Review on infectious bovine keratoconjunctivitis.

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Abstract

Infectious Bovine Keratoconjunctivitis (IBK), sometimes referred to as 'pink-eye' or New Forest disease, is a highly contagious condition affecting the superficial structures of the eyes. This disease is most commonly caused by *Moraxella bovis.* The main aim of this seminar is to review on the general aspect of infectious bovine keratoconjunctivitis. Infectious bovine keratoconjunctivitis is considered the most important ocular disease in cattle production, due to the decreased growth performance of infected individuals and its subsequent economic effects. This ocular disease is characterized by excessive tearing and ulceration of the cornea and perforation of the cornea may also occur in severe cases. Cattle of all ages are susceptible and ource is carrier cattle, with transmission by mediate contagion and by flies. The disease is more common in summer months and *Bos taurus* breeds appear to be more susceptible to infection than *Bos indicus* breeds. The current treatment and prevention measures can be unrewarding and often do not circumvent the economic losses. Despite the efficacy of antimicrobial therapy, treatment of affected cattle has many disadvantages and prevention of infectious bovine keratoconjunctivitis through application of appropriate management, control of the vectors and vaccination plays a vital role in combating the disease is preferable. Even though the presence of disease has been reported in Ethiopia, yet no consideration has been given for this disease. Therefore, Studies should be conducted to determine the prevalence, incidence, economic impact and the disease status in Ethiopia.

Keywords: *Infectious bovine; Keratoconjunctivitis; Moraxella bovis; Pink-eye.*

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Introduction

Infectious Bovine Keratoconjunctivitis (IBK), sometimes referred to as 'pink-eye' or New Forest disease, is a highly contagious condition affecting the superficial structures of the eyes. Pink eye disease is an economically important disease of cattle and may infect up to 80% of herd within 3 weeks. This disease is a highly contagious, painful ocular disease that affects cattle of all ages and occurs worldwide. The bacterium adheres to the cells via its fimbriae and pili proteins, and produces β-haemolysin toxins which lyse the corneal epithelial cells [1].

The most common causative agent of IBK is *Moraxella bovis*. It sticks to the surface of the eyeball with hair like projections called "pili". Once attached, it releases a toxin that kills the cells on the surface of the cornea, leading to the characteristic corneal ulcers seen with this disease. The organism is located in the eyes and nasal cavities of infected cattle. Cattle are the only known reservoir of *M. bovis* and infected carrier animals may harbor this organism year round without showing any signs of eye problems. Apart from the etiologic agent *Moraxella bovis*, many factors including exposure to UV light, accumulation of dust and trauma at ocular region etc., predisposes the infection. Pinkeye is a highly contagious and infectious disease. The ability of *M. bovis* to cause the disease is influenced by host (the cattle) and environmental factors.

Breed differences in susceptibility of IBK have been established. In a number of studies, Hereford, Jersey and Holstein breeds appear to be more susceptible to infection than *Bos indicus* breeds. The disease is influenced by many factors, such as season, mechanical irritation (dust, grass, and weeds), host immune response, eye pigmentation, and concurrent presence of pathogenic bacteria, environment, and strain of *M. bovis*. Pathogenic strains of *M. bovis* are piliated strains that initially bind through their pili to receptors on the surfaces of corneal epithelial cells. The face fly Musca autumnalis is the important species in transmission of *M. bovis*. Flies may serve as mechanical vectors of the bacteria. Pathogenic strains of *M. bovis* are piliated strains that initially bind through their pili to receptors on the surfaces of corneal epithelial cells. Moreover, the ocular and nasal discharges of infected animals can carry the pathogens, hence direct transmission from animal to animal contact, contaminated equipment and animal handlers can also transmit the disease. The disease is characterized by increased ocular secretion, conjunctival hyperemia, edema, corneal opacity and ulceration of the infected eyes. *M. bovis* possesses virulence factors that allow it to colonize the eye and result in infection. However, current treatment and prevention measures are unrewarding and often do not circumvent the economic losses [2].

Infectious bovine keratoconjunctivitis is considered as the most important ocular disease in cattle production, due to the decreased growth performance of infected individuals and its subsequent economic effects. It can also cause great economic impact such as reduction in growth rate of growing calves, reduction in milk production of dairy cows, reduction in the market values of afflicted cows, decreased weight gain in beef breeds, short term disruption of breeding programs, and treatment costs. Tremendous economic losses also stem from inappetance, suffering from ocular pain, visual impairment. Other major losses resulting from IBK include the direct costs of repetitive drug treatments. In spite of the above situation there is scarcity of information. Therefore the main objective of this seminar is to review etiology, epidemiology, pathogenesis, clinical sign, diagnosis, treatment, and control and prevention of infectious bovine keratoconjunctivitis. Furthermore, the current stutus of IBK is also highlighted.

Litarature Review

Etiology

The most common causative agent of IBK is *M. bovis*. Although *M. bovis* is the most commonly isolated pathogen from IBK, the occurrence and clinical severity is mediated by various factors such as the environment, season, *M. bovis* strain and host immune response. *M. bovis* is a gram-negative coccobacillus, non-motile, free-living bacteria measuring between 0.6-1.0 µm in diameter, lacking flagella with varying amounts of pili. *M. bovis* is able to use colonial morphology as a way to adapt to environmental changes Colonies can interchange between spreading corroding and non- corroding morphologies. The more virulent and common colony type is that of the SC form which the bacteria grow in a flat cylindrical disk. This organism is non-motile, aerobic and usually catalase-positive and oxidase-positive. Although proteolytic, it is unable to utilize sugars. Growth, which is enhanced by the addition of blood or serum to media, does not occur on MacConkey agar. Virulent strains, when isolated from cases of infectious bovine keratoconjunctivitis, are fimbriate, haemolytic and grow into the agar.

Epidemiology

Distribution and occurrence: *M. bovis* is disseminated worldwide. Infections with these bacteria are most commonly seen in hot months. Summer and early fall are the peak seasons for pinkeye, although it has been reported in all seasons. This is the time when the *M. bovis* organism can be recovered from cattle eyes at the highest rates. The disease occurs in most countries of the world and, although it can occur in all seasons, is most common in summer and autumn. The prevalence and severity of the disease vary greatly from year to year, and it may reach epizootic proportions in feedlots and in cattle running at pasture. Only cattle are affected, the young being most susceptible, but in a susceptible population, cattle of all ages are likely to be affected. There is no mortality, and cases in which there is permanent blindness or loss of an eye are rare. However, the morbidity rate can be as high as 80%, with the peak infection rate at weeks 3-4 of the outbreak. Severe outbreaks can be experienced in winter, especially if the cattle are confined in close quarters such as barns or intensive feedlots.

Susceptibility: Variation within the clinical picture of the disease may be related to the input of the several possible enhancing factors, but may also be due to the susceptibility of the animal to infection and the type of *Moraxella bovis* present in its conjunctival sac. Older cattle are more resistant to infection and disease is more commonly seen in young animals and is usually also more severe in these age groups. Pinkeye is an infectious eye disease of cattle that is found in nearly all breeds throughout the world. However, there is some evidence of breed related differences in susceptibility. Bos indicus breeds, such as Zebu and Brahman, seem to be more resistant than Bos taurus breeds such as Hereford and Angus. There also has been some evidence to suggest that breeds with little or no pigmentation around the eye have a higher incidence of the disease [3].

Risk factors

Agent risk factor: *M. bovis* is a Gram negative diplobacillus with many different strains identified with varying virulence. Virulence is associated with fimbriated strains and which may include β-hemolytic corneotoxic and leucotoxic cytotoxin factors. Other virulence factors include phospholipases, iron acquisition systems, hydrolytic enzymes and proteolytic enzymes. *M. bovis* needs to attach to the bovine corneal epithelium, the first step required for establishment of infection. Two functionally distinct types of pili, termed Q and I, have been identified which enhance pathogenicity. Q pili aid in the attachment of bacteria to the bovine cornea whilst I pili allows for local persistence and maintenance of an established infection on the corneal surface. In different studies it has been demonstrated that *M. bovis* strains expressing Q pili type are more efficient at establishing infection, and are generally more pathogenic than strains expressing only I type pili. Haemolytic strains of *M. bovis* secrete a pore forming cytotoxin which enhances the development of corneal ulcers by lyses of the corneal epithelial cells and host neutrophils. The net result is extensive release of neutrophilic enzymes leading to fragmentation and aggregation of Collagen fibrils the later stages of disease delaying corneal healing. *M. bovis* has high affinity iron-uptake systems, which can remove iron from bovine lactoferrin and transferring, which in turn support protein production and DNA and RNA synthesis.

Host risk factor: Several host defense mechanisms are in place and if disrupted may enhance infection. These include the intact healthy corneal epithelium, tear film and complement system, phagocytic cells and local secretory antibodies. The epithelial surface of the cornea and conjunctiva is generally an effective barrier against disease. Continuous turnover of epithelial cells occurs in five to seven day cycles. As the bacteria must adhere to the epithelial cells in order to establish infection, this continuous cycling inhibits adherence. The tear film represents the most important part of the non-specific defense of the ocular surface as a number of substances that have antimicrobial actions, such as B-lysine, complement, transferring and lactoferrin are transported in tears and washing by may inhibit adhesion. IgA is the major of the ocular surface and tears. Protective IgM and IgG were also detected in bovine lacrimal secretions. The cornea is avascular and combined with its lack of lymphatic drainage, humoral antibodies and or cytotoxic lymphocytes have limited access to the cornea.

Clinically healthy carriers thus play an important role in maintaining the infection in herds by harboring the acteria in the ocular and nasal tracts and carrier animals may be responsible for transferring the disease in to previously unaffected herds. Although the morbidity rate may be high mortality rates are usually negligible [4].

Environmental risk factors: Disease is most common during the summer months when increased UV light exposure results in increased epithelial cell degeneration and epithelial defects which all favour establishment of infection. Transmission of infection may be either direct or indirect. The feeding habit of face flies results in ocular damage and increased eye secretions favoring the growth and therefore spread of infection. Noctuid eye frequenting moths Bacteria may survive for as long as three days on the legs of face flies which make it an ideal transmitter of infection. Increased insect populations are reported to be associated with increased levels of transmission and mechanical irritants and crowding of animals also favours outbreaks. Outbreaks of disease have, however, been described during the winter months as well when excessive wind and dust would be predisposing factors. Any form of stress such as transport and concurrent or prior infection with other infectious agents such as Chlamydophilapecorum, infectious bovine rhinotracheitis, parainfluenza-3 and adenoviruses as well as certain Mycoplasma species may aggravate clinical disease.

Source of Infection and Transmission: Cattle are the reservoir and the organism is carried on the conjunctiva and also in the nares and vagina of cattle. Persistence of the disease from year to year is by means of infected animals, which can act as carriers for periods exceeding year 1. Receptors for I-pili may be found on tissues other than the cornea and facilitate colonization of noncorneal tissue and inapparent infection, and the organism can switch from expression of one pilus type to the other. The same strain can remain on the farm in carrier animals from year to year. *M. bovis* is transmitted by animal handlers or direct contact with eye and nose discharges of infected animals, contact with fomites aerosol and most commonly by mechanical vectors.

Pathogenesis

The primary step in the pathogenesis of infectious bovine keratoconjunctivitis is the adherence by *M. bovis* to corneal epithelium. By scanning electron microscopy, surface epithelium of the bovine cornea is composed of light and dark cells. Light cells are younger and possess surface microplicae (ridges); dark cells are older and relatively devoid of microplicae. Virulent strains of M. bovis selectively colonize the dark cells *in vitro*. “Pit-like depressions” on the surface of these cells are believed to result from activity of a “pitting factor” produced by *M. bovis*. Piliation and elaboration of this “pitting factor” appear to be necessary for *M. bovis* to damage corneal epithelium early in infection but adhesions other than pili may contribute to pathogenicity. Pathogenesis of the disease is influenced by season, mechanical irritation (dust, grass, weeds), host immune response, eye pigmentation, and concurrent presence of pathogenic bacteria, environment, and strain of *M. bovis*. Prerequisites for induction of ocular lesions by *M. bovis* include microbial adhesion to the corneal surface and cytotoxicity, both mediated by several virulence factors. *M. bovis* invades the lacrimal and tarsal glands of the eye, causing keratitis, opacity, uveitis, aqueous flare and corneal ulcers. The bacterium adheres to the cells via its fimbriae and pili proteins, and produces β-haemolysin toxins which lyse the corneal epithelial cells. *M. bovis* also secretes cytotoxic toxin and pathogenic fibrinolysin, phosphates, hyaluronidase and amino peptidases. The bacterial membrane proteins and LPS are also pathogenic. However, this process can be reversible as the corneal epithelium may regenerate once ocular *M. bovis* infection is cleared [5].

Clinical signs

An incubation period of 2-3 days is usual, although longer intervals, up to 3 weeks, have been observed after experimental introduction of the bacteria. Injection of the corneal vessels and edema of the conjunctiva are the early signs and 'are accompanied by a copious watery lacrimation, blepharospasm, and photophobia and, in some cases, a slight to moderate fever with fall in milk yield and depression of appetite. The clinical presentation of the disease, persistence of infection, and rate of progression will vary from animal to animal under field conditions. One or both eyes may be involved. M. bovis exhibits several virulence factors, but only two cause clinical disease: the presence of fimbriae on the bacteria cell surface and the secretion of a beta hemolytic toxin which damages the cornea. The incubation period is usually two to three days, but has extended to three weeks in experimental trials. There are four stages of pinkeye. The disease may resolve at any of these stages while, without treatment the most severe cases will progress through all four stages.

Stage one: Cattle have excessive tearing and increased frequently and there is redness along the eyelids. Cattle will often seek shade, which will decrease their grazing time. Pain associated with pinkeye also decreases their feed intake. Stage I will progress to a small ulcer in the center of the cornea which appears as a small white spot. The cornea develops a slightly cloudy grey appearance due to inflammation. One or both eyes may be affected.

Diagnosis

The usual diagnostic procedures for *M. bovis* infections include: history of the animal, clinical sign, direct microscopic examination of stained conjunctival smears, isolation on enriched media and, identification by morphology and biochemical tests such as catalase test and reaction in litmus milk. If corneal opacification was observed, the inferior subconjunctival sac was swabbed with a sterile Dacron swab. Such eyes were then stained with fluorescein and examined with a flashlight to determine if a corneal ulcer was present; if so, the swabs were further processed as described below. A corneal ulcer was considered to be associated with IBK unless it appeared to be mechanically induced (the pattern of fluorescein staining was linear or stellate), or was present in an eye from which a plant awn was removed. If a mechanically induced ulcer was still present at the next weekly observation, it was considered to be associated with IBK, and in such cases, that observation day was counted as the starting day for the ulcer. Corneal ulcer scores were assigned according to the widest ulcer diameter as measured with a ruler held next to the affected eye: 0=no ulcer; 1=less than or equal to 5 mm; 2=greater than 5 mm; and 3=perforated corneal ulcer. Eyes with ulcers were digitally photographed with a ruler held next to the eye for subsequent determination of the corneal ulcer surface area as described.

Bacterial culturing of ulcerated eyes Ocular swabs collected from eyes of calves with confirmed ulcers were streaked onto 5% sheep blood agar plates that were stored at 4°C during transport back to the laboratory; these plates were subsequently streaked for isolation. Plates were incubated at 35°C for 24 hour and examined for the presence of beta hemolytic. Beta hemolytic colonies were sub cultured and identified using biochemical criteria established for *M. bovis*.

All were showing symptoms like ocular discharge, corneal opacity, conjunctivitis and drastic reduction in feed intake. Sterile ocular swabs were obtained from further the laboratory results were correlated with clinical evidence such as blepharospasm, epiphora, photophobia, chemosis, corneal edema, corneal ulceration and blindness. Characteristic hemolytic colonies were observed on blood agar but no colonies were developed on MacConkey agar plate. The pattern of hemolysis was very peculiar 1-2 mm diameter with corrosion of the agar at the edges of colony. Further, some of the colonies were found to be surface spreading. The organism was gram negative diplococci resembling tumbles and was non motile, catalase and oxidase positive. Gelatin agar was liquefied by the organism within in 24 hrs of stab culture and was able to auto agglutinate normal saline in sugar tubes. Bacteriological examination revealed the production of virulent factors such as hemolytic and proteolysis enzyme production which could have caused opacity or cloudiness of the affected eye. However, fimbriae also help in colonization of the organism in cornea along with capsule, the main virulence factor of *M. bovis*.

Differential diagnosis of ibk and concurrent diseases

Squamous cell carcinoma: Squamous cell carcinoma is an example of a disease that can occur concurrently with IBK. It forms small raised area (cyst) on the nictitans in addition to the corneal ulceration which can be an early indication of squamous cell carcinoma topical and systemic antimicrobial and NSAID therapy should be used followed by re-examination 5 to 7 days later. Removal of the affected tissue, or even enucleation, may be required if feasible.

Bovine iritis (silage eye): Bovine iritis is a disease primarily affecting the iris. Inflammation of the iris or uveitis is the predominant feature. It is common with IBK, however, blepharospasm, photophobia, and lacrimation are often seen together with corneal lesions and, thus, the conditions may be difficult to differentiate. History of big bale silage use and occurrence in winter rather than summer are differences between IBK and silage eye. The affected eye exhibits corneal edema, hyphema, hypopyon, and vascularization migrating from the limbus and clearly appears different from those in IBK. Corneal ulceration is not typical of silage eye; however, in clinical practice, varying degrees of corneal ulceration are often seen.

Control and prevention

Addition of vitamin A in feed may help in preventing the occurrence of IBK as vitamin A helps in the maintenance of the integrity of the surface of the eye, mucous membranes and skin are effective barriers to bacteria and viruses, reducing the risk of eye infections, respiratory problems and other infectious diseases. Fly control is very essential in preventing IBK from occurring as the fly (face fly, stable fly, and house fly) are important vectors of the transmission of IBK pathogenic organisms. The flies usually carry the organisms and cause damage to the eye lining using their proboscis thereby creating a point of entry for the organisms to proliferate and cause IBK infection. Vaccination may help in prevention of IBK as this will reduce the number of susceptible hosts in a herd therefore less outbreak of IBK may occur. Another important concept for working with IBK control is primary, secondary and tertiary prevention practices. Primary prevention options are directed towards preventing exposure to causal factors, such as vaccination and quarantine. Secondary preventive practices intervene after the disease has occurred but before pathology (clinical disease) is obvious, and aim to intervene early in the disease process. Tertiary preventive practices are implemented after clinical disease is diagnosed and attempt to limit the outcome of the disease; it prevents death or decreased productivity.

Vaccination is a commonly used primary preventive practice in veterinary medicine. Depending on the mechanism of immunity, vaccines may offer primary protection by rendering the animal immune to the level of the challenge by *M. bovis* under field conditions. Effective vaccination against a necessary cause (or major component cause) such as *M. bovis* would be the most effective primary preventive practice. However for IBK, multiple commercial vaccines are available with no consensus on vaccine efficacy. It is possible that the vaccines do work on some herds, due to the differences in sufficient causes. As mentioned, ideally disease control programs should use primary prevention practices against a necessary cause as this will be most effective. Due to the short course of disease, there are unlikely to be any realistic secondary preventive practices for IBK unless a herd owner mass-medicates all calves in a group because a threshold number of cases have been identified. In this situation, treatment may occur after the disease process has begun, but before it is detected in a percentage of calves. Tertiary control programs occur after disease has been diagnosed and aim to limit the disease impact, therefore antibiotic therapy can be thought of as a tertiary preventive practice. The cost of IBK on production is substantial. The economic losses occur through decreased growth rates, as affected calves display an average of 17-18 kg decrease in weaning weight. Furthermore, lower performance in post weaning cattle has also been documented.

Treatment

The practical problem of treating several individuals within a herd 2 or 3 times daily has inspired several studies in search of alternate, more cost-effective therapies, for treating both ulcerative keratitis and *M. bovis* in carrier animals. Early treatment of cattle with pinkeye is important, not only for a successful outcome of the individual animal affected, but also to stop the shedding of the bacteria to decrease the risk of transmission to other cattle. Ideally, treatment of IBK will achieve elimination of *M. bovis* infection. If IBK is treated successfully corneal ulcers heal at a constant rate; however, if *M. bovis* is not eliminated from ocular tissues ulcer reoccurrence may follow initial healing. The ideal therapy for a herd problem demands effective one-time dosage and, as such, topically applied preparations, which require frequent administration to maintain therapeutically effective levels in the Precorneal Tear Film (PCTF), cannot be very effective. Their short contact time is reduced further by the presence of ocular discharge and lacrimation, and blepharospasm renders their application difficult. Currently, increased contact time as the result of specific formulation is claimed for several antibiotic preparations including cloxacillin, cephalonium and a penicillin and streptomycin combination.

Subconjunctival treatment: Subconjunctival administration of antimicrobials aims to reduce treatment costs and total dosages of drug while achieving higher ocular drug concentrations. Subconjunctival administration of antimicrobials probably leads to some direct diffusion across the sclera and choroid; alternatively, the drug may gradually leak from the injection site, entering the tear film and eventually the eye via the cornea as if it were applied topically. Although subconjunctival drug dosages are variable, they are generally given in volumes up to 1 ml and typically maintain therapeutically effective tear concentrations for 24 hours or longer. Nonetheless, the elimination of an ocular *M. bovis* infection may be more dependent on achieving therapeutic drug concentrations in infected ocular tissues rather than tears. In calves given a bulbar subconjunctival injection of a conventional (100 mg/ml) oxytetracycline formulation, the antibiotic concentration in tears was above MIC for 24 hours. Although a single subconjunctival dose of a long-acting oxytetracycline formulation achieved tear concentrations above MIC for longer than 72 hours, severe tissue necrosis at the injection site precludes such therapy.

Topical treatment options: Topical administration of antimicrobial formulations has been recommended as a potentially cost effective and less labour intensive method for treatment of IBK. However, topically applied aqueous antimicrobial suspensions have a short tear half-life. Antimicrobials sprayed into the eye may prove irritating and remain only a few minutes before tears wash them away. Although topical application of oxytetracycline aerosol or powder is potentially effective based on MIC data and application is required three to four times daily for 4 to 7 days. Its advantage is that it has a nil withdrawal periods. Two doses of topical benzathine cloxacillin (minimum of 250 mg up to 375 mg, 72 hours apart) should be administered to affected cattle early in the disease process. Both eyes should be treated even if only a single eye is clinically affected and, preferably, unaffected eyes should be treated prior to affected eyes. Procedures should be in place to limit transmission between animals. Again, blanket administration of the drug to all animals simultaneously may not prove a practical or economic means for reducing IBK incidence. Advantages of this therapeutic regime include nil meat and milk withdrawal times.

Systemic treatment: Systemic antimicrobial therapy has been recommended as a means of targeting M. bovis located within lacrimal glands and nasal passages. Drugs administered systemically may enter the eye via the tear film or through the perilimbal or intraocular circulation. Generally, lipophilic drugs achieve higher intra corneal and intra ocular concentrations and are more effective at penetrating the blood tear barrier than hydrophilic drugs. Nonetheless, the attainment of bacteriostatic tear concentrations is not necessarily predictive of efficacy. Elimination of *M. bovis* in calves with IBK has been demonstrated following parenteral treatment with oxytetracycline or florfenicol. Treatment with two doses of long-acting oxytetracycline has been shown to ameliorate clinical signs of naturally occurring IBK through a reduction in *M bovis* ocular infection and consequent decrease in corneal ulcer healing time and recurrence. Similarly, treatment of all herdmates with IM long-acting oxytetracycline followed by daily.

Conclusion and Recommendations

Infectious bovine keratoconjunctivitis is highly contagious, painful ocular disease that affects cattle of all ages and occurs worldwide. The disease is caused by *M. bovis* and there are Pathogen, host and environmental related factors involved that aggravates the disease. It is characterized by excessive tearing and ulceration of the cornea and erforation of the cornea may also occur in severe cases. Mechanical vectors play a vital role in the transmission of the disease. IBK is one of the most economically important diseases in cattle of all ages. Economic losses arising from decreased weight gain in beef breeds, loss of milk production, short term disruption of breeding programmes and treatment costs. Good managmental practice, control of the vectors and vaccination plays a vital role in combating the disease. The status of infectious bovine keratoconjunctivitis in Ethiopia is not clearly understood. But the presence of disease has been reported. Based on the above conclusion the following points are forwarded:

- Studies should be conducted to determine the prevalence, incidence, economic impact and status of the disease in Ethiopia.
- There should be risk mitigation measures to limit the spread and emergence of the disease, thus close collaboration between animal owner and veterinary experts, couple with appropriate education is necessary to develop treatment, control and prevention guide lines.
- Affected animals should be segregated from normal cattle and mechanical vectors should be controlled using insecticides.

- Exposure to environmental irritants such as grass awns and dust should be limited.
- Preventive vaccination should be warranted.

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