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Article Review: The Role of Blood-sucking Insect Vectors in the Spread of Jembrana Disease in Bali Cattle

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ABSTRACT. *Jembrana disease is a contagious disease in cattle and causes enormous economic losses for farmers. This disease, known as bovine lentivirus, was first reported in 1964 in Jembrana District. Jembrana disease specifically attacks Bali cattle at various ages. The cause of this disease is a lentivirus from the Retoviridae family. Jembrana disease transmission occurs by direct contact and is mediated by blood-sucking insect vectors. Not all blood-sucking insects can be vectors for Jembrana disease. Some blood-sucking insects capable of spreading Jembrana disease are Tabanus rubidus flies Culicoides sp mosquitoes, and Aedes lineatopennis, which act as mechanical vectors. As for the Boophilus microplus tick, it is suspected that it acts as a biological vector. Even after re-testing the tick, it could not prove its role as a biological vector for JDV. Prevention and control of this disease can be carried out by vaccination, biosecurity, biosafety, administration of vitamins, isolation of infected livestock, and control of blood-sucking insect vectors by administering insecticides to cages.*

Keywords: Bali cattle, blood sucking insects, jembrana disease

INTRODUCTION

Jembrana disease is one of the infectious diseases that is a big enemy for cattle farmers especially the Bali species in Indonesia. Other animals such as bufallo, goats, sheeps and pigs are immune and do not show any clinical symptoms of the disease, but are capable of carrying the virus that causes Jembrana disease for six months. For experimental animals such as rabbits, mices, white rats, and marmuts are resistant to Jembrana disease. A Bali cattle who recovers from Jembrana disease will be immune to re-infection but at the same time capable of acting as a carrier (Ditkeswan, 2015).

Jembrana disease caused by a lentivirus from the Retoviridae family (Firison *et al*., 2022). The disease was first reported in 1964 in Jembrana District, known as bovine lentivirus (Krisnayanti *et al.*, 2020). The disease has now spread throughout the Bali region and also to various parts of Indonesia such as Sumatera, Java, and Kalimantan. The deaths of more than 60,000 cows per year were observed during the first outbreak and the disease became endemic throughout Indonesia (Kusumawati *et al*., 2015).

Even by 2023, the disease will be spreading again in the West and South Sulawesi provinces (Kementerian Pertanian Republik Indonesia, 2023). By mid-2023, as many as 996

cows were infected with the Jembrana disease virus in South Sulawesi (Beritakotamakassar, 2023) and in West Sulawesia as much as 280 Balinese cows have been confirmed infected by the disease in the period from January to February 2023 (Kompas, 2023).

The Jembrana disease is not a zoonosis but can cause huge economic losses because of its high rate of pain and death. According to Oktarianti & Purnama (2018) stated that Jembrana disease in Bali cattle has a morbidity rate of 60% and mortality of 10%. According to Su *et al.,* (2018) Jembrana disease causes acute disease with a 20% mortality rate in Bali cattle. In the experimentally infected Balinese cattle, the mortality rate was 21% and occurred within 1-2 weeks of infection (Kusumawati *et al.*, 2015). The incubation period of the disease in Bali cattle ranges from 5 to 12 days. Cows infected with membrane disease will show clinical symptoms such as fever, anorexia, lethargy, and swelling of the superficial lymph nodes, bleeding sweat, diarrhea with blood in the stools, and excessive saliva discharge (Firison *et al*., 2022). The disease also causes leukopenia, thrombocytopenia, anemia, increased concentrations of urea in the blood, and a decrease in plasma proteins (Soyi & Kusumawati, 2016).

The transmission of membrane disease occurs through direct contact between sick cows and healthy cows. Vertical transmission does not occur, so cows infected with Jembrana disease are still able to produce normal pedets (Balai Besar Peternakan Batu, 2016). Besides, the disease is also supposed to be chained by blood-sucking insects like flies, ticks and mosquitoes (Horri *et al*., 2023). This transmission is due to the transmission of viruses carried by insects. Some of the insects capable of being vectors of the spread of Jembrana disease are *Culicoides sp* and mosquitoes (Katamtama *et al*., 2018). Experimentally, it has been that Tabanus rubidus has the potential for mechanically transmitting Jembrana disease, whereas *Boophilus microplus* has transovarial potential (Putri & Purnama, 2018). Infection by insects often occurs in cattle extensively kept in repatriation farms (Firison *et al.*, 2022). So the farmer needs to control the vectors that can transmit the Jembrana disease.

This article discusses the role of blood-sucking insect vectors in the spread of Jembrana disease in Bali cattle. It will also discuss the etiology, clinical symptoms and control and prevention of Jembrana disease.

ETIOLOGY

Jembrana disease is a disease caused by the *Jembrana Disease Virus* (JDV) member of the Lentivirus and the Retroviridae family (Astawa *et al.,* 2006). This virus is capable of causing acute and sometimes fatal diseases in infected animals *(*Chen *et al.,* 2000). JDV is a virus with genetic material on single stranded Ribonucleic Acid (ss-RNA), icosahedral shaped with base length 7732 pairs of base (pb) (Indriawati *et al.,* 2013). The virus also has an envelope, measuring 80-120 nm, with four main proteins: p26, p16, p100, and p38-42-45 (Direktorat Kesehatan Hewan, 2014). Based on reverse transcriptase activity, the virus gets into the retrovirus group. The JDV reverse transcriptase gene has an amino acid sequence similarity of about 68% to the bovine immunodeficiency virus (BIV) so BIV is called the bovin lentivirus type 1 and the membrane virus is called bovine lenvirus type 2 (Berata, 2015).

Jembrana Disease Virus only attacks Bali cows, while other breeds of cows are more resistant to the virus (Suwiti, 2009). JDV is capable of infecting by sticking to the surface of the target cell and then inserting RNA genetic material into its target cell. Viral RNA will undergo reverse transcription to become cDNA. Viral DNA will migrate from cytoplasm to nucleus, and the enzyme integrase will

integrate viral DNA into the host cell DNA to form a provirus. The provirus (DNA) can be found in the blood of target animals with infected cells, e.g. in Bali cattle (Irwanto *et al.,* 2021). The provirus that is formed will remain in the body of the cow, so that the cow will be a carrier. When these carrier cows are in an unhealthy condition and humoral immunity begins to decline, it is suspected that the cDNA of the virus can turn into re-active and can infect the surrounding animals (Krisnayanti *et al*., 2020). JDV Provirus DNA has a typical retroviral genomic structure containing *gag*, *pol*, and *env genes* with the presence of long terminal repeats (LTRs) in 5′ and 3′ terminals. The env gene encodes the proteins that are present in the outermost parts of the Jembrana disease virus, namely the Surface Unit (SU) and Transmembrane proteins. (TM). SU proteins play an important role at the beginning of the replication process and interact with host cells by binding Jembrana virus particles on the host cell surface (Indriawati *et al.,* 2013). Besides, the virus also contains a number of regulating genes that encrypt proteins, some of which are involved in regulating the expression of viral genes (Marchand *et al*., 2019).

The Jembrana disease virus has a sensitivity to chloroform as well as ether and is resistant to deosicolate sodium. (1:1000). The virus is also inactive by formalin and sensitive to extreme pH. (3.0 dan 12.0). The virus is immediately denaturated when heated to a temperature of 550C for 15 minutes. The virus is also resistant to antibiotics. JDV is able to stay in the blood and tissues of the patient for a long time. The virus is only able to grow on experimental animals, especially Bali cows. The virus is incapable of growing and developing in primary cell breeding such as lungs, kidneys, testicles, spleen, fetal muscles, peripheral blood macrophages as well as Vero (vero-E6 and CV1) (African green monkey), embryonic bovine tubinate (EBTI), Mardin Darby bovine kidney (MDBK), HeLa and baby hamster kidney (BHK21) and does not cause cytopathogenic eect (CPE) (Berata, 2015; Ditkeswan, 2015).

CLINICAL SYMPTOMS

Cows suffering from Jembrana disease will show clinical symptoms such as high fever, blood diarrhea in stools, excessive salivation and swelling of the lymph glands (Firison *et al*., 2022). According to de Pablo-Maiso *et al.,* (2018) stated that the clinical symptoms of Jembrana disease in cows are fever, anorexia, frostbite, lymphadenopathy, and lymphopenia. In addition, they found symptoms such as blood stains on the skin so it looks like bleeding sweat and also this disease causes death due to an operational infection because the Jembrana virus is able to lower the immune system, so the body becomes vulnerable and susceptible to disease (Krisnayanti *et al*., 2020). Miswati *et al.,* (2017) in his research proved that as many as 7 of the 9 cows in the Kinali district, Western Pasaman district confirmed positive infection with Jembrana disease based on the results of testing using the method of Polymerase Chain Reaction (PCR). The cows showed clinical symptoms such as fever, decreased appetite, hypersalivation, hyperlacrymation, swelling of superficial lymph glands and blood sweating. While the results of the investigation of the death of Bali cattle by Anggy & Srihanto, (2019) in the Pubian district, Lampung Tengah district showed that as many as 15 Bali cattle who had a confirmed positive death were infected with Jembrana disease based on results of laboratory examinations. Prior to death, the cows showed clinical symptoms such as anorexia, diarrhea, hypersalivation and collapse. Jembrana disease causes acute disease with a mortality rate of 20% in Bali cattle (Su *et al.,* 2018). In the experimentally infected Balinese cattle, the mortality rate was 21% and occurred within 1-2 weeks of infection (Kusumawati *et al*., 2015).

The incubation period of the disease in Bali cows ranges from 5 to 12 days.

A typical symptom of Jembrana disease is blood sweating, and 93% of all cases have been. Bleeding sweat can occur in the flank, back, abdomen, legs and scrotum. But the symptoms of bleeding sweat did not appear in experimentally infected cattle in a cage without bloodsucking insects. This proves that the bleeding sweat in the Balinese cows infected with Jembrana disease has something to do with blood-preparing insects (Soesanto *et al*., 1990). Jembrana disease can lead to a decrease in the number of platelets (Direktorat Kesehatan Hewan, 2014). As a result of the detection, bleeding occurred near all the organs and even the injury was crushed by the bite of a bloodsucking insect, resulting in symptoms such as bleeding sweat (Ditkeswan, 2015). In addition to causing thrombocytopenia, the disease also causes leukopenia, anemia, increased concentration of urea in the blood, and a decrease in plasma protein (Soyi & Kusumawati, 2016).

Table 1. Clinical symptoms of Jembrana disease according to several references

No	Clinical Symptoms	References
1	High fever, blood diarrhea in stools, excessive salivation and swelling of the lymph glands	Firison *et al*., 2022
2	Fever, anorexia, frostbite, lymphadenopathy, and lymphopenia	de Pablo-Maiso *et al*., 2018
3	Fever, decreased appetite, hypersalivation, hyperlacrymation, swelling of superficial lymph glands and blood sweating	Miswati *et al*.,2017; Krisnayanti *et al*., 2020
4	Bleeding sweat can occur in the flank, back, abdomen, legs and scrotum	Soesanto *et al*., 1990
5	Anorexia, diarrhea, hypersalivation and collapse	Anggy & Srihanto, 2019
6	Thrombocytopenia, leukopenia, anemia, increased concentration of urea in the blood, and a decrease in plasma protein	Direktorat Kesehatan Hewan, 2014; Soyi & Kusumawati, 2016

THE TRANSMISSION OF JEMBRANA DISEASE BY VECTOR ATTACKS OF BLOOD SUCKER INSECTS

During the fever phase, JDV was found in both blood cells and blood plasma and showed high titers of up to 10^8 ID_{50}/ml. Besides, the virus can also be detected in secreted fluids such as saliva, milk, and nasal fluids. The transmission of membrane disease occurs through direct contact between infected animals to vulnerable animals. This is demonstrated by research by Soeharsono *et al.* (1995) that there is transmission of Jembrana disease through direct contact between infected cows to other vulnerable cows via the conjunctive, intranasal and oral route. Besides, the transmission of Jembrana disease is suspected by bloodsucking insects (Hartaningsih *et al*., 1993). Hori et al (2023) states that the transmission of Jembrana disease in the field can occur through blood sucking insects. Besides that Firison (2022) states that Jembrana disease can be transmitted directly through interaction between sick and healthy livestock, or indirectly through intermediaries. Kusumawati *et al*., (2014) also stated that, because of the high virus titers in the blood of the victim, the possibility of transmission can also occur mechanically through bloodsucking insects. This is in line with Irwanto *et al.* (2021) statement that membrane disease can be contagious through contact transmitted by bloodsucking insects. This transmission occurs due to the transmission of the virus causing the disease of the membrane carried by blood sucking insects after biting and sucking the blood of infected cattle (Sumbarprov, 2017). Transmissions

transmitted by bloodsucking insect vectors often occur in extensive cattle breeding. Maradesa *et al.,* (2022) also stated that the maintenance system is extensively one of the factors that cattle are susceptible to diseases as well as to vectors like flies.

Not all bloodsucking insects are capable of being vectors of Jembrana disease. Some of the bloodsucking insects that are believed to be capable of spreading the disease are flies, ticks and mosquitoes (Horri *et al.,* 2023). Insects capable of being vectors of the spread of Jembrana disease are *Culicoides sp* and mosquitoes (Katamtama *et al*., 2018). Experimentally, it has been that *Boophilus microplus* tick has potential in transovarial transmission of jembrana disease (Dennig, 1977). Inside the body of *Boophilus microplus*, the virus multiplies, causing Jembrana disease, before spreading to other vulnerable cattle. Tick *Boophilus* microplus is a species of insect that sucks blood in cattle (Rustam *et al.,* 2021). In addition to sucking blood, it can also act as a vector of the spread of various diseases in cattle (Ariman *et al.,* 2021). However, after a re-testing of *Boophilus microplus*, a result was obtained that it could not prove the role of the caplak as a biological vector of JDV. Field observations also did not provide any indication that every case of Jembrana disease has always been associated with *Boophilus microplus* (Sulistyana and Putra, 1993).

In addition, it has been experimentally that *Tabanus rubidus* flies are potentially mechanically transmitting Jembrana disease (Putri & Purnama, 2018). The mechanical transmission of the disease occurs because the insect/anthropod blood sucker is disrupted in the interrupted feeding of the infected animal and then sucking the blood back to the other healthy animal. That's when there's mechanical transmission of Jembrana disease as a result of virus contamination in the mouth of the bloodsucking insect. In addition to these vectors, there are also other vectors such as mosquitoes *Aedes lineatopennis* (Guntoro *et al*., 2018) and *Culicoides sp* (Katamtama *et al*., 2018).

Mosquitoes have an average maximum flight distance of 50 m to 50 km, depending on their species and are able to be carried even further by the wind (Verdonschot & Besse-Lototskaya, 2014). As for flies, they can fly up to eight kilometers (Puspitarani *et al.,* 2017). The viral disease membrane has a low viability outside the body so it requires a transmission process in a short time. This is in line with Hastutiek & Fitri (2013) statement that mechanical transmission must take place within a short period of time in order to be able to effectively infect, due to the very short resistance of some disease-causing agents while in the carrier vector. Therefore, mosquitoes and flies can play an important role in the transmission of Jembrana disease. The close distance between sick and healthy cows is one of the key factors in the spread of the disease.

PREVENTION AND CONTROL

Preventive and control measures must be taken promptly to prevent the spread of the disease and prevent the economic losses caused by the disease. For the prevention and control of Jembrana disease in threatened areas can be done with vaccination (Margawati, 2020). The Jembrana disease vaccine has been successfully produced from plasma and spleen (Direktorat Kesehatan Hewan, 2014). In addition, intensive maintenance, biosecurity, biosafety, multivitamin administration, and also control of vectors capable of spreading membrane disease are needed (Firison *et al*., 2022). Fighting flies and mosquito vectors can be done by spraying insecticides into the cage. In addition to the elimination of pathogen vectors, it is also necessary to isolate infected animals so that they do not become a source of transmission of

Jembrana disease to other vulnerable animals (Putri *et al.*, 2019).

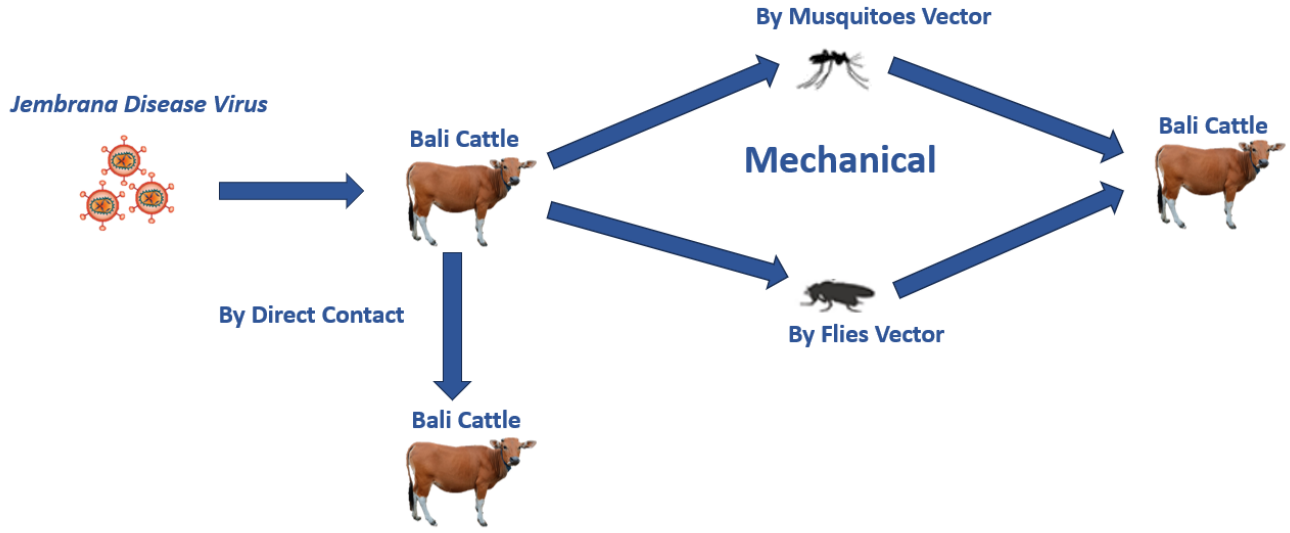

Figure 1. Mechanism of Spread of Jembrana Disease in Bali Cattle (*source: private document*)

CONCLUSION

Jembran disease is a disease caused by Lentivirus in the family Retoviridae. Cows suffering from membrane disease will show clinical symptoms such as high fever, blood diarrhea in the stools, lymphadenopathy, Lymphopenia, and bleeding sweat. In addition, membrane disease can lead to thrombocytopenia, leukopenia, anemia, increased concentration of urea in the blood, and a decrease in plasma proteins. The disease is transmitted through direct contact and also by vectors of bloodsucking insects. Some of the bloodsucking insects that are believed to be capable of spreading Jembrana disease are *Tabanus rubidus* flies and *Culicoides sp* mosquitoes as well as Aedes lineatopennis that act as mechanical vectors. The *Boophilus microplus* tick is supposed to act as a biological vector. Even after re-testing the tick could not prove its role as a biological vector of the JDV virus.

CONFLICT OF INTEREST

All authors declare that there is no conflict of interest in the research.

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