworming of dogs, improving sanitation to reduce environmental contamination with eggs, educating communities about the risks of consuming undercooked or raw meat, and promoting safe slaughtering practices. Additionally, public health campaigns can raise awareness about the disease and encourage preventive behaviors among at-risk populations.

In conclusion, hydatid cyst disease is a growing concern in the northeastern region of India due to the socio-economic and cultural practices of the people. It is a neglected zoonotic disease with significant implications for both animal and human health. Addressing this issue requires a comprehensive approach that includes improved diagnostic methods, molecular

characterization of the parasite, and effective control measures to reduce the transmission and prevalence of CE. By doing so, it is possible to mitigate the economic losses in livestock and improve public health outcomes in affected regions.

CLINICAL SYMPTOMS

Hydatid cyst disease, caused by the larval stage of the tapeworm *Echinococcus granulosus*, manifests with a variety of clinical symptoms depending on the size, location, and number of cysts within the affected organs. The disease progresses slowly, often remaining asymptomatic for years until cysts reach a significant size or cause complications. The liver is the most commonly affected organ, accounting for 50-70% of cases. Symptoms of hepatic hydatid disease include abdominal pain, a palpable mass in the right upper quadrant, and hepatomegaly. If a cyst ruptures, it can lead to severe complications such as anaphylactic shock, secondary bacterial infection, or biliary obstruction, which may present as jaundice. The lungs are the second most commonly affected site, representing 20-30% of cases. Pulmonary hydatid disease may cause chest pain, cough, hemoptysis (coughing up blood), and dyspnea (shortness of breath). In severe cases, a ruptured cyst can lead to hydropneumothorax, a condition where both air and fluid accumulate in the pleural cavity, causing respiratory distress. Other less commonly affected organs include the spleen, kidneys, heart, and brain. Splenic hydatid disease may present with left upper quadrant pain and splenomegaly. Renal hydatid disease can cause flank pain, hematuria (blood in urine), and urinary tract infections. Cardiac involvement, though rare, can lead to chest pain, arrhythmias, and heart failure. Cerebral hydatid disease can cause neurological symptoms such as headaches, seizures, and focal neurological deficits.

In some cases, cysts can also rupture into the peritoneal or pleural cavities, leading to secondary echinococcosis, which complicates the clinical presentation with additional symptoms such as generalized pain, peritonitis, or pleuritis.

Overall, the clinical presentation of hydatid cyst disease is highly variable and depends largely on the cyst's location, size, and whether complications such as rupture or secondary infection occur. Early detection and treatment are crucial to preventing severe complications and improving patient outcomes.

DIAGNOSIS AND TREATMENT

The asymptomatic nature of cystic echinococcosis (CE) infection in livestock presents significant challenges for surveillance and diagnosis. Diagnosing hydatidosis antemortem is difficult, as infected animals typically do not display overt clinical signs. However, post-mortem examinations often reveal the presence of hydatid cysts, making it easier to diagnose the disease after the animal has died. Molecular techniques have advanced the identification of *Echinococcus granulosus* genotypes, utilizing probes targeting regions such as the internal transcribed spacer (ITS) region, ribosomal DNA, and mitochondrial enzymes like Cox 1 and Nad1. These molecular methods have facilitated the detection and differentiation of various genotypes of the parasite.

Interestingly, *E. ortleppi*, a highly zoonotic strain of the parasite, has been identified in Mithun, underscoring the zoonotic risk posed by this infection. The diagnosis of extrahepatic echinococcal disease has become more accurate with the advent of advanced imaging techniques such as ultrasound, computed tomography (CT), and magnetic resonance imaging (MRI). These tools allow for the precise localization and characterization of hydatid cysts in organs other than the liver, such as the lungs, spleen, kidneys, and brain. Current treatment options for CE include surgical intervention, percutaneous drainage, and chemotherapy with antiparasitic drugs like Albendazole and Mebendazole. Surgical removal of cysts is often necessary in cases where cysts are large, symptomatic, or at risk of rupture. Percutaneous techniques, such as PAIR (puncture, aspiration, injection, re-aspiration), have been developed to manage cysts in less invasive ways. Chemotherapy with Albendazole and Mebendazole is used to reduce cyst size and prevent recurrence, particularly in cases where surgery is not feasible or as an adjunct to surgical treatment. Wildlife plays a critical role in the epidemiology of CE, as sylvatic cycles involving wild animals can intersect with domestic cycles involving livestock and sheepdogs. This interaction complicates control efforts, as wild animals can serve as reservoirs for the parasite, maintaining the infection in the environment and facilitating its transmission to domestic animals and humans. Effective control and prevention strategies require a comprehensive approach, including regular deworming of dogs, improving sanitation to reduce environmental contamination with *Echinococcus* eggs, educating communities about the risks of consuming undercooked meat, and promoting safe slaughtering practices. Additionally, monitoring and managing wildlife populations to minimize their interaction with domestic animals can help reduce the incidence of CE.

In conclusion, the asymptomatic nature of CE in livestock complicates diagnosis and surveillance efforts. Molecular techniques and advanced imaging methods have improved the accuracy of diagnosis, while current treatment options include surgery, percutaneous drainage, and chemotherapy. Addressing the zoonotic risk posed by wild animals and implementing effective control measures are essential to managing and preventing CE in endemic regions.

4. Sarcocystis infection

Sarcocystis is regarded as one of the significant emerging zoonotic parasites found in both domestic and wild animals, as well as in humans. Herbivores and omnivores serve as the intermediate hosts, while carnivores and humans act as the final hosts for Sarcocystis species. These parasites form septate cysts in various organs of the body, including the heart, diaphragm, tongue, esophagus, eyes, and skeletal muscles, and rarely in the brain of domestic animals such as cattle, buffaloes, pigs, sheep, goats, and other animals. Infection with certain species of Sarcocystis, such as *S. bovicanis*, has been associated with decreased feed intake and milk production. Poor productivity, lameness, recumbency, and abortion have also been attributed to Sarcocystis species. In dogs, *S. canis* has been linked to encephalitis, hepatitis, and generalized coccidiosis.

In humans, Sarcocystis hominis, derived from beef, and *Sarcocystis suis hominis*, from pork, cause infections that can result in the development of fever, headache, and myalgia, along with elevated serum enzyme levels and eosinophilia. The diagnosis is often confirmed by finding Sarcocystis in muscle biopsy specimens. In Assam, the prevalence of sarcocystosis in cattle was assessed using various diagnostic tests, including the Modified Agglutination Test (MAT), Agar Gel Precipitation Test (AGPT), Counter Immunoelectrophoresis (CIEP), and Indirect Haemagglutination Test (IHAT). It is suggested that all livestock are intermediate hosts for several Sarcocystis species with varying degrees of pathogenicity. The early phase of infection can lead to acute disease in the intermediate host, with the most pathogenic species causing more severe disease in these hosts than in the definitive hosts. Generally, species transmitted by canids or primates tend to be more pathogenic. The severity of the disease is closely related to the dose of infection and the immune status of the host. The developmental stages of the parasite in the vascular endothelial cells of the internal organs are more harmful than those in the muscular or nervous tissues.

Species affecting ruminants are considered the most pathogenic, and new infections are of particular concern. Animals that experience or survive primary infections develop immunity against the homologous species. However, chronic infection or low-grade infection can still lead to significant economic losses due to the reduced quality of livestock.

In summary, *Sarcocystis* poses a substantial threat to both animal health and human health. The economic impact on livestock production due to infections with *Sarcocystis* species underscores the need for effective diagnostic methods and control measures to mitigate this parasitic threat. Continued research and surveillance are essential to better understand the epidemiology, pathogenicity, and control of *Sarcocystis* infections in various host species.

DIAGNOSIS

Diagnosing *Sarcocystis* infection in domestic livestock involves various laboratory and clinical methods. The choice of diagnostic technique depends on the stage of infection and the specific *Sarcocystis* species involved. Here are some key diagnostic methods:

1. Microscopic Examination:

- **Histopathology:** Tissue samples from suspected animals are examined under a microscope to identify *Sarcocystis* cysts in muscles and organs. This method can reveal the presence of characteristic cysts, which are septate and vary in size.
- **Muscle Biopsy:** A muscle biopsy is taken from animals showing clinical signs of sarcocystosis. The biopsy is then stained and examined microscopically to detect cysts.

2. Serological Tests:

- **Modified Agglutination Test (MAT):** This test detects antibodies against *Sarcocystis* spp. in the serum of infected animals. It is useful for screening large populations.
- **Agar Gel Precipitation Test (AGPT):** This test identifies antibodies in the serum by precipitating antigen-antibody complexes. It is relatively simple and cost-effective.
- **Counter Immunoelectrophoresis (CIEP):** This method enhances the precipitation reaction by applying an electric field, increasing the test's sensitivity.
- **Indirect Hemagglutination Test (IHAT):** This test detects specific antibodies by measuring the agglutination of red blood cells coated with *Sarcocystis* antigens.

3. Molecular Techniques:

- **Polymerase Chain Reaction (PCR):** PCR is a highly sensitive and specific method for detecting Sarcocystis DNA in tissue samples. It can identify different Sarcocystis species and differentiate between closely related species.

4. Clinical Diagnosis:

- Clinical signs such as muscle pain, weakness, reduced productivity, and, in severe cases, neurological symptoms, can suggest sarcocystosis. However, clinical diagnosis alone is not definitive and should be supported by laboratory tests.

5. Enzyme-Linked Immunosorbent Assay (ELISA):

- ELISA is used to detect specific antibodies against Sarcocystis antigens in the serum. It is highly sensitive and can be used for both individual diagnosis and large-scale epidemiological studies.

Accurate diagnosis of Sarcocystis infection in livestock is crucial for effective management and control of the disease. Combining clinical signs with laboratory tests such as histopathology, serological assays, and molecular techniques enhances the reliability of the diagnosis. Early detection and intervention can mitigate the economic losses associated with sarcocystosis in domestic animals.

CONCLUSION

Zoonotic parasitic diseases that are prevalent in India and pose serious concerns include taeniasis, cysticercosis, and toxoplasmosis. These diseases are transmitted either through direct contact or by ingestion of pathogens, leading to chronic infections that affect various vital organs in the body. They are often associated with significant morbidity and mortality. Despite the availability of a wide array of diagnostic tests, ranging from microscopic detection of pathogens to sophisticated molecular diagnostic methods, serological tests remain the mainstay of laboratory diagnosis in the Indian context due to their cost-effectiveness. Serological tests can efficiently detect the presence of specific antibodies or antigens, making them suitable for large-scale screening and diagnosis. Taeniasis is caused by the ingestion of tapeworm eggs, leading to intestinal infections. Cysticercosis, a more severe form, occurs when larvae invade

tissues and form cysts, particularly in muscles and the central nervous system. Toxoplasmosis, caused by the protozoan *Toxoplasma gondii*, can lead to serious complications, especially in immunocompromised individuals and pregnant women. Effective management of these diseases requires rigorous surveillance to monitor and control outbreaks. Mass deworming programs using suitable anthelmintics are crucial for reducing the parasite burden in the population. These programs should be coupled with public health education to raise awareness about preventive measures, such as proper sanitation, safe food handling, and avoiding contact with contaminated soil or water. In the North-Eastern Hill (NEH) region of India, safeguarding the population from these parasitic infections involves coordinated efforts between healthcare providers, government agencies, and the community. Implementing regular deworming schedules, improving diagnostic facilities, and enhancing surveillance systems are essential steps towards achieving this goal. By focusing on these strategies, the NEH region can significantly reduce the incidence and impact of zoonotic parasitic diseases, ultimately improving public health outcomes.

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Chapter 7

Impact of climate change on parasitic diseases **J.K. Chamuah, Plabita Goswami, A.P. Milton¹, L.S Devi and Mahak Singh,**

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INTRODUCTION

Global warming and its impact on health have become widely discussed topics in recent times. Climate change is a major concern due to its implications for increased human exposure to vector-borne diseases that can be transmitted between humans and animals. Rising temperatures associated with climate change can extend the disease transmission season and increase the susceptibility of both humans and animals to various epidemic diseases. Climate change can be defined as a long-term and statistically significant shift in weather patterns, including temperature, humidity and rainfall in a particular geographic area over an extended period of time. The 21st century has witnessed an average global temperature rise of -1.5 to 5.8°C, accompanied by more frequent occurrences of extreme temperatures as projected by the Intergovernmental Panel on Climate Change (IPCC). The changing climate can have indirect effects on pathogens, vectors, hosts and their environments through alterations in their biology. Similarly, the occurrence and prevalence of livestock diseases in Great Britain have been extensively investigated in the context of climate change. It is evident that the unstable climate plays a significant role in the emergence of epidemic and infectious diseases affecting both domestic animals and human populations. The lengthened disease transmission seasons along with changes in the biology of pathogens, vectors and hosts contributes to this increased risk of epidemic diseases. Therefore, addressing climate change and stabilizing the climate is crucial for mitigating the impacts on disease dynamics and protecting the health of both animals and humans.

DIRECT IMPACT OF CLIMATE CHANGE ON DISEASE VECTORS

Climate change has direct effects on disease vectors, which are organisms that can transmit diseases from one host to another. These effects can significantly impact the transmission dynamics of vector-borne diseases and pose a risk to human and animal health. Climate change results in the changes of temperature, precipitation and humidity which influence the

reproduction, development, behavior and population dynamics of insects, different pathogens including microbes and different parasitic agents. Insects by physiology cannot regulate body temperature and thus causing shifting vector range patterns and risk to new threat or mutation of particular insect in order to accommodate particular environment, according to particular situation. Humidity and water is crucial for vector breeding, thus areas with standing water and high precipitation become favorable for more and rapid insect hatching. The incubation period of pathogens within vector is also temperature-dependant and become shorter in warmer condition. Some of the factors which affect climate change are:

1. **Distribution and Range:** Climate change can alter the geographical distribution and range of disease vectors. As temperature and precipitation patterns change, vectors may expand into new areas previously unsuitable for their survival. This expansion can introduce diseases to new regions and populations that were previously unaffected.
2. **Population Dynamics:** Climate change can influence the population dynamics of disease vectors. Warmer temperatures can lead to increased breeding rates and shorter life cycles for many vectors, allowing them to reproduce more rapidly and increase their population size. This can lead to higher transmission rates of diseases.
3. **Phenology and Seasonality:** Changes in climate can disrupt the phenology and seasonality of vectors. Phenology refers to the timing of biological events, such as the emergence of adult mosquitoes or the hatching of vector eggs. Climate change can alter these timings, leading to mismatches between the life cycles of vectors and their host availability. This mismatch can affect disease transmission dynamics.
4. **Behavior and Biting Rates:** Climate change can impact the behavior and biting rates of disease vectors. Rising temperatures may alter the activity patterns and feeding behaviors of vectors, such as mosquitoes or ticks. Changes in precipitation patterns can also affect their breeding sites and availability of water sources, influencing their abundance and biting rates.
5. **Vector Competence:** Climate change can influence the vector competence of disease vectors, which refers to their ability to acquire, maintain and transmit pathogens. Changes in temperature, humidity and other climatic factors can impact the vector's ability to carry and transmit diseases, potentially increasing or decreasing transmission rates.
6. **Molecular biology of the pathogen:** The ability of the microorganism to mutate the corresponding gene due to adverse effect of climate is the potential impact of climate

change, *e.g.*, the single amino acid substitution of glycoprotein envelope of venezuean equine encephalitis virus helps to adapt to epizootic vector of mosquito (*Ochlerotatus taeniorhynchus*). Due to climate change, RNA virus could easily respond to changes in vector population. In comparison to DNA viruses, RNA viruses have higher evolutionary rate due to their proof-reading capacity of gene. Mutation of virus microbes⁻¹ may easily change the nature of various biotic and abiotic factors that affect the geographical transmission of the organism.

7. **Wildlife habitat:** The habitat of wildlife and food sources will change along with change in feeding pattern to climate change which will ultimately change their social structure along with new introduction of domestic livestock. Climate change also promotes biological invasion of non-indigenous species with their adapted variety of newly introduced pathogens.

It is important to note that the specific effects of climate change on disease vectors can vary depending on the vector species, the disease they transmit and the local environmental conditions. Nonetheless, understanding and monitoring these direct effects are crucial for developing effective strategies for vector-borne disease prevention, control and mitigation in the context of a changing climate.

IMPACT OF CLIMATE CHANGE ON HELMINTH PARASITES

Climate change has contributed to the steady rise in the prevalence and intensity of helminth infections in both temperate and tropical regions. Changes in climate, including relative humidity and rainfall patterns, have impacted the epidemiology and transmission of these damaging parasites. Infections in host animals result in weight loss, lower milk yield, loss of condition, abortion, infertility and sporadic deaths. Climate change projections indicate an expected increase in summer temperatures by 2.0°C to 2.8°C by 2050 in different regions. The survival and development of parasitic free-living stages are heavily influenced by climatic conditions, which in turn affect the epidemiology and transmission of the parasites. Temperature is a crucial factor in the increased development of helminth larvae, although extreme temperatures can be lethal and reduce pasture contamination. Moreover, temperature fluctuations impact the population dynamics of parasites on livestock, influencing the seasonal incidence and overall epidemiology. Relative humidity also plays a vital role in larval survival and development. High humidity inside the host species provides a favorable environment for

hatching and full larval development. Climate not only affects larval survival and development within a grazing season but also influences over-winter survival, consequently affecting the availability of infective larvae in the following grazing season. Long-term climatic trends affect the distribution of different parasites in various geographical areas, while the timing and intensity of outbreaks depend on the presence of favorable environmental conditions. Climate change, along with increased weather variability, may also influence the spread of emerging parasites and lead to changes in species composition. Helminth infections in livestock were traditionally limited to species adapted to colder climates. However, with climate change, there has been an increase in the prevalence of parasites like *Haemonchus contortus* in warm regions, causing severe mortality in ruminant animals. In addition, the emergence of anthelmintic resistance and the potential for changes in the timing and duration of transmission seasons are concerning associated with climate change. The impact of climate change on animal and zoonotic helminth infections is a significant concern for the emergence and re-emergence of diseases. There is a need to address these challenges by implementing effective management strategies, monitoring parasite burdens and considering the development of new anthelmintic treatments. Additionally, research and surveillance efforts should be intensified to better understand the interactions between climate change and helminth infections, allowing for more targeted interventions and adaptation strategies. The climate change could affect the transmission cycle of helminthes parasites in different ways, such has been summarized below:

***Fasciola hepatica*:** Fasciolosis is one of the important ruminants' fluke disease which is characterized by weight loss, decreased milk yield, diarrhea, severe fibrosis of liver, clay pipe appearance of larvae and occasionally mortality. The prevalence and incidence of disease has been increased in entire globe due to changing epidemiology and infection pattern of flukes. There is also report of increase in its outbreak in sheep cattle⁻¹. There was an exceptional rise in *F. hepatica* and *F. gigantica* infections in both temperate and tropical climate due to increase construction of dam and irrigation facilities which helps in propagation of snail intermediate host. Summer rainfall is quite congenial for growth and propagation of snail epidemiology and the completion of their life cycle. The long-term changing distribution is attributed to warmer average temperatures year-round, increased rainfall in winter and autumn and increased humidity that leads to 4-week extension of the herbage growing season over the years.

***Nematodirus battus*:** *N. battus* is a highly pathogenic nematode found in the small intestine of ruminants, including cattle, buffalo and mithun. Unlike other livestock parasites, development of the free-living stages into infective larvae was thought to only occur after prolonged periods of exposure to low temperatures (around 0°C) followed by a rise in temperatures. Changes in

the pattern of infection and their epidemiology have been observed and it is now thought that the ability of *N. battus* eggs to hatch without a chilling period coupled with increased temperature is a predisposing factor for increased prevalence in domestic livestock.

***Haemonchus contortus*:** *H. contortus*, an abomasal nematode of ruminants, is typically a parasite of tropical climates, and it's the main cause of calves' mortality. There is a high prevalence and widespread distribution of *H. contortus*, but low outbreak intensity with few clinical cases has been observed in sheep and goat. This could be due to climate change, as warm temperatures lead to high larval activity which leads to increased infection rate. Higher autumn temperatures also encourage the ingested larvae to develop into disease inducing adults rather than enter hypobiosis, increasing the number of eggs released later in the year and extending the transmission window later into the autumn and major cause of spring rise phenomenon. It has been observed that larvae can withstand more desiccation during the hotter and drier summer months. The increased incidence of *H. contortus* has also been attributed to different non climatic factors like, increased anthelmintic resistance and changing of animal husbandry practice along with repeated exposure to anthelmintic.

IMPACT OF CLIMATE CHANGE ON TICKS AND FLEAS

Nowadays, the most severe impact of climate change has been noticed particularly in ticks like, the brown dog tick, *Rhipicephalus sanguineus*, which is endemic not only in Mediterranean regions but also in tropical climatic regions like India. Sporadic occurrence of these ticks in Europe, Asia including India, has also established their role of transmission of rickettsial organism in dogs. Regarding *Ixodes ricinus*, the castor bean tick is the most common among hard ticks, and plays a significant role in their development and transmission pattern among hosts with their epidemiology in domestic livestock in Europe due to suitable climatic conditions. The decrease of winter days with temperature below a certain cut-off was identified by several authors as a key factor in the increased density of ticks of the genus *Ixodes*. Using larva, nymph and adult ticks, documented that it needs certain environment of temperature and humidity for perpetuation of their life cycle. Increased prevalence of these ticks in Scandinavian countries is due to increased temperature which is suitable for its survival, development and completion of their life cycle. Climate change has significant implications for the distribution, abundance and behavior of ticks and fleas which are important vectors of diseases. Here are the key impacts of climate change on ticks and fleas:

Ticks:

1. **Expanded Geographical Range:** Warmer temperatures and changing climatic conditions have allowed ticks to expand their geographical range. Ticks that were once limited to specific regions are now being found in areas previously unsuitable for their survival. This expansion increases the risk of tick-borne diseases in new locations.
2. **Altered Seasonality:** Climate change can affect the seasonality of ticks. Rising temperatures and shifts in precipitation patterns can prolong the active season of ticks, extending the period during which they can transmit diseases. This can lead to increased exposure of humans and animals to tick-borne pathogens.
3. **Increased Abundance:** Favorable climate conditions can promote the survival and reproduction of ticks, resulting in increased tick populations. Warmer temperatures and increased humidity can enhance tick survival rates and contribute to higher tick densities in certain areas.
4. **Changes in Host-Vector Dynamics:** Climate change can influence the interactions between ticks and their hosts. Alterations in the behavior and distribution of host species, such as mammals and birds, can impact tick feeding patterns and their ability to acquire and transmit pathogens.

Fleas:

The impact of climate change on fleas is very difficult to ascertain due to its continuous life cycle through an entire year. A particular temperature $>25^{\circ}\text{C}$ and humidity $> 85\%$ is quite congenial for growth and propagation of different flea particularly of cat flea *Ctenocephalides canis*.

1. **Favorable Conditions for Reproduction:** Fleas thrive in warm and humid environments. As temperatures rise due to climate change, it can create more favorable conditions for flea reproduction and population growth. Increased temperatures and humidity can support their life cycle, leading to higher flea infestations.
2. **Prolonged Activity Period:** Rising temperatures can extend the activity period of fleas, allowing them to remain active for longer durations throughout the year. This prolongation of flea activity increases the risk of flea-borne diseases and the potential for infestations in both humans and animals.
3. **Impact on Wildlife:** Climate change can also affect the behavior and distribution of wildlife hosts that serve as reservoirs for fleas. Changes in the abundance and distribution of wildlife populations can influence flea populations and their potential for transmitting diseases.

Overall, climate change has the potential to significantly impact the distribution, abundance and disease transmission dynamics of ticks and fleas. Monitoring and understanding these effects are crucial for implementing effective prevention and control measures to mitigate the risks posed by tick and flea-borne diseases in a changing climate.

IMPACT OF CLIMATE CHANGE ON VECTOR BORNE DISEASES

The impact of climate change on vector-borne diseases and their consequences for public health and research policies have been extensively studied. Researcher Lendrum *et al.*, 2015 have investigated the effect of climate change on the risk of vector-borne diseases, particularly in the UK. Authors Rossati (2017) and Kenneth (2008) have discussed the different climatic factors that influence the transmission and distribution of these diseases. Climate change, which encompasses changes in temperature, precipitation, humidity and other climatic factors, significantly affects the reproduction, development, behavior and population dynamics of arthropod vectors. It can also impact the development of pathogens within vectors and the population dynamics and ranges of nonhuman vertebrate reservoirs for many vector-borne diseases. Whether climate change increases or decreases the incidence of vector-borne diseases in humans depends not only on climatic conditions but also on man-made non-climatic epidemiological and ecological factors. It is important to consider the contributions of other global change agents, such as increased trade and travel, demographic shifts, civil unrest, changes in land use, water availability and other factors, in predicting the overall impact of sustained climate change on vector-borne diseases. Adapting to the effects of climate change requires the development of adequate response plans, enhanced surveillance systems and the implementation of effective and locally appropriate strategies to control and prevent vector-borne diseases. Vector-borne diseases continuously contribute to the global burden of disease, causing epidemics that disrupt health security and have wide socioeconomic impacts worldwide. The ongoing variability in weather patterns threatens to undermine the progress made in combating these diseases globally. Efforts by the World Health Organization (WHO) and its partners have been reviewed in addressing the challenge posed by climate change and vector-borne diseases. While there has been a focus on attributing past changes in disease rates to climate change and using scenario-based models to project future changes in risk, the uncertainty associated with such analyses and the dependence on other socioeconomic and public health determinants limit their utility as decision support tools. The priority for health

agencies is to strengthen control efforts, reduce current disease rates and manage short-term climate risks, which will ultimately increase resilience to long-term climate change.

The WHO and its partner agencies are working through a range of programmes and initiatives to

- (iii) achieve political support and financial investment in preventive and curative interventions in cutting down current disease burdens;
- (ii) Promote a comprehensive approach to climate risk management;
- (iii) Support applied research, through definition of global and regional research agendas, and targeted research initiatives on priority diseases and population groups.

IMPACT OF CLIMATE CHANGE ON MOSQUITO

Temperature plays a crucial role in the lifecycle of mosquitoes and the transmission of diseases like malaria. Each stage of the mosquito's lifecycle is influenced by temperature with minimum and maximum thresholds determining their development and survival. For instance, above a certain temperature threshold, Anopheles mosquitoes which are the vectors for malaria cannot survive. Higher temperatures result in a faster mosquito lifecycle that does not allow enough time for the development of the Plasmodium parasite within the mosquito's salivary glands. Therefore, temperature acts as a limiting factor for the transmission of malaria. Temperature affects both the vector population and the parasite within the vector. While the availability of water and moisture primarily impacts the vector, temperature influences the development and survival of both the mosquitoes and the malaria parasite they carry. In the last decades, outbreaks of malaria have been observed in mountainous regions of Kenya, Uganda and Rwanda. However, attributing these resurgences solely to regional climate changes can be overly simplistic. East Africa experiences significant temporal and spatial variations in climate, suggesting that other factors may contribute to the reported malaria outbreaks. Factors such as migration, breakdown in health service provision and vector control operations and deforestation have also been identified as contributing factors to the increase in malaria cases. These factors can disrupt vector control measures, reduce access to healthcare and alter the habitat and breeding patterns of mosquitoes, leading to an increase in malaria transmission. Understanding the complex interactions between climate, vectors, parasites and human activities is essential for effectively addressing the resurgence of malaria and implementing appropriate prevention and control strategies. Climate change is just one piece of the puzzle,

and a comprehensive approach that considers multiple factors is necessary to combat the disease effectively.

NATIONAL ACTION PLAN ON CLIMATE CHANGE

1. Assessment of increased burden of disease due to climate change.
2. Providing high-resolution weather and climate data to study the regional pattern of disease.
3. Development of a high-resolution health impact model at the state level.
4. GIS mapping of access routes to health facilities in areas prone to climatic extremes.
5. Prioritization of geographic areas based on epidemiological data and the extent of vulnerability to adverse impacts of climate change.
6. Ecological study of air pollutants and pollen (as the triggers of asthma and respiratory diseases) and how they are affected by climate change.
7. Studies on the response of disease vectors to climate change.
8. Enhanced provision of good health care facilities and implementation of public health measures and awareness, including vector control, sanitation and clean drinking water supply.

CONCLUSION

Parasitic infections have a significant impact on livestock production, leading to reduced meat and milk yields. The life cycle and propagation of parasites are often restricted to specific climatic conditions. With the changing climate scenario, there is an increased prevalence and intensity of parasitic infections making their control more challenging. It is crucial to anticipate these changes and prepare for methods to limit outbreaks and the prevalence of parasitic diseases on a global scale in the context of climate change. Scientists have developed forecasting models to predict the occurrence of different parasites in specific areas in response to climate change. These predictions can help inform proactive measures to mitigate the impact of climate change on parasitic infections. Additionally, there are predictions of zoonotic diseases, which are diseases transmitted between animals and humans in response to climate change. Such forecasts emphasize the importance of addressing climate change as a means to prevent the spread of these diseases. Clinical practitioners play a vital role in addressing the challenges posed by climate change. They can take urgent steps to mitigate climate change issues by implementing effective management programs and promoting the planting of vegetation that reduces greenhouse gas emissions and acts as a carbon sink. Furthermore, it is

essential for the scientific community to unite and make efforts against climate change. Scientists can contribute by developing biological interventions and strategies to mitigate climate change and its impact on parasitic infections. Applied research is crucial in reducing the burden of vector-borne diseases in the face of climate change and other environmental and social determinants. To ensure relevance to current health programming, there is a need to bridge the gap between research and the demands of health policy-makers. This involves complementing research on detecting and attributing health effects to climate change with a more applied approach to assessing and managing climate-related risks in the present. Policies and frameworks on health adaptation to climate change, including the prioritization of vector-borne diseases, present opportunities to strengthen the connection between applied research and public health policy.

In conclusion, addressing the impact of climate change on parasitic infections requires proactive measures, collaboration between clinical practitioners and scientists and the integration of applied research into public health policies. By taking these steps, we can manage the risks better posed by parasitic diseases in a changing climate and safeguard livestock production and human health.

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Chapter 8

STRATEGY FOR MANAGEMENT OF CALF DIARRHOEA IN FARM CONDITIONS

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Mithun (*Bos frontalis*), 'the cattle of the mountains', is the pride of the north-eastern hill region of India, which is distributed in four states of Arunachal Pradesh, Nagaland, Manipur, and Mizoram, and is known to be a symbol of status and prosperity, peace and harmony among the tribal people of this region. Mithun is not only found in India, but also in the Indo Himalayan countries of Myanmar, Bhutan, Bangladesh, China, and Malaysia. Traditionally it is a ceremonial animal and used for sacrifice during festivals and social ceremonies by various tribal communities. Mithuns have a browsing habit. Their draught potential has been demonstrated in the remote hilly terrains. Due to congenial atmosphere and suitable climate, they are susceptible to wide variety of diseases including bacterial, viral and parasitic diseases. Among these, calf diarrhoea is very common due to bacterial, viral and parasitic causes. Calf diarrhoea due viral (rotavirus infections) and bacterial (*Escherichia coli*) are the primary causes of neonatal mortality. Therefore, a comprehensive understanding of the causes of calf diarrhoea and strategies available for its control is crucial to avoid economic losses in a farm.

ETIOLOGY OF DIARRHOEA:

1. Bacterial causes:

E. coli, *Citrobacter freundii*, *Proteus mirabilis*, *C. koseri* and *Klebsiella pneumonia* were the most frequent, followed by *Staphylococcus sp.* *Enterococci*, *Enterobacter aerogenes*, *Alkaligenes dispar*, and *Providencia sp.* are important causes of calf diarrhoea.

2. Parasitic causes:

Toxocara vitulorum, *Haemonchus contortus*, *Bunostomum phlebotomum*, *Amphistome spp.*, *Fasciola gigantica*, *Cryptosporidium parvum*, and different Eimerian species are common etiologies that cause diarrhoea in mithun calves.

3. Viral causes:

Rota virus, Corona virus, Bovine viral diarrhoea virus and enteric calicivirus are also commonly associated with diarrhoea in mithun calves.

4. Nonspecific causes:

Indigestion, Overfeeding or Underfeeding, Sudden Diet Changes, Poor Quality Feed, Environmental Stress, Inadequate Colostrum Intake, Improper Housing and Overusage of Antibiotics are the non-specific causes precipitating diarrhoea in calves.

SIGNS OF CALF DIARRHOEA:

- ❖ Loose or Watery Stools
- ❖ Dehydration
- ❖ Sunken Eyes
- ❖ Dry Mucous Membranes
- ❖ Reduced skin elasticity, where a pinch of skin takes longer to return to its normal position.
- ❖ Weakness and Lethargy
- ❖ Weight Loss and Poor Growth
- ❖ Abdominal Pain and Discomfort
- ❖ Arching of the Back Indicative of pain.
- ❖ Kicking at the Belly
- ❖ Reduced Appetite
- ❖ Depending on the underlying cause, calves may have an elevated body temperature.
- ❖ Diarrhea can make calves more irritable, restless, or lethargic.
- ❖ Calves may drink more water in an attempt to compensate for fluid loss.
- ❖ In some cases, calves may strain to defecate, which can indicate more severe gastrointestinal issues.
- ❖ In severe cases, diarrhea can lead to systemic illness
- ❖ Shock Characterized by cold extremities, weak pulse, and rapid heart rate.
- ❖ Sepsis showing Signs of a severe systemic infection, including severe lethargy, fever, and rapid deterioration.

MANAGEMENT OF CALF DIARRHOEA:

- ❖ Replace the lost water and electrolytes at the earliest -Feed 2-4 litres of electrolyte solution every day
- ❖ The electrolyte solution provided should be over and above the normal feeding.
- ❖ Consult a veterinarian at the earliest to determine the cause of diarrhoea and to provide appropriate treatment.

On-field electrolyte solution composition

(For 1 litre of warm water)

Glucose * - 5 teaspoons

Soda bi carb - 1 teaspoon

Table salt - 1 teaspoon

10 Preventive Strategies to Mitigate Calf diarrhoea:

1. Provide high-quality colostrum (with high immunoglobulin content) and in sufficient quantities (usually 10% of the calf's body weight within the first 24 hours).
2. Maintain a clean and dry environment in the calving area to reduce exposure to pathogens.
3. Regularly clean and disinfect feeding equipment, housing, and other tools that come into contact with the calves.
4. Provide a balanced diet that meets the nutritional needs of the calves, including adequate energy, protein, vitamins, and minerals. Avoid sudden changes in diet, housing, or management practices to minimize stress.
5. Ensure that calves have access to clean and fresh water at all times. Contaminated water can be a source of infectious agents.
6. Consider the use of probiotics and prebiotics to support gut health and enhance the immune response of the calves.
7. Implement a vaccination program strictly in the mithun farms
8. Strict monitoring of calf health regularly in mithun farms
9. Maintenance of strict biosecurity measures in the farms

Chapter 9

A Comprehensive Guide to Herd Health Management in a Commercial Farm

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INTRODUCTION

Herd health management involves proactive and systematic approaches to disease prevention, early detection, and treatment within a livestock population. This approach helps maintain high levels of productivity and animal welfare while minimizing costs associated with disease outbreaks.

KEY COMPONENTS OF HERD HEALTH MANAGEMENT

2.1. Disease Prevention

Biosecurity Measures: Implement stringent biosecurity protocols to prevent the introduction and spread of infectious diseases. This includes controlling access to the farm, disinfecting equipment, and quarantining new or sick animals.

Vaccination Programs: Develop and follow a comprehensive vaccination schedule tailored to the specific diseases prevalent in the region and species of livestock.

Nutrition and Feed Management: Provide balanced and adequate nutrition to support the immune system and overall health of the herd. Ensure feed is stored properly to prevent contamination.

2.2. Monitoring and Early Detection Regular Health Assessments: Conduct routine health checks to monitor the physical condition of the animals. Look for signs of illness, lameness, and abnormal behavior.

Record Keeping: Maintain detailed records of health status, treatments, vaccinations, and production data for each animal. This helps in identifying trends and making informed decisions.

Surveillance Programs: Implement disease surveillance programs to detect early signs of outbreaks. This can involve regular testing of blood, milk, or feces samples.

2.3. Disease Control and Treatment

Quarantine Protocols: Isolate sick animals to prevent the spread of disease. Designate separate areas for treating and housing quarantined animals.

Veterinary Care: Establish a relationship with a qualified veterinarian who can provide regular health checks, emergency care, and guidance on treatment protocols.

Medication Management: Use antibiotics and other medications judiciously to avoid resistance. Follow the prescribed dosage and withdrawal periods to ensure food safety.

2.4. Reproductive Management

Breeding Programs: Implement selective breeding programs to enhance genetic resistance to diseases and improve overall herd health.

Reproductive Health Monitoring: Regularly check reproductive organs and monitor fertility indicators to ensure successful breeding and calving.

3. Specific Herd Health Strategies for Different Types of Livestock

3.1. Dairy Cattle

Mastitis Control: Implement strict milking hygiene practices, regular udder health monitoring, and appropriate treatment protocols.

Lameness Prevention: Maintain proper flooring, provide regular hoof trimming, and ensure balanced nutrition to prevent lameness.

Calf Health Management: Ensure colostrum intake within the first few hours of birth, vaccinate calves, and monitor their growth and development closely.

3.2. Beef Cattle

Nutritional Management: Provide high-quality forage and supplements to support growth and immune function.

Parasite Control: Implement regular deworming and control external parasites through appropriate treatments and pasture management.

3.3. Swine and Poultry

Biosecurity: Strictly control access to the farm, disinfect equipment, and implement all-in/all-out systems to reduce disease transmission.

Respiratory Health: Monitor for respiratory diseases, vaccinate against common pathogens, and ensure proper ventilation in housing facilities.

Vaccination Programs: Follow vaccination schedules for diseases like Newcastle disease, avian influenza, and Marek's disease.

Environmental Control: Maintain optimal temperature, humidity, and ventilation in poultry houses to prevent respiratory and heat-related illnesses.

4. Integrating Technology in Herd Health Management

4.1. Precision Livestock Farming

Wearable Technology: Use sensors and wearable devices to monitor vital signs, activity levels, and other health indicators in real-time.

Data Analytics: Employ data analytics to interpret health and production data, enabling early detection of issues and informed decision-making.

4.2. Software Solutions

Health Management Software: Utilize farm management software to keep track of health records, treatments, and vaccination schedules.

Automated Systems: Implement automated feeding, milking, and climate control systems to ensure consistent and optimal care.

5. Training and Education

5.1. Staff Training

Health Protocols: Train farm staff on biosecurity measures, disease detection, and proper handling of animals.

Emergency Procedures: Ensure staff are well-versed in emergency response procedures for disease outbreaks and other health crises.

5.2. AWARENESS PROGRAMS

Workshops and Seminars: Attend industry workshops and seminars to stay updated on the latest advancements in herd health management.

Veterinary Consultations: Regularly consult with veterinarians to review health management practices and update protocols as needed.

6. ECONOMIC CONSIDERATIONS

6.1. Cost-Benefit Analysis

Preventive Measures vs. Treatment: Compare the costs of preventive measures against the potential expenses of disease outbreaks and treatments.

Investment in Technology: Evaluate the long-term benefits of investing in technology and automation for health monitoring and management.

CONCLUSION

Effective herd health management is vital for the sustainability and profitability of commercial farms. By implementing comprehensive disease prevention, monitoring, and control strategies, integrating technology, and investing in staff training and education, farmers can ensure the well-being of their livestock and achieve optimal production outcomes. Continuous evaluation and adaptation of health management practices, can help in overcoming challenges and maintaining a healthy herd.

Chapter 10

Prevention, control and management of different bovine diseases for profitable dairy farming system

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Dairy sector plays a significant role in generating employment opportunities, providing a basic food source, improving the country's economy, afford stable income to rural households and helps to combat poverty. The economic effects of the dairy industry are consequential in India due to its major contribution to the country's GDP. The dairy and animal husbandry sector contributes 4.2% of the national GDP (National Accounts Statistics, GoI, 2019). However, the majority of the milk supply channel is informal and lacks maintenance of quality. There is an increased demand for hygienic milk production hence it is imperative to take stringent measures for improved management system so that clean milk may be available for the human consumption.

The management aspect plays a key role various good management practices to be conducted for running a good dairy farm.

The management practices to obtain better results for successful dairy farming are:

DAIRY FARM HOUSING SYSTEM

Mainly there are two systems of housing for dairy animals: Loose housing system and Conventional closed housing system.

In tail-to-tail system, which is mostly favoured, the animals do not face each other and their feeding mangers are different. Tie stall barns require a ventilation system that allows close control of air exchange and temperature to prevent the barn from reaching freezing temperatures. Good air distribution is especially critical because the cows are restrained and cannot move out of drafts or poorly ventilated areas. Naturally ventilated tie stall barns use sidewall openings only or combinations of sidewall and ridge or stack openings. Figures 1-3 illustrate these systems and their requirements. For all systems the barn must be well constructed and insulated similarly to a mechanically ventilated barn. Openings require adjustment in response to wind and temperature changes. Automatic temperature-adjusting

controllers will respond to changes even if no one is in the barn. Each sidewall and ridge or stack should be controlled separately.

- Continuous adjustable sidewall opening, 4 to 6 feet high.
- Large doors or removable panels in end-wall for increased hot weather ventilation.
- Manual adjustment for warm seasons
- Thermostat-controlled automatic adjustment provides better conditions during changeable freezing weather.
- Large doors or removable panels in endwall for increased hot weather ventilation.
- Excellent air/vapor barrier between animal space and attic space.
- Ventilate all enclosed attic spaces above animal areas (1 square inch of opening for each 1 square foot of ceiling area).
- Trouble shooting Low milk production
- Skills in detective work are sometimes more valuable than knowing the ins and outs of nutrition. Today's computer models make ration formulation almost too easy. When troubleshooting "nutrition" problems many people start with the paper ration. However, in reality the problem many times is in the implementation. This is where science and art come into play.

Cost of construction is significantly lower than conventional type. In this system It is possible to make further expansion without change, it better facilitate easy detection of animal in heat. Animals feel free and therefore, proves more profitable with even minimum grazing. Animals get optimum exercise which is extremely important for better health production. Animal health: Poor animal health is one of the principal constraints to increasing small-scale dairy productivity, as it results in high morbidity and low production. Overcoming this constraint could significantly improve productivity and result in real and direct benefits for producers. Good dairy farming practices for animal health are establishing the herd with resistance to disease; preventing the entry of disease on to the farm; establishing effective herd health management; and using all chemicals and veterinary medicines as directed.

CALVING PEN

The Calving Pen is designed to accommodate any animal that requires immediate attention. For sick animals that need to be isolated from the herd, the Calving Pen provides great access to safely perform necessary treatments and procedures. Individual calving pens should be

approximately 12 feet by 12 feet, or approximately 144 square feet. pen, which is easily cleaned after each calving.

NUTRITIONAL REQUIREMENTS FOR THE DAIRY ANIMALS

Nutrition (feed and water): A dairy animal's health and productivity, as well as the quality and safety of its milk, depend largely on providing the right feed and water. The requirements for feed and nutrients of dairy animals depend on factors such as physiological state, milk production level, age, sex, body condition, body weight, weight gain, health condition, level of activity and exercise, climate and season. The feeding of livestock is a major challenge in many developing countries. This challenge is even greater in the tropics because of seasonal fluctuations in the availability of feed – caused by periods without rainfall – and the poor quality of feed. When producers cannot rely on locally available feed resources, the feeding of dairy animals can become more expensive. The feeding methods used by small-scale dairy producers in developing countries are grazing, which requires fairly large areas; tethering, which permits full use of roadside verges, areas around cropland, etc.; and stall or pen feeding, which requires more labour inputs. When supplements are provided, they are either fed to the entire herd or to individual animals. Dairy animals consume large amounts of water for milk production and pregnancy. Access to water therefore has a great influence on milk production. Good dairy farming practices for nutrition are securing feed and water supplies from sustainable sources; ensuring a supply of feed and water of suitable quantity and quality; controlling the storage conditions of feed; and ensuring the traceability of feedstuffs brought on to the farm. There are a range of diseases affecting dairy cows with different impacts on welfare, productivity and profitability, so it makes sense to have a good understanding of common conditions.

Dry fodder is required to get sufficient fat in the milk. The sources of dry fodder are hay, stovers like jowar kadbi and straws like paddy straw, wheat bhussa, Karad grass which contains 10-15% moisture. Total 3-6 kg of dry fodder/day is required to adult cattle. Lactating cow weighing 300 to 500 kg body weight needs approximately 20-25 kg greens for the body development and production. Lactating animal should be given 1 Kg. additional concentrate for every 2.5 Kg to 3 Kg.

ANIMAL HEALTH

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Chapter 11

Lumpy skin disease: epidemiology, prevention and control

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INTRODUCTION

Lumpy skin disease (LSD) is a vector borne pox viral disease of cattle and buffaloes that is characterized by appearance of skin nodules. The natural hosts for this virus are cattle and Asian water buffaloes. Considering the potential for international spread and economic impacts the World Organization for Animal Health classified the disease as notifiable disease. LSD causes huge economic impact on livestock sector due to decreased milk production, loss of draught power, damage to skin, trade restrictions, and loss of body condition, abortions, infertility and the cost of veterinary care. The disease was first identified in Zambia in 1929. The LSD has spread from its origin's in central Africa to the Middle East, Europe and Asia with rapid spread occurring since 2013. India reported first time in the year 2019 in Odisha state. The surrounding countries of India viz., Bangladesh, China, Nepal, Bhutan, Myanmar, Sri Lanka, and Pakistan have also reported the LSD outbreaks. The whole bovine population (cattle, buffaloes, yak and mithun) in the country are at risk of getting infected LSD.

ETIOLOGY

The Lumpy Skin Disease is caused by Lumpy Skin Disease virus (LSDV) that belongs to the genus *capripoxvirus* of the family *Poxviridae*. LSDV is a brick shaped, enveloped virus with a double stranded DNA (151 kbp). The average size of the virus is 294 ± 20 nm in length and 262 ± 22 nm in width (Kitching and Smale 1986). There is only one serotype of LSDV. LSDV is closely related to Sheep pox virus (SPPV) and Goat pox virus (GTPV) but are phylogenetically distinct from each other. All capripoxviruses grow slowly on cell cultures and may require several passages. They can be propagated on a variety of cells of bovine and ovine origin, causing easily recognizable cytopathic effects (CPE). The replication of LSDV occurs in the cytoplasm of the host cell causing intracytoplasmic eosinophilic inclusion bodies. The antibodies produced against one member is serologically cross reactive with the other. Hence, there differentiation among the members of genus capri poxvirus is possible through molecular techniques like PCR and sequencing. LSDV is susceptible to sunlight and detergents containing lipid solvents, but in dark environmental conditions, such as contaminated animal sheds, it can persist for many months. The virus can survive for long in cold conditions and as

per the current information available, there is no evidence about difference in virulence of the different LSDV strains.

HISTORY

The initial description of the LSD clinical indications was made from Zambia, in 1929. Up to 1949, there were ongoing reports of disease, which caused severe economic losses in South African nations. Disease first appeared in West Africa in 1974 and East Africa in 1957. In a succession of epizootics that were documented, the disease has continued to spread across the majority of the African continent. LSD is now present throughout the Africa (with the exception of Libya, Algeria, Morocco, and Tunisia). Outside of the sub-Saharan African region, Egypt (1988), Kuwait (1991), Yemen (1995), United Arab Emirates (2000), Bahrain (2003), and Oman (2010) had all reported outbreaks of lumpy skin disease. In India, first outbreaks of lumpy skin was reported in Mayurbhanj and Bhadrak Districts of Odisha in the month of August 2019.

EPIDEMIOLOGY

Typically, in endemic countries LSD outbreaks occur in epidemics with several years apart. The existence of a specific reservoir for the virus is not known, nor is how and where the virus survives between epidemics. The presence of growing numbers of naïve animals, abundance of active blood feeding vectors and uncontrolled animal movements are usually the drives for LSD outbreaks. The first outbreak in an unaffected region is usually attributable to either legal or illegal movement of host (cattle/buffaloes). Generally, the long distance jumps are due to host movement and local spread is due to vectors. Outbreaks are usually seasonal and co-inside with abundance of vector population during wet and humid conditions. But, may occur at any time because in many affected regions no season is completely vector free. The morbidity (10-20%) is high and mortality is very low (1-5%). Infected animal eventually clears the infection and there is no scientific evidence for establishment of carrier status in the recovered animals. Even though LSD reported mainly in cattle, the disease has been reported in Asian water buffaloes and African antelope species. *Bos taurus* is more susceptible to clinical disease than *Bos indicus*. However, even within the herd maintaining the same breed, the clinical manifestation varies from subclinical to fatal disease. Although sheep and goats co-exist with cattle and buffalo, the role of small ruminant as reservoir of LSDV has not yet been confirmed have not been implicated as reservoir of LSDV. The role of wildlife in the epidemiology of LSDV requires further investigation. LSDV is not known to infect humans.

Risk factors for disease occurrence includes effect of agro climate, share of the same grazing and water bodies and unrestricted movement of animals across different borders that would facilitate the spread of outbreaks in various localities. High occurrence of LSD was recorded during wet seasons when biting fly populations are abundant (Table 1). Necrotic skin and air-dried hides are the potential source of virus since the virus persists for month at ambient temperature. Imports of any animal product like meat, milk, or hides originating from a LSD affected country/ies have potential to spread in LSD free countries. During the infective phase, virus secretes in nasal, lachrymal, pharyngeal secretions, semen, milk and blood and may act as source of infection to others animals.

Factors	Variables	Effect
Host	Species	<ul style="list-style-type: none"> • Cattle are more susceptible than buffaloes
	Sex	<ul style="list-style-type: none"> • Both sexes are equally susceptible
	Age	<ul style="list-style-type: none"> • Very young and old animal are more suspectable than adults
	Breed	<ul style="list-style-type: none"> • The cross breeds are more susceptible than native Indian breeds however the native breeds are more reactive
Pathogen	Serotype	<ul style="list-style-type: none"> • Only one serotype • Not much variation in pathogenicity between strains
	Stability/viability of virus	<ul style="list-style-type: none"> • Stable and viable in both dry and cold conditions. • In biological samples (Blood: 5-16 days, Skin nodule: 39 days, Scabs: several years and semen: 22-42 days) the virus viability varies.
Environment	Temperature	<ul style="list-style-type: none"> • LSDV can survive both cold and warm conditions. But, inactivated by heat easily.
	pH	<ul style="list-style-type: none"> • Sensitive to both acidic and alkaline pH
	Season	<ul style="list-style-type: none"> • Incidence is more in wet and humid seasons due to abundance of vector population • Incidence is less during the dry and cold seasons. • Incidence is more during migratory season and livestock fairs or sales or ritual/festivals

Table.1: Various risk factors and their effect on of the lumpy skin disease.

LUMPY WAVES IN INDIA:

First wave (2019-2020): First outbreaks occurred in Odisha in 2019, where the morbidity was higher and mortality was very low. The disease was also spread to other neighboring states like including all southern states. But the disease was mild and mostly dermal form of disease was reported in 2019-2020.

Second wave (2020-2021): During this period most of southern states were affected along with other central states but the disease again was mostly of dermal form and caused many deaths in Maharashtra state compared to other affected states including MP and UP.

Third wave (2021-2022): This is ongoing and the disease has turned into more virulent form especially in this wave respiratory form of the disease was more prevalent along with dermal form and maximum mortality was reported from states especially northern India where more of stary or owned cattle population is more. The mortality was also attributable to comorbidities in these stary and unowned or even in animal shelters which are mainly due to managerial practices (Fig.1).

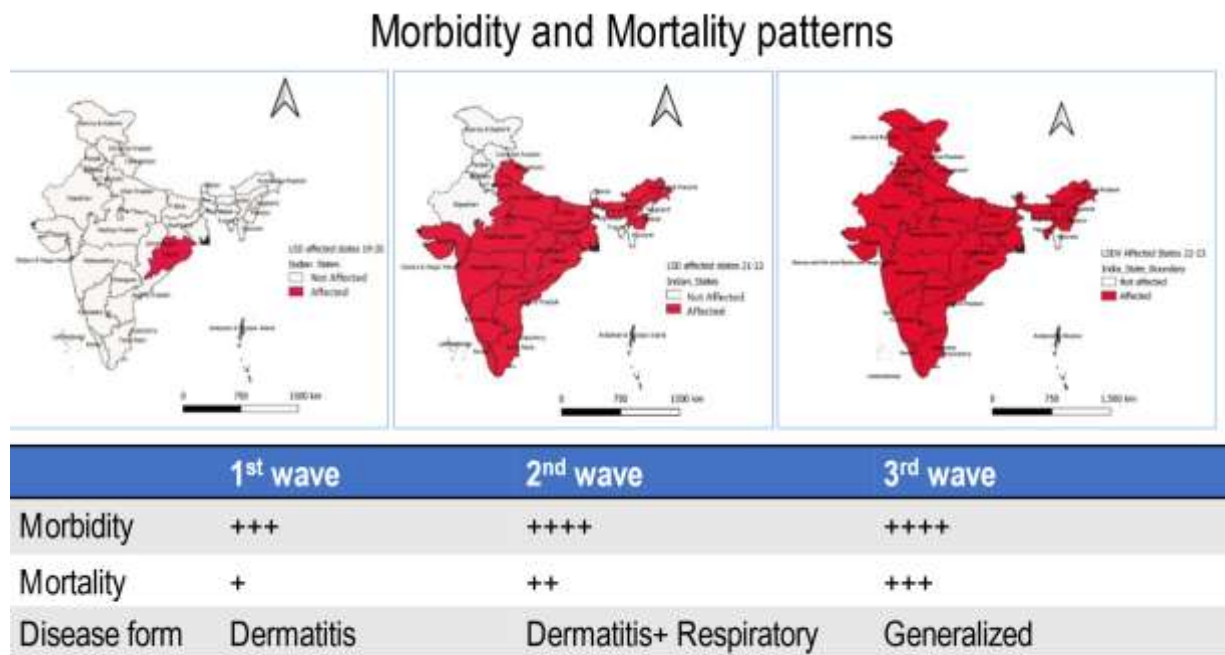


Fig.1: Patterns of LSD in three different waves in India

ECONOMIC IMPACT

Lumpy skin disease impacts cattle and buffaloes production by causing substantial economic losses. Different stakeholders being affected are animal breeders, dairy farmers, agriculturist (drought power), milk and meat processing and selling industries, wholesale and retail dairy and meat sellers, organic farming community, cattle transport and feed manufacturers etc.,. Among the stakeholder the highest impact will be on landless or marginals poor, small scale and backyard farmers who’s entire earning depends on animal farming only. The economic loses are attributed to decreased milk production, decreased body condition, damage to hide/skin, drought power loss in bullocks, fertility problems and abortions, emaciation, veterinary treatment, secondary complication like maggot wound, mastitis,

temporary or permanent blindness. Above all the disease also causes mortality (deaths) upto 5% of affected animals which account direct loss. Other costs include national cost of surveillance and control measures like awareness campaigns, monitoring and surveillance program, compensation program for culled or slaughtered animals, cleaning and disinfection of affected farms, sampling and laboratory testing, purchase of vaccines and implantation of a vaccination program, vector control, restriction on movement of live animals and products and carcass disposal. The traders will be affected due to ban on export of live animals and animal products in and out of country.

Transmission

The following are the routes of transmission

- Blood sucking/biting of vectors - Stable fly (*Stomoxys calcitrans*), Mosquito and Ticks
- Contaminated feed/fodder and water with saliva, nasal and ocular discharges.
- Direct contact with infected animals.
- Vertical transmission: Contaminated semen
- Intrauterine and Trans-mammary

The long-distance jump or transmission of virus is mainly due to movement host and rarely due to transport vehicle is carrying the vectors and short distance jump or transmission due to mainly vectors and other modes of transmission.

CLINICAL SYMPTOMS

The incubation period may vary between 4-14 days in experimental conditions, however in field conditions it may go up to five weeks. The clinical signs include nasal and ocular discharge, pyrexia, brisket oedema (Fig 2), upper limb oedema, drastic reduction in milk production and characteristic skin lesions. Initially, subscapular and pre-femoral lymphnodes become enlarged and are easily palpable (Fig 2). High fever (>40.5) usually precedes the appearance of skin lesions and may be accompanied by a sharp drop in milk yield. The fever may persist for a week. Mild clinical cases may show a few nodules. In severe cases the entire body may be covered with skin lesions (Fig. 2). Pneumonia due to secondary bacterial infection is a common sequela. Sometimes ulcerative lesions may also appear in the cornea of the one or both eye, leading to blindness in some cases. Skin nodules usually start to appear within 48 hours of the onset of fever. They are most commonly found at head, neck, udder, genitalia (Fig. 2), perineum and lateral parts of ribcages (Fig 2). Silent or subclinical infections can occur up

to one third of infected cattle which do not show any clinical signs although all might have viraemia.

The morbidity and mortality depending the control measures adopted after detection of first case. During the typical course of disease, cattle die several weeks after infection. Thus the outbreak is likely to have been going on for a minimum of 2-3 weeks and probably longer. Severe cases of LSD are highly characteristic and easy to recognize or diagnose. However early stages of infection and mild cases may be difficult to distinguish even for the most experienced veterinarians.



Fig.2. The LSD confirmed animal showing brisket oedema, enlarged prescapular lymph node, and various sized nodules all over the body in local and cross breed affected animals.

PATHOGENESIS

The Lumpy Skin Disease (LSD) virus infects the host through the skin or the mucosa of the gastrointestinal system. Primary multiplication of virus causes lymphadenitis when it enters the local lymph nodes. Viraemia occurred after the initial febrile reaction and persisted for two weeks. After viremia, virus spread through monocytes and localized in the skin and developed the inflammatory nodules as a result of its rapid multiplication in the cells. LSDV

replicates inside the host cells such as macrophages, fibroblasts, pericytes and endothelial cell in the lymphatics and blood vessels walls lead to developing vasculitis and lymphangitis, while thrombosis and infarction may developed in severe cases. Virus multiplication in cells produce hyperplasia and ballooning degeneration of keratinocytes, formation of epidermal micro vesicles, inflammatory cell infiltration into dermis. Four to seven days after infection coalescence of epidermal micro-vesicles occur into large vesicle and ulceration of tissue developed.

IMMUNITY

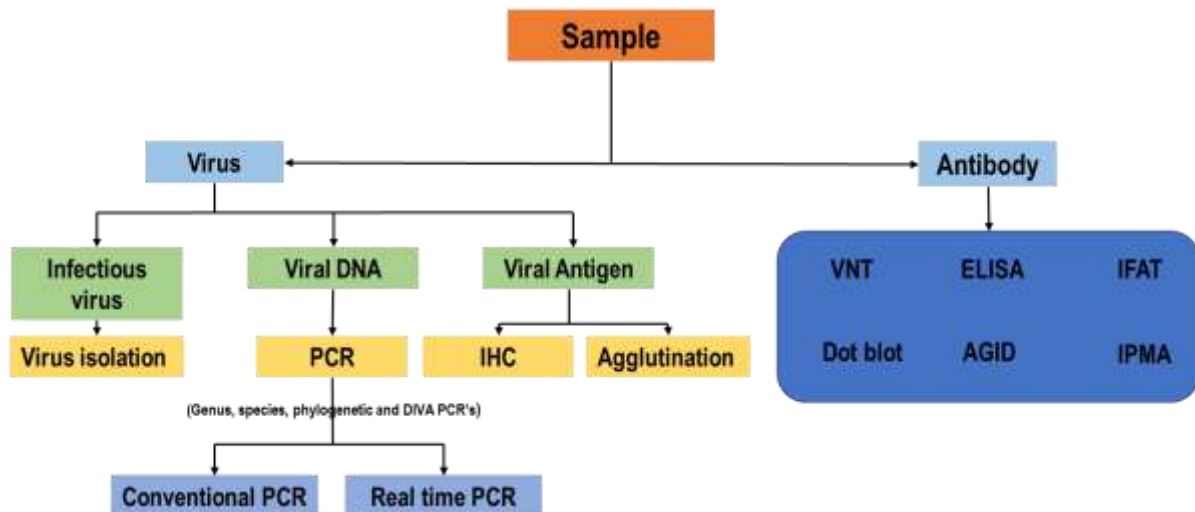
Young calves, lactating cows, and malnourished animals seem to develop more severe disease that may be due to an impaired humoral immunity. A lifelong cell- mediated immunity is developed in most animals that recover from clinical disease. Calves are born from the infected cow acquire maternal antibodies that may protect them from clinical diseases for approximately six months.

DIAGNOSIS

The early diagnosis of disease is very important for initiating treatment, control and preventive measures. The early and mid stage of LSD is difficult to diagnose by clinical sings. However, the late stage of disease is comparatively easy for clinician to diagnose at field level. Hence, quick and early laboratory diagnosis is very important. The details of suitable samples for laboratory diagnosis are provided in table depending on stage of disease. There are different testes used for virus/antigen and antibody detection in various laboratory depending on the availability of facilities (Table. 2).

Type of sample/ stage of disease	Early	Mid	Late	All the samples should be shifted to lab at the quickest time possible at 2-8° C
Swabs (Nasal/lacrima)	++++	+++	+	
Blood	+++	++	-	
Skin biopsy	+++	++++	+++	
Scab	+	+++	++++	
Serum	-	+	++	
Milk	-	+	++	
Rectal swabs/faecal	-	+	++	
Vectors	+	+	+	

Table.2: Details various clinical samples to be sent for laboratory diagnosis or confirmation in suspected cases of LSD.



GROSS AND HISTOPATHOLOGICAL FINDINGS

The postmortem examination of the dead animals will reveal grossly the skin nodular lesions all over the body depending on the severity of the disease. Internally, the respiratory system will show lesions on lungs typical gunshot wounds as observed in case of sheep and goat pox and even upper respiratory tract will show necrotic pock lesions (Fig 3). Liver may show necrotic foci and digestive system may show pock lesions very rarely. Histopathology of skin shows varied degree of mononuclear cell infiltration especially dendritic cells, lymphocytes and macrophages in dermis and hypodermis (Fig4, 6). Even though there are reports of presence of intracytoplasmic inclusion bodies in LSD affected organs, we did not observe any inclusions till date which is very peculiar finding especially with respect to LSDV Indian strains.



Fig 3: The lungs and trachea upper portion showing pock lesions.

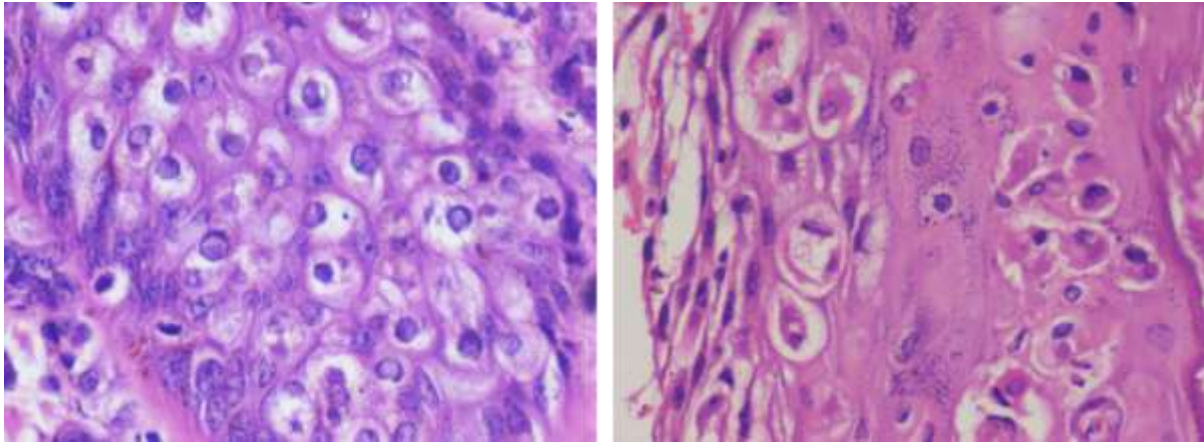


Fig 4: The epithelial cells showing hydropic degeneration. The epithelial cells showing intracytoplasmic inclusion bodies.

Virus isolation and characterization

Virus isolation from clinical sample is critical in the confirmation of disease. Virus can grow on number of primary cells or cell lines of bovine, ovine, or caprine origin. Chorioallantoic membrane of embryonated chicken eggs and African green monkey kidney (Vero) cells are the most commonly used to grow the virus. Virus grows slowly in cell cultures, and produce the cytopathic effect (CPE) after five to seven days after inoculation. LSDV induces a specific cytopathic effect (CPE) and intracytoplasmic inclusion bodies in cell culture.

Electron microscopy

Transmission electron microscopic (TEM) can be used for detection of virus directly from clinical sample. Diagnosis can be confirmed within a few hours of receipt of specimens. Mature capripox virions have an average size 320 x 260 nm and are a more oval profile and larger lateral bodies than orthopox virions.

Molecular detection methods

With advancement of molecular techniques like PCR, diagnostic of LSD become easy and specific. The molecular testing is critical for monitoring the spread of these viruses and controlling disease outbreaks. LSD virus confirmation may be done utilizing a Capri poxvirus specific PCR approach on conventional and real-time PCR technologies (Bowden et al., 2008). For differentiating virulent LSDV from the vaccine strain, Restriction Fragment Length Polymorphism (RFLP) has also been used. Molecular assays employing loop-mediated isothermal amplification to identify capripoxvirus genomes have been shown to have

sensitivity and specificity comparable to real-time PCR, with a simpler approach and a cheaper cost.

Serological tests

The only serologically approved test available is the Virus neutralization test (VNT) for monitoring of LSDV antibodies. Besides Indirect fluorescent antibody testing (IFAT), viral neutralization, enzyme-linked immunosorbent assays (ELISA), and immunological blotting (Western blotting) are used in different research laboratory. Neutralizing antibodies occur 3-4 days after the onset of clinical symptoms and reach maximal titer levels in 2-3 weeks. The agar gel immune diffusion test (AGID) and IFAT are less specific than VNT due to cross-reactivity with antibodies to other poxviruses. Western blotting is sensitive and specific, but it is difficult and expensive to perform.

Differential diagnosis

The differential diagnosis of LSD includes pseudo-LSD, dermatophilosis, dermatophytosis, bovine farcy, photosensitisation, actinomycosis, actinobacillosis, urticaria, insect bites, besnoitiosis, nocardiasis, demodicosis, onchocerciasis, pseudo-cowpox, and cowpox, foot and mouth disease, bovine viral diarrhoea, malignant catarrhal fever, infectious bovine rhinotracheitis, and bovine papular stomatitis. In case, if homologues vaccination is practiced the vaccine induced mild disease should be considered for differential diagnosis.

Treatment

There is no specific treatment to treat Lumpy Skin Disease. The treatment is by means of supportive care. The following protocols may be followed in case of LSD outbreak.

1. Separation and isolation of suspected/affected animals.
2. Symptomatic treatment of affected animals based the experience and availability of medicine in the locality.
3. Feeding management: Give easily digestible preferably liquid feed/food and succulent green fodder.
4. Proven or indigenous technology knowledge based herbal preparations may be used for wound and dermatitis treatment, as immunomodulators and antioxidants and fly repellents.

Immunization

As the LSDV is quite stable and can remain for months, a long-term vaccination regimen should be developed and made mandatory to control LSD covering the entire bovine population. A live attenuated vaccine has been developed and used in endemic country to control the disease. Members of the capripoxvirus are known to provide cross-protection. Hence, homologous (Neethling LSDV strain) and heterologous (sheeppox or goatpox virus) live attenuated vaccines can all be used to protect cattle (OIE, 2013). Most of the vaccine manufacturer in African countries use Neethling strain for virus for production LSD vaccine. This LSD vaccine is attenuated by culturing sixty times in lamb kidney cells followed by twenty times sub-culturing in chorio-allantoic membrane of embryonated chicken eggs. Attenuated virus provides immunity for three years after administration in bovines. Dose of vaccine varies from manufacturer to manufactures generally from $10^{2.5}$ to 10^4 TCID₅₀/dose. Attenuated LSDV vaccinations may produce minor adverse reaction known as the "Neethling response. Besides use of capripox virus vaccines in bovines in also recommended due to cross protective nature of vaccine. SPPV vaccines at a higher dose (three, five and ten-fold) have been used in cattle against LSDV in those regions where LSD and SPP are both present. Kenyan or Romanian sheep pox virus strains have been used to confer LSD immunity in bovines. These live vaccines could potentially serve as a source of infection for the susceptible sheep and goat populations, hence they are not recommended in nations free of sheep and goat pox. In India as per the guideline of Department of Animal Husbandry and Dairying, Ministry of Fisheries, Animal Husbandry and Dairying goat pox vaccine (Uttarkashi strain) is used @ $10^{3.0}$ TCID per animal subcutaneously. Recently India has also developed the LSD vaccine from local strain named Lumpi-ProVac^{Ind}. The vaccination plan should be chosen depending on the needs of the country and the resources that are available. Animals that have just been introduced to a farm should be immunised. Similar to this, calves from cows who have received a vaccination or have recovered from a natural infection need to receive one between the ages of three and four months. Vaccinations can be administered annually to all susceptible cattle and buffaloes especially in high risk groups and areas.

Prevention and control

Like several other viral diseases of livestock, the LSDV can be effectively controlled by implanting the following measures.

1. Animal movement restriction: the animal movement in or out from infective zone should be banned. If possible, the live animal markets, fairs, gatherings, migration etc., should be stopped.
2. Preventive vaccination: The ring vaccination (5km radius) should be carried in all (cattle and buffaloes) healthy animals which are aged above 4 months as per the GoI guidelines with goat pox vaccine ($10^{3.0}$ TCID₅₀).
3. Bio-security measures: The bio-security measures like quarantine, isolation, disinfection of premises, deworming, disposal of farm waste and carcasses, people movement, insemination, health records etc., should be regularly practiced/monitored.
4. Vector Control: The measures should be adopted for control of biting or blood feeding vectors through application of insecticide or repellents and also through reducing the breeding grounds for these vectors in farm premises.
5. Cleaning and disinfection: The affected areas including the farm equipments and fomites should be cleaned then disinfected with appropriate chemicals / disinfectants like ether (20 %), chloroform, formalin (1 %), phenol (2 % for 15 minutes), sodium hypochlorite (2-3 %), iodine compounds (1:33 dilution) and quaternary ammonium compounds (0.5 %).
6. Creating awareness among different stake holders through radio, TV, press releases, posters, newspapers / print media, digital media, social media posts, webinars, workshops etc.,
7. Prepare SOPs at state/regional level
8. Prepare and train the rapid response teams
9. Keep networking with line departments for better and quick response.
10. Monitoring and surveillance especially in unaffected areas.

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Chapter 12

An Overview of Protozoan Diseases of Domestic Animals in India

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Domestic animals in India often harbour a variety of pathogens including parasites, which account for a considerable economic loss to livestock farming due to severe morbidity and occasional mortality. Indigenous animals are less affected because of multiple reasons. Apart from breed resistance, strain variation of the parasite, immune and physiological status of the animals, co-infections with other pathogens determine the outcome of an infection. Many individual hosts are infected with multiple parasite species, and this may increase or decrease the pathogenicity of the infections. This phenomenon is termed heterologous reactivity and is potentially an important determinant of both patterns of morbidity and mortality. Of late, with the help of mathematical modeling, scientists found out that concurrent co-infection with less pathogenic species of *Theileria* (*T. mutans* & *T. velifera*) resulted in an 89% reduction in mortality associated with *T. parva* infection. Conversely, concurrent infection with *Babesia*, *Anaplasma*, *Theileria* and *Trypanosoma* in any combination or in combination with concurrent bacterial, viral or rickettsial diseases will certainly be more fatal. Hence, one should be vigilant to check for concurrent infection(s) and treatment for all individual pathogens should be taken into consideration. The following description on common protozoan diseases, commonly encountered in domestic animals India, is a brief account, which might be useful to field veterinarians and students of parasitology.

TRYPANOSOMOSIS

Only two species, viz., *Trypanosoma evansi* and *T. theileri* are known to occur in livestock in India, the former being of considerable economic importance. The disease caused by *T. evansi* is fairly widespread in India and is called 'Surra' (rotten). It is transmitted through hematophagous flies, viz., tabanids and *Stomoxys* spp., which act as mechanical vectors. This extracellular parasite is found in the blood plasma (and may also be found in lymph) and chiefly affects horse, mule, donkey, camel, cattle, buffalo, sheep, goat, pig, dog, cat, elephant and a number of wild animals. Laboratory animals (rat, mice, guinea pig and rabbit) are also susceptible. The most important clinicopathological finding in surra is

progressive anaemia, which is multifactorial. Muscle fibre degeneration, oedema and degeneration of endocrine glands also occur. In advanced cases the parasite may invade central nervous system. Thus, chronically infected animals suffer from a multisystemic disease. In camel, the disease is more chronic ('Tei-barsa' which means three year duration) than in horses and comparatively a few parasites may be found in peripheral circulation. However, a fairly high mortality occurs in camels if left untreated. In dogs also the disease is severe. Though surra is common in cattle, water buffalo and pig, these animals show a low parasitaemia devoid of clinical signs with a few fatalities. These animals can, therefore, serve as important 'carriers' for other susceptible livestock.

The pathological changes include enlargement of spleen, kidney, liver and lymphatic tissues, marked anaemia, emaciation, leukocytic infiltration of parenchyma with some necrosis or with petechial haemorrhages.

CLINICAL SIGNS

In horses, there is oedematous swelling along the belly, intermittent fever (lasting for 3-4 days) with high parasitaemia, anaemia, ecchymotichaemorrhage in the eyes, nostrils and anus, urticaria, muscular weakness resulting in reluctance to walk and unsteady gait. The urine becomes viscid and dark yellow. The disease in cattle and buffalo may be peracute to chronic. Peracute cases are characterized by high fever, dullness, sonorous breathing, salivation and abdominal pain. Reduced body weight, milk yield and poor reproductive performance is seen in chronically affected animals. Surra affected dogs may show corneal opacity and change in voice. Affected camels become progressively emaciated and the hump disappears. The disease in elephants is similar to camel surra.

Diagnosis: Conventional parasitological examinations are dependent on identification of the parasite in wet blood films as well as thick and thin blood smears and lymph node aspirate stained with Romanowsky stain. This method is 100% specific but poor in sensitivity. In cases of chronic and subacute trypanosomosis where parasite cannot be demonstrated, concentration techniques are used. In chronic or cryptic cases other methods like animal inoculation tests are used. Blood (0.25 ml) from the suspected animal is injected intraperitoneally into a suitable laboratory host (mouse or rat). The blood of the experimentally infected laboratory animal is examined starting 3 days post inoculation, and it is continued for 3-4 weeks if necessary, for the presence of trypanosomes.

The serology based tests are important for study of epidemiological surveys. The serological tests that are being widely used include, card agglutination trypanosomiasis test (CATT), indirect fluorescent antibody test (IFAT), enzyme linked immunosorbent assay

(ELISA) and latex agglutination test (LAT).

Polymerase chain reaction (PCR), based tests detect the trypanosome DNA in the blood. A number of sensitive primers targeting the internal transcribed spacer 1 (ITS1), nuclear repetitive sequences, kinetoplast DNA and RoTat 1.2 have been found to be highly specific.

TREATMENT: The following drugs are used for the treatment of surra in animals.

Sl. No.	Compound	Dose	Route	Remarks
1.	Suramin	7-10 mg/ kg bw in horse 10 mg/kg bw in cattle	IV	Administered in 3 divided doses at weekly intervals for 3 wks
2.	Quinapyramine sulphate or Quinapyramineprosolt	3-5 mg/kg bw	SC	Horses and dogs may suffer from local or systemic reaction
3.	Homidium chloride or Homidium bromide	1 mg/kg bw (2.5% solution)	IM	Protective effect upto 5 wks
4.	Isometamidium chloride	0.5-2 mg/kg bw	IM	Protection for 3-6 months
5.	Diminazine aceturate	3.5-7.0 mg/kg bw	IM	Administered in 2-3 divided doses at 4 hr intervals, toxic to horses, dogs and camels.

Control: Since the disease is mechanically transmitted by haematophagous arthropods (especially the tabanids), control of flies during monsoon season would reduce the incidence of infection. The tabanids are most active in sunny weather. Animals can be protected in endemic areas by chemoprophylaxis using antrycideprosolt and isometamidium during peak fly season.

BABESIOSIS

Nearly 16 to 18 species of *Babesia* are found in domestic animals. The disease is also known as 'piroplasmosis'. All species of *Babesia* have more or less similar pathogenesis.

This is mainly due to massive destruction of host's erythrocytes not only by invading merozoites, but also due to release of pharmacologically active substances and immunologically mediated erythrophagocytosis of infected and uninfected erythrocytes. Some species of *Babesia* (*B. bovis* and *B. canis*) clog the brain capillaries, which is invariably fatal. This condition is known 'cerebral babesiosis'.

BOVINE BABESIOSIS

In India, the disease is exclusively caused by *Babesia bigemina*. Inverse age resistance is characteristic of *B. bigemina*. The disease is transmitted through several species of ticks with *Rhipicephalus (Boophilus) microplus* being the main vector. Transmission is mainly transovarian since one host ticks serve as vectors. *Babesia* sp. can also be transmitted between animals by direct inoculation of blood, although this method of transmission is thought to be of minor importance. Recently, few cases of a small form of *Babesia* have been reported from cattle. Genetic characterization of these isolates revealed closeness with *B. bovis*. Bubaline babesiosis is also not uncommon in India. It is axiomatic to consider bubaline *Babesia* species as *B. bigemina*. However, our preliminary study on genetic characterization of these isolates indicated the existence of *B. orientalis* like genotype in our country.

Clinical Signs: The disease is characterized by high fever, haemoglobineuria and severe anaemia. The severity of anaemia depends upon the severity of infection. The liver, spleen and kidneys, may also be involved. There is severe splenomegaly and jaundice. Antigen-antibody complex mediated hypersensitivity leads to glomerulonephritis. The released haemoglobin is excreted with the urine which becomes red to almost black in colour. Accordingly, the disease is also called 'red water fever'.

Diagnosis: It is based on clinical signs and confirmation is done by demonstration of the parasite in stained blood smear. The disease may be confused with leptospirosis, anaplasmosis or cowdriosis.

Treatment: The following drugs are highly effective against bovine babesiosis.

1. Berenil, 3-5 mg/kg by deep intramuscular injection.
4. Quinuronium sulfate 1-2 mg/kg by intramuscular or subcutaneous injection.
5. Imidocarb dipropionate (imizole) as subcutaneous or intramuscular injection @ 1-3 mg/kg.

Control: Effective tick control is strongly recommended. No vaccine is available in India. Serially passaged attenuated organisms are used as vaccine in some developed countries.

EQUINE BABESIOSIS

B. caballi and *B. equi* (*Theileria equi*), infect equines. The vector in India is probably *Hyalomma* sp. *B. equi* is much smaller than *B. caballi* and is more pathogenic. It characteristically divides into four daughter organisms which form a 'Maltese cross'. It causes fever, anaemia, anorexia, jaundice, depression, enlargement of liver and spleen. Oedema of the head, limbs and ascites occur but haemoglobinuria is not a constant feature. In acute form, the disease is fatal within 2 weeks.

Diagnosis: It is based on clinical signs and demonstration of the parasite in blood, especially during high fever. Serological tests like complement fixation (CFT), precipitation, passive haemagglutination and indirect fluorescent antibody (IFAT) can be used.

Treatment: For *B. equi*, imidocarb dipropionate (4 mg/kg given intramuscularly or subcutaneously four times at 72 hr intervals) and diminazene aceturate (6-12 mg/kg given twice at 24 hr interval) are effective.

OVINE AND CAPRINE BABESIOSIS

Four species, viz. *B. motasi*, *B. ovis*, *B. foliata* and *B. taylori* have been reported from sheep and goat. *B. motasi* is a large form, resembling *B. bigemina* and may cause acute or chronic form of disease. The parasite is transmitted through various ticks belonging to *Dermacentor*, *Haemaphysalis* and *Rhipicephalus*. The parasite causes high fever, haemoglobinuria, anaemia and prostration, often resulting in death. The chronic type of infection does not show any characteristic signs. *B. ovis* is smaller than *B. motasi* and also less pathogenic. In acute stage it may, however, cause fever, haemoglobinuria and anaemia. Not much is known about the remaining two species, which are evidently not pathogenic.

Treatment: The drugs mentioned under treatment of bovine babesiosis are also effective in ovine and caprine babesiosis.

CANINE BABESIOSIS

At least six species of *Babesia*, viz. *B. canis*, *B. vogeli*, *B. rossi*, *B. gibsoni*, *B. vulpes* and *B. conradae* infect dogs worldwide. The formerly mentioned three species are large forms while the rest three are small forms of *Babesia* of dogs. *B. vogeli* is the only large form reported from India and is less pathogenic than *B. canis* and *B. rossi*. It is a large piroplasm,

often several of them are found in an erythrocyte and in endothelial cells of lungs and liver. Both pups and adult dogs may show clinical signs of the disease, which is commonly called 'tick fever' or 'biliary fever'. The clinical signs are fever, listlessness, anaemia, jaundice, debility, anorexia, etc. In the later stages, there may be a drop in temperature and the animal may suddenly collapse and die. The diagnosis is made by making a blood film, especially from the ear vein. *B. gibsoni* is the small form and usually associated with chronic infections. It produces fever, anaemia, weight loss, thrombocytopenia and marked enlargement of liver and spleen.

THEILERIOSIS

Bovine tropical theileriosis in India is caused by *T. annulata*. The parasite occurs as round, ovoid, rod, dots or comma-shaped piroplasms within erythrocytes and are found as intracellular parasites in lymphocytes (and other cells of reticulo-endothelial system). *Theileria* need two obligatory hosts for the completion of life-cycle. In vertebrate host, the parasites multiply asexually by schizogony in lymphocytes and by binary fission in erythrocytes. The invertebrate host (vector) is a hard-tick (*Hyalomma* spp.) Transmission of the parasites by the tick vector is through stage-to-stage/transstadial transmission (i.e. if larva picks up infection nymph transmits and if nymphs pick the infection then adult transmits it to a new host). During the last decade atypical cases of theileriosis in cattle and buffaloes and existence of *Theileria buffeli/orientalis* appeared in literature. There is controversy over the pathogenicity of this apparently mildly pathogenic species. More concerted efforts in the form of dedicated research are required to establish the fact.

CLINICAL SIGNS: The disease is characterized by high rise in body temperature followed by lacrimation, nasal discharge, cough, depression, anaemia, icterus, swelling of superficial lymph nodes, spleen and liver and tumefaction of eyelids. Haemoglobinuria may occur. In affected animals, the gums, oral cavity, abomasum and intestine may get inflamed accompanied by diarrhoea. Punched out 'button-shaped' or 'cigarette-burn' ulcers, 2-12 mm in diameter, in the abomasums, intestine and lymph nodes are the pathognomonic lesions of the disease. This destruction of connective tissue could be due to digestion of the extracellular matrix by MMPs secreted by macroschizont-infected cells known to be present in these lesions. Bovine leukocytes are induced to express eight novel metalloproteinase activities. These are the virulence factors and responsible for the

pathology. Immunologically mediated pneumonia and lung oedema are the immediate causes of death in severely affected animals. The indigenous breeds do not suffer much possibly because of innate immunity and preimmunity, though calves below three months of age occasionally succumb to the infection.

Diagnosis: It is made by finding the intraerythrocytic piroplasms in Romanowsky stained blood smears and lymphocytic stages (Koch's blue bodies) in aspirate obtained from the enlarged superficial lymph nodes. Schizonts are also found in mononuclear cells in peripheral circulation. In *T. annulata* about 80% of intraerythrocytic forms are ring-shaped or annular. Very low levels of infection can be detected unequivocally by PCR.

Treatment: Buparvaquone (@ 2.5 mg/kg, administered intramuscular) is the drug of choice. It may be repeated, if need be, after 48 hours in severe cases. Resistance against buparvaquone has been reported from many countries.

Control: Currently control of theileriosis is based upon regular tick control in endemic areas and vaccination with cell culture attenuated schizont-infected lymphoid cells. Approximately, 2×10^6 schizont-infected lymphoblasts are inoculated as vaccine.

OVINE AND CAPRINE THEILERIOSIS

Among *Theileria* species affecting small ruminants, *Theileria lestoquardi* causes malignant ovine theileriosis, characterized by fever, cessation of rumination, swelling of superficial lymph nodes, cardiovascular problems, diarrhoea, jaundice and haemorrhages, and mortality rates of 46–100%. *T. uilenbergi* and *T. luwenshuni* are also considered highly pathogenic. Diagnosis, treatment and control are the same as in *T. annulata*.

COCCIDIOSIS

Cattle: Bovine coccidiosis is an acute or chronic disease characterized by haemorrhagic or mucoid diarrhoea in young calves up to 3 months of age. Adult cattle act as carriers and continue to shed oocysts without showing any clinical signs. Out of 21 species infecting cattle, *E. bovis* and *E. zuernii* are the most pathogenic and cause haemorrhagic diarrhoea. Severe changes are seen in caeca and colon which become congested and oedematous. In acute form of disease (associated with *E. zuernii*), there is bloody diarrhoea, hence also known as 'red-dysentery'. The affected animals go off food, become dehydrated, emaciated and die within a week of the appearance of clinical signs. In heavy infection, *E. ellipsoidalis*, *E. auburnensis* and *E. alabamensis* may also become pathogenic and cause mucous diarrhoea.

Buffalo: Buffaloes share almost all the species of *Eimeria* found in cattle. Additionally, they also harbour *E. bareillyi*, which is moderately pathogenic. Like cattle, *E. bovis* and *E. zuerni* are the most pathogenic species in buffaloes and the course of disease is also similar.

Diagnosis of coccidiosis in calves is made by seeing the clinical signs and by demonstration of oocysts in faeces. It should be differentiated from other diarrhoea causing agents (Rotavirus, Coronavirus, colibacillosis etc.).

Some of the commonly used anticoccidials are Sulfaguanidine @ 125 mg/kg bw (33% solution) x 13 days (on alternate days), Amprolium @ 20-25 mg/kg bw for 4-5 days (therapeutic), Toltrazuril @ 20 mg/kg bw (single dose). Rehydration is recommended in severely dehydrated calves. Coccidiosis is a disease of organised farms, where overcrowding in calf pens occurs and hygienic measures are not followed. Avoidance of housing adult animals with calves must be avoided.

CRYPTOSPORIDIOSIS

Like intestinal coccidia, *Cryptosporidium* spp. also infect the intestinal epithelial cells. Cattle and buffaloes are the natural host of at least 4 species, out of which *C. parvum* causes one of the most severe diseases of young ruminants, particularly neonatal calves. Infected animals may suffer from profuse watery diarrhoea, dehydration and in severe cases death can occur. Calves which fail to get sufficient amount of colostrums are more susceptible to infection. Infection can be detected as early as 5 days of age, with the greatest proportion of calves excreting organisms between days 9 and 14. Many reports associate infection in calves with diarrhea occurring at 5–15 days of age. Newborn lambs and kids also suffer from cryptosporidiosis. *C. parvum* is of zoonotic importance and can cause life threatening disease in immunocompromised humans.

As the oocysts are very small (4.5 to 7.0 µm), it is difficult to demonstrate in routine faecal smears. The oocysts can easily be demonstrated in faecal smears stained with modified Ziehl-Neelsen acid fast staining. No effective treatment is available.

Chapter 13

Viral diseases of bovines with special reference to Infectious Bovine Rhinotracheitis (IBR)

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Bovine herpes virus (BoHV-1), a member of the family Herpesviridae, causes diverse ailments, viz. infectious bovine rhinotracheitis (IBR), infectious pustular vulvovaginitis (IPV), infectious balanoposthitis, keratoconjunctivitis and neurological disorders. Though mucous membranes of either upper respiratory or genital tract are the most common routes of entry of BoHV-1, transmission by conjunctival and aerosol routes were also reported. During primary infection, the virus is transported along axons where it becomes latent and persists for life in sacral and trigeminal ganglions. The virus gets reactivated under stress conditions making the animal a potential shedder of virus. Hage et al. (1996) in their study on population dynamics of BoHV-1 defined the reproduction ratio of BoHV-1 to be at a minimum of 7. This study described the virus transmission and seroprevalence of BoHV-1 in breeding cattle, under different housing, feeding and watering practices.

Epidemiology BoAHV-1 is known to predominantly infect epithelial cells in the upper respiratory airway and genital tract mucosa. Infections are common in animals over six months of age, which is most likely due to a decline in passive immunity acquired through the colostrum and possible viral exposure in older animals. BoAHV-1 has a restricted epidemiology primarily affecting cattle, but can also infect goats, sheep and buffaloes. Abortions and genital infections are quite common, and they can happen at the same time as an acute respiratory infection due to reactivation from latency. In BoAHV-1-positive animals the abortion rate may be increased anytime during the second half of gestation. BoAHV-1 can bind to the human poliovirus receptor or weakly to human vascular endothelial cell (HveC) (nectin-1) in vitro, without detectable viral replication; human infection with BoAHV-1 has not been reported yet.

BoAHV-1 can be transmitted between cattle and goats, with a seropositivity rate of 25.5% in cattle and 20.9% in goats. Although cattle remain the leading source of BoAHV-1 infections, serological studies showed higher infection rates in goats, which rank second as a source of BoAHV-1, while sheep have no role in transmitting BoAHV-1. Cross-infection experiments

between Caprine Herpesvirus 1 (CapHV1) in calves and BoAHV-1 in goats revealed that goats shed higher BoAHV-1 during primary infection and experimental reactivation. These results also suggested goats as a prospective BoAHV-1 reservoir as they are kept in close contact with bovines; goats are therefore recommended to be considered during IBR eradication programs. A serological study in northern Turkey using a BoHV-1 gB-blocking enzyme-linked immunosorbent assay (ELISA) also found CapHV-1 and BoHV-1 in circulation among local goats with a seropositivity of 19.33%, backing up the theory that goats play a substantial role in the transmission of BoAHV-1. There are very few studies exploring the transmission of BoAHV-1 in sheep. Hage et al. concluded that BoAHV-1 infection in sheep is probable, but that it does not spread easily to cattle. Although all sheep exhibited nasal discharge and seroconversion, none of them produced antibodies against BoAHV-1 estimating the basic reproduction ratio (R₀) of BoAHV-1 between sheep and calves at 0.1 and among calves at ≥ 9 (1997). Furthermore, one study showed experimental transmission of BoAHV-1 in buffaloes, establishing buffaloes as reservoir of BoAHV-1. The study evaluated serum antibody responses using commercial IBR gB and gE ELISA kits, as well as virus neutralization, and showed viral DNA in the trigeminal ganglia. Similar to sheep very little information exists on BoAHV-1 transmission in buffaloes; however, studies evaluating seropositivity against BoAHV-1 in water buffaloes and Mediterranean buffaloes have been documented; nevertheless, naturally infected buffaloes with BoAHV-1 have not been reported yet.

BoAHV-1 establishes latency in the trigeminal ganglia through cell to cell spread, with intermittent reactivation, which is critical for the maintenance of the virus in cattle. During reactivation the virus gets transported to the primary site of infection, followed by the initiation of the lytic replication cycle, thereby producing infectious virions. Reactivation may occur when the animals are under stress, for example during transport, and can be induced by compounds such as dexamethasone. The latency-related (LR) and ORF-E genes, which are highly expressed, regulate the maintenance of the reactivation cycle. A hallmark of reactivation from latency is the suppression of LR gene expression and the enhanced viral gene expression in the neurons. The viral regulatory protein bICP0 is also involved in reactivation. Moreover, it should also be considered that the commercially available BoAHV-1 ELISA tests show serological cross-reactivity against BoAHV-2 strains. Due to this issue, animals could be incorrectly deemed positive for BoHV-1 thereby causing an impact on control and eradication programs, both economically and socially. A study in

Italy recently demonstrated the occurrence of cross-reactivity against BoAHV-2 and BoAHV-1 in a calf detained at a performance station.

Differential diagnoses in individual cattle

- Pneumonia
- Malignant catarrhal fever
- Mucosal disease (BVD)

Differential Diagnosis in a group of cattle

- Bluetongue.
- Foot and mouth disease

Other causes of respiratory disease in cattle (bovine parainfluenza virus, bovine respiratory syncytial virus, bovine coronavirus, bacterial pneumonia, lungworm)

Genetically engineered vaccines Gene-deleted vaccines Gene-deleted vaccines have a number of advantages over conventional vaccines. Due to well defined deletions, they can be safer and serve as marker vaccines, allowing serologic distinction between infected and vaccinated animals (DIVA). Live gene-deleted vaccines, like MLV, generally elicit strong balanced immune responses, and when delivered IN, mucosal immunity. Several mutant viruses lacking non-essential genes (gG, gE, gI or gE/gI) were significantly less virulent in cattle. The gC-, gE- and gG- mutants elicited better protection than the gI- and gE-/gI- mutants. Since gE induces a robust antibody response in infected cattle, and is antigenically quite stable, the gE- mutant was further developed and commercialized as a DIVA vaccine. A companion serologic competitive ELISA differentiates gE- mutant vaccinated animals from infected ones. A study in three European countries showed that the gE-deletion mutant reduced the rate of Wt BoAHV-1 seroconversion in vaccinated animals, and this vaccine is still used in European countries with BoAHV-1 infections. The gE-deletion mutant induces protection as either MLV or KV vaccine; however, while earlier studies showed that the live gE- mutant is unlikely to be re-activated, subsequent studies suggested that it can cause latency. Furthermore, a potential risk of virulence recovery due to recombination of a gE- mutant with Wt virus was reported by . However, when the gE- mutant was administered IM instead of IN, the vaccine virus was not excreted. To reduce the chance of recombination with Wt virus, a gE-TK-deletion mutant was generated, which was protective in calves after two IM vaccinations; when the MLV was combined with an adjuvant, Polygen, no virus-induced nasal shedding was detected, and almost

no DNA in the trigeminal ganglia, suggesting reduced establishment of latency after Wt BoAHV-1 challenge. A triple deletion mutant BoAHV-1, with deletions in the UL49.5 luminal domain (residues 30-23), the cytoplasmic tail (residues 80-96) of gE and the entire US9, was also generated. The triple deletion mutant and the gE- mutant were both protective against BoAHV-1 infection, but the animals vaccinated with the triple deletion mutant cleared the virus three days earlier. Recently, two inactivated gE-deleted marker vaccines were reported to be safe and to produce strong humoral immune responses in pregnant cows until calving. In the calves passive immunity persisted until 180 days after birth.

Two types of marker vaccines (gE- and gE-TK-) are commercially available. A recent comparison of live BoAHV-1 marker vaccines (gE- IM, gE- IN and gE-TK- IM) showed protection against Wt BoAHV-1- induced disease, but not against Wt BoAHV-1-induced latency reactivation suggesting that new vaccines that protect from Wt BoAHV-1-induced latency, and don't become latent themselves, still need to be developed. A triple gE-gG-TK- mutant was generated and compared to a double gGTK- mutant developed previously. The triple mutant exhibited slower growth kinetics in vitro and replication in vivo in cattle compared to the double mutant and Wt BoAHV-1. No reactivation from latency was detected after dexamethasone treatment, and the triple gE-gG-TK- mutant elicited a protection rate of 64.2% after BoAHV-1 challenge in cattle.

Subunit vaccines Subunit vaccines, consisting of one or more protective antigens, have a number of advantages over conventional vaccines. When formulated with an effective adjuvant, a subunit vaccine induces strong humoral immune responses, and if the adjuvant contains a component promoting cross-presentation, may also elicit cell-mediated immune responses. In addition, subunit vaccines have a good safety profile, and can also serve as marker vaccine. The three major glycoproteins, gB, gC and gD, when formulated with an effective adjuvant, all induce protection from BoAHV-1 challenge in cattle. Glycoprotein D, being most effective, was further evaluated in combination with different adjuvants, and shown to elicit balanced, protective immune responses when formulated with Emulsigen and CpG ODN, a TLR9 ligand. With gD as the immunogen in the subunit vaccine, in addition to the cgE ELISA, the more sensitive cgB ELISA could be used to differentiate infected from vaccinated animals. However, although as little as 7 ug of gD, when combined with an effective adjuvant, can be sufficient to elicit full protein from BoAHV-1 challenge, one of the major drawbacks for veterinary use is the relatively high expense.

CONCLUSION

Overall, MLV and KV BoAHV-1 vaccines are still used most frequently in most parts of the world, whereas marker vaccines are in use in Europe. While studies on the basic biology have led to novel, more defined vaccines, such as gene-deleted DIVA vaccines, and vectored and subunit vaccine candidates, for BoAHV-1 only the new gene-deleted marker vaccines have been commercialized. Most recently, efforts have concentrated on generating new, safer gene-deleted BoAHV-1 vaccines, that are not reactivated when used as live vaccine.

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Chapter 14:

Meat-borne Zoonotic Diseases: An Overview

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INTRODUCTION

Humans are considered omnivores, having consumed meat for about 2.6 million years, making it a prominent protein source in the average diet. Since 1961, global meat production has quadrupled, with pork being the most consumed meat, although poultry is rapidly expanding. Meat consumption varies widely among countries due to economic, cultural, and other factors. Currently, nearly 340 million metric tons of meat are produced annually, three times more than 50 years ago. Meat-borne zoonotic diseases are infections naturally transmitted between vertebrate animals and humans through the consumption or handling of contaminated meat. These zoonoses, such as bacterial infections (salmonellosis, campylobacteriosis, listeriosis), viral infections (hepatitis E, avian influenza), and parasitic infections (toxoplasmosis, trichinosis), pose significant public health risks, leading to illnesses, hospitalizations, and even fatalities. Additionally, outbreaks of these diseases can cause substantial economic losses in the farming sector and food industry, highlighting their profound impact on both human health and the economy. Among the numerous microbes interacting with animals, some pathogens can become zoonotic, posing significant threats to public health and the economy. Animal-derived foods like milk, meat, and eggs are essential to human nutrition, but when contaminated with pathogenic microbes, they can pose serious health risks. These diseases can range from mild self-limiting diarrhoea to fatal conditions. It is estimated that contaminated food causes illness in 1 out of 10 people, with children under five accounting for 40% of cases, thus burdening healthcare systems and hindering socioeconomic development.

MEAT: FROM FARM TO FORK

If we examine the journey of food of animal origin, we observe a complex interplay of priorities at each stage. The primary producer focuses on the production and profitability of their livestock, ensuring the animals are raised efficiently to maximize returns. Veterinarians prioritize the health and well-being of the livestock, aiming to prevent and treat diseases.

Transporters are mainly concerned with reducing mortality during transport, striving to deliver animals safely to their next destination. Slaughterers and abattoirs emphasize high throughput and operational efficiency to process animals quickly and effectively. Processors focus on minimizing production costs while increasing market share, sales, and consumer attention through various strategies. Retail sellers are driven by the need to boost sales and profit margins. Finally, consumers are interested in the cost, taste, nutritional facts, and, to a lesser extent, the safety of the food they purchase. This intricate chain reflects the diverse concerns and objectives of each stakeholder involved in the journey of food of animal origin.

SOURCES OF PATHOGEN CONTAMINATION IN MEAT

Pathogens contaminate meat through various avenues. Animal carriage involves pathogens carried by animals, such as in their gut or on their skin, which can contaminate meat during the slaughter and processing stages, often through viscera spills. The processing environment also plays a role, as contaminated processing equipment, surfaces, or water can introduce pathogens to meat through cross-contamination. Additionally, human handling contributes to contamination risks, with improper hygiene practices such as unwashed hands or cross-contact transferring pathogens to meat. These factors collectively highlight critical points where contamination can occur, underscoring the need for stringent hygiene and handling practices throughout the meat production process.

CHARACTERISTICS AND RISKS OF SMALLHOLDER LIVESTOCK FARMS

Smallholder livestock farms often exhibit several characteristics that contribute to increased disease risks. These farms typically have limited biosecurity measures, lacking restricted farm access, quarantine protocols, and adequate hygiene practices, which heightens the risk of disease introduction and spread. Due to limited land availability, small farms have high stocking densities, promoting disease transmission among animals due to close contact and increased stress. Inadequate housing conditions with limited ventilation, space, and sanitation further create a favourable environment for disease pathogens to thrive and spread. Additionally, smallholders may face challenges in accessing veterinary services, leading to delayed diagnosis, treatment, and prevention of diseases. Lack of regular vaccination programs due to cost constraints or awareness issues leaves animals vulnerable to vaccine-preventable diseases. Limited access to quality feed and nutrition weakens immune systems of animals, making them more susceptible to infections. Poor water quality, whether due to contamination

or scarcity, can contribute to the spread of waterborne diseases among livestock. Climate variability, extreme weather events, and environmental pollution also stress animals and increase their susceptibility to diseases. Moreover, small farms often lack systematic disease surveillance systems, resulting in underreporting and delayed response to disease outbreaks, further compounding the challenges faced in managing disease risks.

CHARACTERISTICS AND RISKS OF TRADITIONAL MARKETS

Traditional food markets, also known as wet markets for their use of water or ice, embody cultural heritage and community dynamics. They offer a diverse range of perishable goods and operate through decentralized open-air stalls with basic infrastructure. However, sanitary facilities like waste disposal are often lacking. In-situ animal slaughtering and trading of illegal wildlife add vibrancy but raise hygiene concerns. Stray animals and birds are common, and street food hygiene can be questionable. Yet, these markets are vital for fresh, affordable food and unique regional products, supporting livelihoods and serving economically disadvantaged communities. Trust-based transactions prevail, but issues like hygiene awareness, and limited regulation persist. Improved infrastructure, hygiene standards, and regulatory frameworks are needed for economic vitality and public health in these vibrant cultural hubs.

CHARACTERISTICS AND RISKS OF INFORMAL SLAUGHTERHOUSES

Informal slaughterhouses, characterized by makeshift structures and lacking proper amenities like flooring, drainage systems, and waste disposal mechanisms, present significant challenges to food safety and animal welfare. The absence of clean water for washing carcasses and inadequate sanitation practices contribute to contamination risks, while the lack of cold storage facilities compromises meat preservation and increases the likelihood of spoilage and foodborne illnesses. Manual labour without mechanization leads to inconsistent meat quality and handling risks, further raising concerns about the safety of meat produced in such environments. These issues are compounded by prevalent inhumane practices, often due to a lack of awareness and training in animal welfare standards. Animals are frequently handled and slaughtered inhumanely, prompting ethical concerns. The absence of veterinary inspection before or after slaughter elevates the risk of diseased meat entering the food supply, posing serious health hazards to consumers. Moreover, many informal slaughterhouses operate without the required licenses and permits, circumventing regulatory oversight and accountability. Non-compliance with standards and guidelines for slaughtering and meat

processing set by food safety authorities such as FSSAI exacerbates these risks, as does poor waste management practices leading to environmental pollution and contamination of soil and water sources. The operations of these slaughterhouses can also result in foul odours and attract pests, creating nuisances for nearby communities.

FACTORS FACILITATING ZONOTIC PATHOGENS IN MEAT

Meat serves as an optimal growth medium for various pathogens due to its nutrient-rich composition, providing an environment conducive to their growth and multiplication. Improper storage and handling practices, such as inadequate refrigeration and cross-contamination during storage and reheating, further facilitate pathogen proliferation in meat products. The food danger zone of 4°C to 60°C, where pathogens thrive, highlights the importance of maintaining proper refrigeration and avoiding frequent reheating, as this can lead to temperature fluctuations that promote bacterial growth. Additionally, unsanitary conditions prevalent in slaughterhouses can lead to contamination of meat with pathogens from animal intestines and other sources. Human consumption practices, particularly the consumption of undercooked or raw meat, directly expose individuals to zoonotic pathogens, underscoring the importance of maintaining kitchen hygiene standards to prevent cross-contamination and ensure thorough cooking. High-density animal farming practices increase stress and disease susceptibility among livestock, creating environments that promote pathogen multiplication and transmission. The inclusion of wild animals in the meat trade introduces new and often unknown zoonotic pathogens into the human food chain, adding to the complexity of food safety challenges. Weak food safety regulations and enforcement further contribute to increased contamination and transmission of pathogens through meat products.

OCCUPATIONAL HEALTH RISKS IN THE MEAT INDUSTRY

Slaughterhouse workers, meat handlers, and processors play vital roles in the meat industry, but their work puts them at significant occupational health risks due to close contact with live animals, carcasses, and raw meat. This proximity exposes them to a variety of zoonotic diseases, which are infections transmitted from animals to humans. These diseases pose substantial health risks to individuals working in these environments. Key diseases they face include:

- **Brucellosis:** Caused by *Brucella* species, transmitted through contact with infected animals or products, leading to symptoms like fever, joint pain, and fatigue.

- **Leptospirosis:** Caused by *leptospira* spp., contracted through contact with infected animals' urine, causing flu-like symptoms or severe disease with liver and kidney damage.
- **Q Fever:** Caused by *Coxiella burnetii*, contracted through inhalation of contaminated dust, with symptoms including high fever, severe headache, and pneumonia.
- **Anthrax:** Caused by *Bacillus anthracis*, transmitted through contact with infected animals or products, with varying symptoms depending on the route of infection.
- **Tuberculosis:** Caused by *Mycobacterium tuberculosis* complex, transmitted from cattle to humans, causing chronic lung disease.

CONSUMER HEALTH RISKS

Consumers face foodborne illnesses due to contamination in meat products, leading to gastrointestinal and systemic symptoms. Examples include:

- **Foodborne infection:** Caused by pathogens growing in the intestinal tract after consuming contaminated food. Examples include *Salmonella* and *Campylobacter jejuni*.
- **Foodborne intoxication (poisoning):** Resulting from pre-formed toxins in food produced by pathogens, such as *Clostridium botulinum* and *Staphylococcus aureus*.
- **Toxin-mediated infection:** Occurs when pathogenic bacteria in food produce toxins in the intestinal tract, causing illness, as seen with *E. coli* O157 and *Vibrio cholerae*.

SYMPTOMS OF MAJOR MEAT-BORNE GASTRIC ILLNESSES

Pathogen	Incubation Period	Symptoms
<i>Staphylococcus aureus</i> and its enterotoxins	1-7 h, mean 2-4 h	Nausea, vomiting, retching, diarrhea, abdominal pain, prostration
<i>Bacillus cereus</i> (emetictoxin)	8-16 h (2-4 h if emesis predominant)	Vomiting or diarrhea, depending on whether diarrhetic or emetic toxin present; <i>Bacillus cereus</i> (emetic toxin) abdominal cramps; nausea

Norovirus	12-48 h	Nausea, vomiting, watery non-bloody diarrhea, dehydration
<i>Clostridium perfringens</i>	2-36 h (mean 6-12 h)	Abdominal cramps, diarrhea, putrefactive diarrhea (<i>CL perfringens</i>), sometimes nausea and vomiting <i>Clostridium perfringens</i>
<i>Salmonella</i> spp., <i>Shigella</i> spp., <i>E. coli</i>	6-96 h (usually 1-3 days)	Fever, abdominal cramps, diarrhea, vomiting, headache
Enterohaemorrhagic <i>E. coli</i> , <i>Campylobacter</i> spp.	1-10 (median 3-4) days	Diarrhea (often bloody), abdominal pain, nausea, vomiting, malaise, fever (uncommon with <i>E. coli</i> 0157:H7)
Rotavirus, Astrovirus, enteric Adenovirus	3-5 days	Fever, vomiting, watery non-inflammatory diarrhea
<i>Yersinia enterocolitica</i>	3-7 days	Fever, diarrhea, abdominal pain
<i>Taeniasaginata</i> , <i>Taenia solium</i>	3-6 months	Nervousness, insomnia, hunger pains, anorexia, weight loss, abdominal pain, sometimes gastroenteritis
<i>Clostridium botulinum</i> and its neurotoxins	2 h to 6 days, usually 12-36 h	Vertigo, double or blurred vision, loss or light reflex, difficulty in swallowing, dry mouth, <i>Clostridium botulinum</i> and its neurotoxins weakness, respiratory paralysis
<i>Trichinella spiralis</i>	4-28 days	Gastroenteritis, fever, oedema around eyes, perspiration, muscular pain, chills, <i>Trichinella spires</i> prostration, laboured breathing
<i>Toxoplasma gondii</i>	10-13 days	Fever, headache, myalgia, rash
<i>Listeria monocytogenes</i> , <i>Campylobacter jejuni</i>	varying periods	Fever, chills, headache, arthralgia, prostration, malaise, swollen lymph nodes and other <i>Listeria monocytogenes</i> , <i>Campylobacter JeJuni</i>

VULNERABLE POPULATIONS TO MEAT-BORNE ZONOTIC DISEASES

Certain populations are particularly vulnerable to meat-borne zoonotic diseases. Children and infants, with their developing immune systems and limited resistance, are more susceptible. Elderly individuals, due to weakened immune responses, are also prone to infections. Immunocompromised individuals, including pregnant women, chemotherapy patients, and those with autoimmune disorders, are highly vulnerable. Frequent travelers, especially to regions with varying food safety standards, face a higher risk of exposure. Similarly, tourists and adventurers exploring exotic cuisines and local delicacies may encounter unfamiliar meats, increasing their exposure to zoonotic pathogens.

PREVENTING FOODBORNE ILLNESSES: COMMON MEAT HANDLING MISTAKES

Common mistakes made by meat consumers that can lead to foodborne illnesses include improper handling practices like not washing hands, utensils, and surfaces after handling raw meat, using plastic containers for washing and rinsing, and using the same knife for cutting raw meat and other foods without proper cleaning. Cross-contamination can occur when using the same cutting board or utensils for raw meat and other foods without washing in between. Incorrect storage methods, such as storing raw meat above ready-to-eat foods in the refrigerator, can lead to dripping and contamination. Undercooking meat and improper thawing techniques like using hot water or leaving meat out at room temperature can also contribute to foodborne illnesses. Other mistakes include not marinating properly, buying compromised meat, ignoring safe handling instructions on packaging, inadequate cleaning of equipment, insufficient washing of produce, reusing leftover meat unsafely, and consuming meat that appears spoiled without considering the risk of harmful bacteria.

PREVENTION AND CONTROL OF MEAT-BORNE DISEASES

- Regular veterinary inspections and health monitoring of livestock should be promoted to detect and prevent the spread of diseases from farm animals to meat consumers.

- Stringent hygiene practices should be implemented at all stages of meat production, including thorough cleaning of equipment, surfaces, and hands to prevent cross-contamination of pathogens.
- Proper storage and refrigeration of meat products should be ensured to maintain safe temperatures and prevent the growth of harmful bacteria.
- Consumers should be educated about safe meat handling practices, including thorough cooking, avoiding cross-contamination, and proper storage to reduce the risk of foodborne illnesses.
- Strict regulations and standards should be enforced in slaughterhouses, processing facilities, and informal markets to ensure food safety and minimize the transmission of zoonotic pathogens through meat products.
- One Health approach should be emphasized to integrate veterinary, medical, and environmental expertise in developing and implementing strategies for preventing and controlling meat-borne zoonotic diseases.

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Chapter 15

Overview of National Animal Diseases Referral Expert System

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Livestock diseases have emerged as a significant constraint on the sustainable growth of the national economy, markedly diminishing livestock productivity and hindering sectoral development. The proliferation of various diseases is often attributed to environmental changes and stress linked to intensified farming practices. According to the epidemiological triad, the environment plays a crucial role in facilitating the transmission of pathogens or vectors to susceptible hosts. Therefore, early recognition of livestock diseases, based on the environmental parameters specific to a given area, is paramount. This early detection is critical for effectively controlling diseases and mitigating their economic and social impacts on the nation. Predicting the occurrence of livestock disease outbreaks can be of considerable value to India's long-term sustainable development. Prior research on disease prediction has essentially depended upon traditional statistical models with varying degrees of prediction accuracy. Futurists have anticipated that novel autonomous technologies, embedded with Artificial Intelligence (AI) and Machine Learning (ML), will substantially influence the livestock health sector. AI & ML are focused on making predictions as accurate as possible, while traditional statistical models are aimed at inferring relationships between variables. The benefits of AI & ML comprise flexibility and scalability compared with conventional statistical approaches, which makes it deployable for several tasks, such as diagnosis and classification, and disease events predictions. The geographic and seasonal distribution of many infectious diseases are associated with climate and therefore the possibility of using seasonal climate forecasts as predictive indicators in disease early warning systems (EWS) is an interest of focus. The development and application of various prominent climatic and remote sensing parameters data mining techniques have led to their adoption in AI & ML environments for the prediction of livestock diseases.

National Animal Disease Referral Expert System (NADRES) is one such expert model which deployed data mining techniques, AI & ML for livestock disease prediction and to provide early warning alerts (https://www.nivedi.res.in/Nadres_v2/). NADRES is a dynamic geographic information system (GIS) and remote sensing-enabled (RS) expert system which is based on district-wise animal disease information collected and collated along with

environmental risk factor data over a long period of time. This initiative was developed and monitored by the Indian Council of Agricultural Research (ICAR)-National Institute of Veterinary Epidemiology and Disease Informatics (NIVEDI), Bengaluru by considering 15 economically important livestock diseases including zoonotic diseases like anthrax. This innovative system leverages a combination of PHP, JS, AJAX, HTML, CSS, SQL, and R tools, showcasing a significant advancement in disease surveillance and response within the agricultural sector. This application represents a state-of-the-art analytics system automated with AI algorithms for data collection, cleaning, alignment, and annotations, as well as modelling, risk analysis, and risk communications. The NADRES captures past disease incidence patterns and the source of data is from all over the country collected through National Animal Disease Epidemiology Network (NADEN) centres and also from the Department of Animal Husbandry and Veterinary Services of all the States on a monthly basis. It provides monthly livestock disease forewarning at the district level for all 15 major livestock diseases 2 months in advance and disseminates this information for early response. The predicted results on important livestock diseases are regularly disseminated through forewarning monthly bulletins, webcasts, mobile applications (LDF mobile app), and automated messaging. This proactive approach empowers farmers and veterinary officers to take appropriate control measures and strategize need-based resource allocation if alerts are provided two months in advance. This web application provides several other features such as analysis results of disease patterns in each state, disease maps, distribution of livestock population, impact analysis etc. Developing an optimal sampling strategy for the surveillance of livestock diseases is important for the early detection of disease incidence and it offers the effectiveness of utilisation of limited resources in the surveillance in addition to offering random, representative and independent sampling units to derive the scientifically valid results. In this direction, ICAR-NIVEDI is also providing a Sampling plan for sero-monitoring and sero-surveillance of important livestock diseases such as FMD, Brucellosis, PPR and CSF etc., (https://www.nivedi.res.in/Nadres_v2/samplingplan.php). The application has been optimized by making it easy to navigate so that users stay on and engage with content. All the results in the application are dynamic and automated. Its significance in the field of veterinary epidemiology and disease management, NADRES has been duly recognised as a technological innovation by the Indian Council of Agricultural Research (ICAR) and the LDF mobile application has received copyright. By leveraging advanced technologies and diverse data sources, this system enables timely intervention and safeguards the livelihoods of farmers and the agricultural economy. The development of NADRES exemplifies a significant stride

towards effectively mitigating the impact of livestock diseases through cutting-edge technological solutions.



Outlook of NADRES v2
Homepage



Copyright received for LDF
mobile app



NADRES V2 is certified as
technology from ICAR

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Chapter 16

Alternative approaches for treatment of diseases with special reference to traditional ethnoveterinary practices

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INTRODUCTION

The market for herbal medicines has grown to a value of \$ 62 billion, and according to the WHO, it will reach \$ 5 trillion in value by 2050. Despite its biodiversity and enormous potential benefits, India, one of the emerging nations, has a very little market share due to ineffective pharmacovigilance for herbal products. Since the dawn of time, herbs have been employed all over the world as therapeutic agents for infectious and non-infectious disorders affecting both humans and animals. The most widespread misconception about herbal remedies is that they are perfectly safe and may be used by patients without even a doctor's prescription. This has led to widespread self-medication around the world, frequently with unsatisfactory results, negative effects, or both. Currently, reports of the negative effects of herbal goods and medicines are common. When used alone or in conjunction with other pharmaceuticals, herbal medicines present unique toxicological issues due to differences in how they are identified, perceived, sourced, and used compared to their synthetic counterparts. The development of pharmacovigilance methods for herbal medications is necessary to address these problems. Applying the current pharmacovigilance paradigm and its related technologies to monitor the safety of herbal medications offers special difficulties because they were established in connection to synthetic drugs. To combat inadequate regulatory measures, weak quality control systems, and mostly unregulated distribution routes, the practise of pharmacovigilance of herbals has already started in India. This presentation will offer a brief summary of the recent trends and challenges posed in the practice of pharmacovigilance of herbal drugs especially in the Indian context.

GLOBAL SCENARIO

With an average annual growth rate of around 7%, the global market for herbal medicines has grown to a value of \$ 62 billion, but India's contribution has shockingly remained quite little at just under \$ 1 billion. Despite having a small market share for herbal pharmaceuticals, India enjoys a strong reputation on the international stage and is acknowledged as one of the world's major biodiversity hotspots. By 2050, the WHO projects that the global market for herbal

products would increase to \$5 trillion. Despite its biodiversity and numerous benefits, India, one of the developing nations, lacks effective pharmacovigilance for herbal goods. Herbal drug authorizations in Europe are very similar to new drug approvals in the United States, where medicines are recognised for their efficacy, quality and safety. The major way to monitor the safety of herbal medicines in the UK is through the "yellow card" Adverse Drug Reaction (ADR) reporting system run by the Medicines and Healthcare products Regulatory Agency (MHRA). Additionally, in order to find previously unknown safety risks or side effects, they simultaneously compare yellow card reports they receive with additional sources of information including clinical trial data, medical literature, or data from international medicines regulators.

INDIAN SCENARIO

The Indian Central Drugs Standard Control Organization [CDSCO] has a similar system to the yellow card programme, but it is only for use by healthcare professionals and is intended to encourage "voluntary" reporting of ADRs. If we're being completely honest, we haven't seen a lot of people in India aware of the aforementioned system. They are therefore unsure about "where" and "why" to report. To make this system successful and functional, it is the duty of the Indian Health Authority to disseminate this knowledge throughout the nation. The UK government established this system in 1964 and made it a legal requirement for pharmaceutical firms to disclose the findings of any allegations of allegedly major adverse effects of their medications. The "yellow card" reporting forms are used to submit these reports. Additionally, they have created a website just for such reports that is open to everyone, including the general public. The perception, on the other hand, is that only healthcare professionals in India have access to this information, and I seriously doubt that many of them are particularly aware of it or diligent about it. Additionally, India lacks any websites that educate the general public about the value of pharmacovigilance and the assistance of healthcare experts.

Adverse effects of all types of treatments are purchased into the "Med Watch" programme in nations like the USA. India may implement similar pharmacovigilance programmes for natural medicines. The amount of adverse reactions to herbal medicines that have been reported to or documented in India's National Pharmacovigilance Programme is quite negligible. One of the factors contributing to this predicament is the widespread belief that herbal or ayurvedic drugs are safe. However, it is a fallacy brought on by herbal practitioners' or healthcare professionals' ignorance of the truth and the idea of pharmacovigilance with regard to herbal products. To

give an example, recent research on the subject have shown just how little people actually know about herbal pharmacovigilance. This inquiry exposed the mindset of many herbalists who maintained that "herbal products do not have any form of detrimental effects" and that very few of them thought there were any. This results in incorrect production processes and illogical prescriptions. Some patients had unintended side effects after receiving a prescription, but they failed to disclose them, and this is where a lack of pharmacovigilance can be seen.

CHALLENGES IN MONITORING THE SAFETY OF HERBAL MEDICINES REGULATION

The national licencing and regulation of herbal medicines varies from nation to nation. Herbal medications can be classified as either prescription or over-the-counter drugs in jurisdictions where they are controlled. Other classifications besides medications are possible for herbal goods. Additionally, the regulatory status of a certain herbal substance may vary between nations. Typically, qualified suppliers and distributors of the relevant substances are also included in the national regulatory framework. Consequently, the distribution method or access to these products is determined by their regulatory status.

QUALITY ASSURANCE AND CONTROL

Every country that regulates herbal medicines should have quality assurance and control measures in place, including national quality specifications and standards for herbal materials, good manufacturing practises (GMP) for herbal medicines, labelling, and licencing schemes for manufacturing, imports, and marketing. These controls are essential for guaranteeing the efficacy and safety of herbal medications. Inadequate regulation and quality control may lead to a high prevalence of unpleasant reactions, particularly when it comes to herbal medications, which may be contaminated with potentially harmful ingredients and/or adulterated with undisclosed powerful drugs.

Compared to other medications, completed herbal products have significantly more complicated criteria and quality control procedures, especially for mixed herbal products. The calibre of the raw materials used has an impact on the quality of such items. Good farming and gathering methods, including careful plant selection and cultivation, are crucial precautions.

SAFETY MONITORING OF HERBAL MEDICINES

Clinical trials and unprompted reports are the most prevalent sources of data on adverse occurrences and reactions to medications (voluntary, unsolicited communications on marketed medicinal products). Over the course of a product's existence, the latter typically outnumber

the former in both numbers and type, especially serious reports. In certain nations, doctors are required to report unfavourable reactions; these reports are viewed as spontaneous.

Other than doctors, dentists, pharmacists, and nurses, herbal medicine providers are frequently left out of reporting systems. National reporting schemes should be developed to include all providers of herbal medicines (both prescribers and dispensers), as well as providers of traditional, complementary, and alternative medicine, depending on national circumstances, if adequate coverage of herbal medicines is to be achieved.

A sizeable number of herbal medications are non-prescription drugs, and many enter this category without having undergone any post-marketing safety testing as prescription drugs. Therefore, it is crucial to take action to boost pharmacovigilance activity in the context of over-the-counter drugs. Despite the fact that many of these items are offered outside of pharmacies, community pharmacists and nurses can be very helpful in ensuring the safety of over-the-counter medications. Consumer involvement in the use of herbal medications and herbal products in healthcare, as well as their worry over potential side effects, should be regarded favourably. Consumer complaints about side effects should be taken seriously as a source of data that might help researchers spot warning signs of potential consequences of herbal medications.

RECORDING AND CODING THE IDENTITY OF HERBAL MEDICINES

It is preferable to use a uniform classification and identification when sending reports to the UMC. It is recommended that the coding of adverse events and adverse responses to herbal medicines be consistent with that for other medications. The WHO Drug Dictionary (WHO-DD), which was created to contain organised, categorised information on the names of herbal products and their constituents in the same way as equivalent information on other medicines, is what the UMC recommends using as a result. The UMC suggests the herbal anatomic-therapeutic-chemical (HATC) classification for the therapeutic classification of herbal products, which is structurally identical to the anatomic-therapeutic-chemical (ATC) classification used for chemical substances in other treatments. In order to integrate with the worldwide WHO database, HATC is being deployed under the WHO-DD framework. All levels of data input, however erroneous, can be managed through the use of the HATC classification and the extended global WHO database structure. The UMC also suggests a comprehensive checklist for cross-referencing the names of components that are given in both botanical and vernacular nomenclature. The national pharmacovigilance centres should find

the WHO-DD, the HATC classification, and the checklist to be helpful resources when posing questions to the reporter in order to improve the clarity and accuracy of reports, according to the UMC.

It is frequently impossible to identify every constituent in herbal medications because they frequently include several. In these situations, it is advisable to write down the product name and refer it to the UMC, which will aid in identification. The product will be added, along with any pertinent data, if it isn't already in the worldwide WHO database. A specific herbal product could have many indications and so show up in various categories under the HATC categorization.

The reporter's local knowledge of the product's accuracy or lack thereof is most valuable. Free text input, comments on the report, the manufacturer's information, or images of the original package can all be used to offer this. As a resource on medicinal plants and their use in the community, a national inventory or catalogue of medicinal plants may be useful. However, the usage of therapeutic herbs and knowledge of them are not widely documented. Therefore, it should be promoted to create a national inventory or catalogue.

The drug regulatory agency of the exporting nation may be able to aid if the finished herbal product in question or its raw materials were imported from other nations.

When feasible, the precise Latin binomial botanical name of the medicinal plant (genus, species, author, as well as name of family) should be given, together with details on the plant parts used and the extraction and preparation techniques used. With the use of this information, other reports can be compared accurately. To prevent the filing of a report from being postponed or cancelled, a popular vernacular name may be employed. Regarding taxonomy (botanical and chemical) identification and botanical and vernacular nomenclature, national pharmacovigilance centres should cooperate with university pharmacognosy departments, botanists, zoologists, and botanical garden employees.

DATA MANAGEMENT

Data quality: Every effort should be taken to guarantee that data processing is subject to quality standards and that report data elements are as comprehensive and accurate as feasible. It is necessary to put in place mechanisms to look for duplicates.

Data storage: To make the data easier to access and utilise, computer databases should be handled to the highest standards available. So that analytical requirements are met, software should be chosen with the help of experts.

Data analysis: It is important to have programmes that offer regular analyses and data output that is suitable for regional requirements.

Analysis of the worldwide WHO database: The suggested "Database management and categorization for coding of herbal medicines," of which the aforementioned HATC is one aspect, is being used to improve the global WHO database controlled by the UMC. [12] Signal detection on reports for herbal medications will be done using data-mining approaches that have proven successful on a very large number of reports for other drugs. The quantity and calibre of data given by the national Pharmacovigilance Centers determines how well these strategies work.

The WHO Collaborating Centre for International Drug Monitoring, UMC (<http://www.who-umc.org/>), offers assistance with technical and data management.

CONCLUSION

Both developed and developing nations use herbal medications extensively in healthcare. However, a number of high-profile herbal safety issues have surfaced recently that have affected the public's health, and there is growing awareness of the need to create pharmacovigilance (safety monitoring) systems for herbal medications. In many ways, pharmacovigilance for herbal medications is in its infancy, and assessing their safety presents particular difficulties.

At both the national and international levels, the purpose of this meeting is to present a thorough and critical evaluation of the current state of pharmacovigilance operations for herbal medicines. It will thoroughly examine the difficulties that herbal medicine pharmacovigilance presents, take into account pertinent new concerns, and assess what actions may and should be taken to improve safety monitoring for herbal medicines in the future.

Chapter 17

Advanced Techniques for Diagnosis of Bovine Diseases

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INTRODUCTION

Currently, culture-based bacterial isolation and identification are the gold standard methods for detection of pathogens. But these methods are relatively time consuming, laborious and technically demanding requiring professional expertise and remains problematic due to the chances of biohazards while handling them. It is evident that culture-based methods are inadequate for rapid detection of pathogens, especially for reducing disease occurrence. The frequent outbreak of microbial diseases and their economic and social implications indicate that analytical methodologies that can rapidly detect and identify pathogens are urgently needed. Advances in biotechnology and bioinformatics have resulted in the development of novel testing technologies that enable more reliable and faster detection of microbial pathogens. These novel methods, although still not applied routinely in everyday practice, are the promising alternative which can replace or can be used as an addendum to current reference methods. As such, many researchers devote themselves to developing more advanced detection methods that can identify pathogens accurately and rapidly in a timely manner so that appropriate treatment may be initiated at the earliest and suitable strategies may be adopted for their control.

CONVENTIONAL METHODS FOR DETECTION OF PATHOGENS

Conventional methods for the detection and identification of microbial pathogenic agents rely mainly on microbiological, biochemical and/or serological identification which include culture of the target organism, visualization of colony morphology (macroscopic morphology). Staining (microscopic morphology) followed by biochemical characterization and serotyping for confirmation of the pathogenic organism. These methods are specific, in-

expensive and can provide both qualitative and quantitative information of the pathogens present in the clinical sample however, they are disadvantageous as time taken in the assay is usually high in order to confirm the pathogens. Moreover, media preparation, handling of infected samples and their processing are labor intensive, technically demanding and may be hazardous. Therefore, the diagnostic industry is in need of more rapid methods which are: rapid to provide immediate information on the possible presence of pathogen in samples, to detect pathogens which are in low numbers, to reduce human errors and to save time and labor cost.

RAPID METHODS OF PATHOGEN IDENTIFICATION

Any modification that reduces the time taken in the confirmation of pathogens by an assay can technically be called as a rapid method. Advances in biotechnology, bioinformatics and engineering have led to the development of a diverse array of assays for detection of pathogens that have reduced the assay time. These assay procedures include automated conventional assays, nucleic acid-based tests, lateral flow assays, biosensors, next generation sequencing (NGS) and MALDI-TOF MS based methods.

NUCLEIC ACID BASED ASSAYS

Nucleic acid-based techniques offer more sensitivity and specificity than culture-based methods as well as considerable reduction in the time taken for getting results. There are many nucleic acid-based assays but only DNA probes and PCR based assays have been developed commercially for detecting various pathogens.

NUCLEIC ACID HYBRIDIZATION METHODS

DNA probes

A DNA probe is a short single-stranded sequence of nucleotides, usually 14-40 bp long which will bind to specific regions of single-stranded target DNA. The homology between the target and the DNA probe results in stable hybridization. Hybridization is monitored by labelling probes, by attachment to or incorporation with compounds into the probe that can be detected visually or chemically. Isotopes such as ^{32}P can be incorporated into the structure of the probe. Enzymes such as alkaline phosphatase or horseradish peroxidase (HRP) are often linked to the probe and hybridization can be detected visually by addition of substrates. Fluorescently labelled compounds such as fluorescein isothiocyanate (FITC) can also be attached directly to probes. DNA probes for detection of several pathogens have been commercialized.

DNA MICROARRAYS (GENE CHIPS)

Microarrays or gene chips are miniaturized system for the simultaneous analysis of hybridization of fluorescent-labelled single strand nucleotide chains to an array of oligonucleotide probes immobilized on a support such as glass or a synthetic membrane. PCR amplification is often used prior to hybridization to increase sensitivity of detection. DNA microarrays may be very useful for detecting multiple microbes simultaneously on a single chip. The diverse nature of the clinical samples is major drawback of microarrays to be used as a detection method. Recently, a microarray system for *Salmonella* detection has been introduced by which serotype/genotype of *Salmonella* can be known within few hours. Development of this approach is continuing at a rapid pace and for microbiologists, this technology has potential to become one of the major diagnostic tools in future.

NUCLEIC ACID AMPLIFICATION METHODS

Polymerase Chain Reaction (PCR)

Since its invention in 1985 by Kary Mullis, Polymerase Chain Reaction (PCR) has emerged as the most powerful molecular biology technique in the area of diagnostics. It is now being increasingly used for monitoring of potential pathogens in clinical samples, so that the early detection can lead to implementing appropriate control measures by authorities.

Using PCR, the target DNA sequence can be selectively amplified, thus increasing the amount of target available for detection. The PCR technique is a three-step process including denaturation, annealing and extension. A typical amplification of 30-40 cycles results in million-fold multiplication of the targeted DNA, with theoretical sensitivities down to a single target pathogen. Nucleic acid-based assays are claimed to be highly sensitive and specific, if suitably designed and optimized. PCR based methods are used in detecting wide range of pathogens like *S. aureus*, *L. monocytogenes*, *Bacillus cereus*, *Yersinia enterocolitica*.

By using multiplex PCR, it is now possible to simultaneously detect more than one pathogen in the suspected samples and the results are available in less than 24 h. Rapid test kits for detection have been developed and now commercially available from various reputed firms.

Real-Time PCR

Real-time PCR also called quantitative PCR (qPCR) or kinetic PCR is a laboratory technique in which identification and simultaneous quantification of a targeted pathogen can be done. The procedure follows the general principle of PCR. Its key feature is that the

amplified DNA is detected as the reaction progresses in real time. The quantification can be either absolute (number of copies of target DNA) or a relative (when normalized to DNA input or additional normalizing genes).

Various chemistries are available for Real-time PCR including 1. SYBR Green dye (non-specific fluorescent dyes that intercalate with any double-stranded DNA); 2. Hydrolysis probes (e.g. TaqMan probes which are sequence-specific DNA probes consisting of oligonucleotides that are labelled with a fluorescent reporter at one end and quencher at other which permits detection only after hybridization of the probe with its complementary sequence followed by its cleavage and subsequent release of reporter); 3. Hybridization probes (e.g. Molecular Beacons and Scorpion probes) and 4. FRET probes.

Measuring the kinetics of the reaction in the early phases of PCR cycles has a distinct advantage over conventional PCR based detection in which post-PCR processing reduces sensitivity and increases time. With the ability to collect data in the exponential growth phase, the power of PCR has been expanded into applications such as: microbial quantification, quantification of gene expression, array verification, drug therapy efficacy, DNA damage measurement, quality control and assay validation, pathogen detection and genotyping. Advantages of using Real-Time PCR over traditional PCR include; 1. Traditional PCR is measured at End-Point (plateau), while Real-Time PCR collects data in the exponential growth phase; 2. An increase in reporter fluorescent signal is directly proportional to the number of amplicons generated and thus copy number of target pathogen; 3. Increased dynamic range of detection; 4. No-post PCR processing is involved and therefore require much less time for results; 5. Detection is capable down to a 2-fold change.

In recent years Real-Time PCR has emerged as a leading technology for rapid identification of pathogens due to its speed and high degree of sensitivity and specificity. During the nCoV pandemic, TaqMan probed based PCR was successfully used globally as a gold standard for diagnosis of SARS-CoV-2.

Isothermal Amplification Methods

Loop mediated isothermal amplification (LAMP) is a novel technique originally developed by Notomi *et. al.*, 2000. The principle of LAMP is auto-cycling strand displacement DNA synthesis in the presence of *Bst* DNA polymerase with high strand displacement activity under isothermal conditions at 60-65⁰C for 30-60 min. The final amplification products are mixtures of different sizes of stem-loop DNAs with several inverted repeats of the target sequence and cauliflower-like structures with multiple loops.

The advantages of LAMP over PCR technique are; 1. A set of two specially designed inner primers and two outer primers recognize six distinct sequences on target DNA, so the method is expected to amplify the target sequence with high specificity; 2. As the reaction is under isothermal condition, no thermal cycler is required and no time is lost in thermal change; 3. The amplification efficiency of LAMP is extremely high; 4. Continuous amplification produces great amount of target DNA (to 10^9 level) as well as large amount of by products leading to formation of white precipitate of magnesium pyrophosphate, within 30-60 min, allowing easy and rapid visual detection; 5. The presence of the non-target DNA and inhibitors are less likely to affect the results in LAMP assay. Therefore, LAMP constitutes a potentially valuable tool for rapid detection of microbial pathogens in either commercial or field laboratories.

Immunological Methods

The basic principle for an immunological method is the specific binding of the antibody to a target antigen followed by the detection of the antigen–antibody complex. Immunological methods are promising because of their sensitivity, rapidity and simplicity, and also because testing can be carried out directly from clinical sample or enrichment media without cumbersome sample preparation steps. There are many different commercially available antibody-based assays for microbial pathogens or their toxins.

Latex Agglutination Test (LAT)

LAT is the simplest among the immunological assays, which detects the presence of an antibody or an antigen using latex beads coated by an antibody (or antigen). If the corresponding antigen (or antibody) is present in the sample, it is attached to the latex beads, and they agglutinate due to the formation of molecular cross-bridges. The sensitivity of the method is 10^7 – 10^8 cfu ml⁻¹ of sample. There are commercially available LATs for several bacterial pathogens such as *Campylobacter*, *E. coli*, *Listeria* spp., *Salmonella* spp., *Shigella* spp., *S. aureus* and *Vibrio cholerae*. Many of the LAT assays are performed manually, and the results are given by visual observation. However, these manual assays suffer from poor sensitivity and reproducibility. During the last years, visual assays have been replaced by spectrophotometers and nephelometers that measure the absorbed or scattered light.

Reverse Passive Latex Agglutination (RPLA)

It is a modification of the LAT. Antibodies that are attached to latex beads react with soluble antigens in test tubes or in wells of microtitre plates. If binding occurs, a diffuse pattern

is observed at the bottom of the tube or well. If not, a ring is observed. RPLA is more suitable for testing for toxin production. Commercially, there are tests for detection of enterotoxins and other toxins produced by *S. aureus*, *B. cereus*, *Cl. perfringens* and *E. coli*.

ENZYME LINKED IMMUNOSORBENT ASSAY (ELISA) AND ENZYME LINKED FLUORESCENT ASSAYS (ELFA)

ELISA couples an immunoassay with an enzyme assay. For most of the pathogen (antigen) detection, a sandwich format is used utilizing two antibodies to trap the target antigen followed by the addition of an enzyme-labelled secondary antibody. In the final step a substrate is added that the enzyme can convert to a detectable signal proportional to the concentration of the analyte (antigen).

The ELISA detection itself only takes 2-3 h. Now-a-days, many ELISA tests are available as automated systems to reduce the hands-on time and improve reproducibility. While many ELISA-based methods rely on chromogenic substrates for end-point detection of the target antigen, ELFA employs fluorescence for endpoint detection.

LATERAL FLOW ASSAYS

Lateral flow assays (LFAs) also known as immunochromatographic tests (ICTs) or strip tests are the simplest immunological assays. They have been a popular diagnostic platform as they are cheaper, user friendly and less time consuming. The main driver for their popularity has been pregnancy diagnosis and glucose monitoring kits available in the market. LFAs are the simplest to use of all the test formats requiring the user to place the test strip in the specimen or pour specimen directly onto it. The sample may be from a clinical case, food or environment. Now-a-days, LFAs are in use for the specific qualitative or semi-quantitative detection of one or more analytes (multiplexing). Therefore, there is a growing demand for development of lateral flow assay devices for fast, accurate, reliable and economic detection of pathogens or their toxins.

LFAs detect the presence or absence of a target analyte in the sample. These are a form of immunoassay in which the test sample flows along a solid substrate via capillary action. A colored reagent then mixes with the sample and then transits the substrate encountering lines or zones pretreated with an antibody or antigen. Depending upon the analytes present in the sample, the colored reagent can become bound at the test line. Some LFA strips are available for detection of pathogens or their toxins in clinical and food samples from Merck-Millipore, Dupont and other reputed vendors.

FLOW CYTOMETRY (FCM)

It is a powerful technique based on the same principle as immunofluorescence. The instrument detects fluorescent cells that are moving in a fluid stream past an optical sensor. The flow cytometer measures the light scattered or the fluorescence emitted by the cells as they pass through the laser beam in a single file (obtained by hydrodynamic focusing). The light energy is converted into an electrical signal by photomultiplier tubes. FCM is sensitive, does not require culturing or enrichment procedures and can be both qualitative and quantitative. It can be used as a tool to determine product quality. FCM combined with immunofluorescent labelling has been used to detect *L. monocytogenes* and other pathogenic bacteria in samples.

BIOSENSORS

A biosensor is an analytical device typically consisting of a bio-recognition component, transducer, a signal amplifier, processor and computer display. The recognition component, called a bioreceptor uses biomolecules to interact with the analyte of interest. This interaction is measured by the transducer which converts it in to digital signal which in turn gets amplified by the amplifier and finally displayed by the data processing unit/computer. Biosensing methods for pathogen detection are centered on four basic physiological or genetic properties of microorganisms: metabolic patterns of substrate utilization, analysis of phenotypic expression of signature molecules by antibodies, nucleic acid analysis and analysis of interaction of pathogens with eukaryotic cells. However, antibody-based methods are the most popular because of their versatility, convenience and relative ease in interpretation of the data. Majority of biosensors use antibody for capture and detection of the target analyte.

Biosensors can be classified according to common types of bioreceptor interactions involving: antibody/antigen, enzymes, nucleic acids/DNA, cellular structures/cells, or biomimetic materials. Biosensors can also be classified by their transducer type. The most common types of biosensors are 1) electrochemical biosensors (amperometric, potentiometric and impedimetric), 2) optical biosensors, 3) electronic biosensors, 4) piezoelectric biosensors, 5) gravimetric biosensors, 6) pyroelectric biosensors 7) Surface Plasmon Resonance (SPR) biosensor, 8) Cell based biosensors, 9) DNA based biosensors 10) Bioluminescence biosensors (ATP bioluminescence and Bacterial bioluminescence) and 11) Immunosensors.

Potential applications of biosensors include 1) Glucose monitoring in diabetes patients (historical market driver) and other medical health related targets 2) Environmental applications e.g. the detection of pesticides and river water contaminants such as heavy metals

3) Remote sensing of airborne bacteria e.g. for countering bioterrorism threats 4) Detection of pathogens 5) Determining levels of toxic substances before and after bioremediation 6) Determination of drug residues in food, such as antibiotics and growth promoters, particularly in meat and honey 7) Drug discovery and evaluation of biological activity of new compounds 8) Detection of toxic metabolites such as mycotoxins etc.

Detection with antibodies, which are specific for a particular microorganism, or toxin is the basic approach utilized in immunosensors. In recent years, DNA-based biosensors have been developed for the detection of pathogens. Wang (2002) successfully developed novel genosensors for *Cryptosporidium*, *E. coli*, *Giardia* and *Mycobacterium tuberculosis*. Biosensor technology is advantageous in significantly reducing time for assay, detecting much smaller amounts of target analyte with fewer false positives, high throughput and capability for automation.

OTHER RAPID METHODS FOR PATHOGEN DETECTION

Bacteriophage Based Methods

Bacteriophages (phages) are viruses infecting bacteria. In general, phages are highly host-specific. The specificity of these phages is partly mediated by tail-associated proteins that recognize surface molecules of susceptible host bacteria. The specific bacteriophage tail-associated proteins may be attached to paramagnetic beads to capture bacteria in suspension. The bacteria bead complex can be integrated in fast detection protocols. The *Listeria* Capture kit (Hyglos) can be integrated as part of a rapid detection method in a similar way as IMS. Results of the test are obtained within hours although a prior enrichment of 6-24 h may be required depending upon the type of pathogen.

Fourier Transform-Infrared Spectroscopy (FT-IR)

FT-IR is a rapid non-destructive method and sample identification depends on the available spectral library. This method requires transfer of cells (biomass) from the growth media to an IR reflecting substrate for spectral collection. When IR is absorbed or transmitted through the sample to the detector, it generates a characteristic scan or fingerprint profile. A library of spectral scans can be generated for different bacterial species and strains, which can be used for future comparison to correctly identify an unknown sample. FT-IR has been used for identification of several pathogens viz. *Yersinia*, *Staphylococcus*, *Salmonella*, *Listeria*, *Klebsiella*, *Escherichia*, *Enterobacter*, *Citrobacter* etc. FT-IR photoacoustic spectroscopy was used for the identification of spores of several *Bacillus* species with 100% accuracy.

NEWER TECHNOLOGIES FOR PATHOGEN DETECTION

Matrix Assisted Laser Desorption Ionization-Time of Flight Mass Spectrometry (MALDI-TOF MS)

In the recent years MALDI-TOF MS has emerged as a potential tool for microbial diagnosis. In the MALDI-TOF MS, microbes are identified using either intact cells or cell extracts. The process is rapid, sensitive, and economical in terms of labor and costs involved. The technology has been readily used for microbial identification. The limitation of the technology is that identification of organism is possible only if the spectral database contains peptide mass fingerprints of the type strains of specific genera/species of the target organism to be identified.

The method is technically simple and rapid. Bacterial colonies are removed from culture plates, mixed with an excess of UV-absorbing matrix, and dried on target plates (stainless steel). The dried preparations are exposed to laser light, resulting in energy transfer from the matrix to the non-volatile analyte molecules, with desorption (removal) of analyte into the gas phase. The ionized molecules are accelerated by electric potentials through a flight tube to the mass spectrometer, with separation of the biomarkers determined by their mass/charge ratio (m/z). The profile of biomarkers is then compared with spectral profiles of a collection of well-characterized organisms. The most common reason an isolate is not identified is because it is not included in the database. As experience is gained with MALDI-TOF MS, it is expected that the database will be expanded to include a greater number of organisms.

MALDI-TOF MS offers the possibility of accurate, rapid, and inexpensive identification of microorganisms. Although, only limited work has been reported on the use of mass spectrometry to identify pathogens, the technique is rapidly picking up in the area of diagnostics. More number of systems are being installed in last few years throughout the country due to its versatility. Two major systems are MALDI Biotyper (Bruker Daltonics, USA) and Vitek MS (bioMerieux, France). Other companies have also launched their systems in the recent years including Auto EXS2600 (Zybio, China).

Next Generation Sequencing (NGS)

Another revolutionary discovery in bioinformatics is next generation sequencing. This is a powerful new technology which enables generation of over one million DNA sequences per run, parallel analysis of multiple samples, detection of unknown pathogens in complex

samples and is yet to widen its application in pathogen detection. New chemistries for sequence reading are:

- Pyrophosphate detection (PPi release upon base addition): Roche 454
- Single (reversibly 3'-blocked) fluorescent base (quenchable) added/step: Illumina SolexaMiSeq
- Sequencing by Ligation (ABI SOLiD)

Applications of NGS include: a. Pathogen detection and discovery-Clinical metagenomics b. Polymorphism detection and discovery-Genomic epidemiology and adaptive changes c. Pathogen biology-Gene detection and discovery.

Oxford Nanopore Sequencing Platform:

The recent addition to NGS platforms is Oxford Nanopore Sequencing. Nanopore sequencing is a unique, scalable technology that enables direct, real-time analysis of long DNA or RNA fragments. It works by monitoring changes to an electrical current as nucleic acids are passed through a protein nanopore. The resulting signal is decoded to provide the specific DNA or RNA sequence. The advantages of using long nanopore DNA sequencing reads for researchers are: a. Resolve complex structural variants and repetitive regions b. Simplify de novo genome assembly and improve existing reference genomes c. Study linkage and phasing d. Enhance metagenomic identification of closely related species and distinguish plasmid from genome e. Sequence entire microbes in single reads – in real-time f. Explore epigenetic modifications using direct, long-read DNA sequencing.

This platform has been extensively used for genetic characterization and classification of SARS-CoV-2.

CONCLUSION & FUTURE PROSPECTS

Early screening of clinical samples for target pathogens is an important measure to prevent disease outbreaks therefore, the present era demands highly sensitive, specific, rapid and cost-effective diagnostic assays for microbial pathogens. Conventional, immunological, and nucleic acid-based methods provide reliable results, but require specialized equipment and personnel and not suitable for automation. Flow cytometry, gene chips, biosensors, MALDI TOF-MS and NGS are promising and upcoming technologies, however due to high cost of equipment, their popularization in routine diagnostic use will take some more time. Technologies continues to advance at a great pace and next generation assays already being

developed potentially have the capability for detection of multiple pathogens. As rapid methods are used more frequently, their benefits and limitations also become more apparent.

The global in-vitro diagnostics (IVD) market was valued at \$44 billion (\$ 4400 Crore= Rs. 352000 Crore) in 2011 and is expected to have a modest growth. IVD market include Clinical Chemistry, Molecular diagnostics, Immunoassays, Haematology and Microbiology. Clinical chemistry dominates global IVD market while molecular diagnostics is expected to register the highest growth. The U.S. represented the biggest market for IVD equipment accounting for about a half of the total market. Increased patient awareness, patient self-testing, rapidly aging population globally and automated testing due to technical advances are the major growth drivers of this market. The molecular methods are a promising alternative that can substitute or complement the current reference methods used. Sample preparation problem, contamination, entrenched attitude slow but cannot stop adoption of the molecular method of diagnosis.

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ABOUT ICAR NRC ON MITHUN, MEDZIPHEMA, NAGALAND-797106

ICAR-National Research Centre on Mithun was established in the year 1988 under the aegis of Indian Council of Agricultural Research (Department of Agriculture Research and Education, Ministry of Agriculture and Farmers' Welfare, Government of India). The Institute has two campuses: the main campus is located at Medziphema, Chumukedeima district, Nagaland and the other is located at Porba village of Phek district of Nagaland which is approximately 125 km from the main campus, Medziphema, 81 km from Kohima and 150 km from Dimapur. Krishi Vigyan Kendra (KVK-Phek) of the Institute is also located at the Porba campus. ICAR-NRC on Mithun is the only research organization in the world, exclusively working for the continual improvement and conservation of Mithun (*Bos frontalis*). The Research and Development (R&D) initiatives of the Institute are accomplished by seven sections viz., Animal Genetics & Breeding, Animal Physiology & Reproduction, Animal Nutrition, Animal Health, Livestock Production & Management, Livestock Production Technology and Veterinary Extension. Over the years the Institute has developed state-of-the-art infrastructure facilities which includes Biotechnology Infrastructure Facility-cum-Central Instrumentation Facility, Semen Processing Laboratory and Mithun Farm. The library with online journal search facility and Agricultural Knowledge Management Unit (AKMU) cell caters to the needs of the scientists, Research scholars and others staff members of the Institute as well as neighboring Institutes. The Institute's Mithun Farm, located about 8 km away from the main campus on a hill-top, is housing about 100 Mithuns under semi-intensive system of rearing. Another Mithun Farm is located at Porba campus of the Institute.

During the last more than 35 years, the Institute has not only generated invaluable scientific data towards the understanding of this unique species but also the developed several packages of practices and technologies. Conservation efforts including taming of mithun and demonstrating an alternative system of semi-intensive rearing of mithun like other bovine species resulted in the complete domestication of the species. The Institute has established two conservation units at Khonoma and Thevopisumi. Further, the Institute has seven patents, seven copyrights and seven designs. Further, 16 technologies have been certified by the Indian Council of Agricultural Research, New Delhi and four technology transfer agreements have been signed. Capacity building is an important sphere of Institutional activities. Till date the Institute has reached out to more than 20,000 stakeholders through various awareness and capacity building programs.



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