

Akabane virus infection

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Summary

Akabane virus is a *Culicoides*-borne orthobunyavirus that is teratogenic to the fetus of cattle and small ruminant species. Depending upon the stage of gestation at which infection occurs, and the length of gestation of the mammalian host, a range of congenital defects may be observed. The developing central nervous system is usually the most severely affected, with hydranencephaly and arthrogryposis most frequently observed. Less commonly, some strains of Akabane virus can cause encephalitis in the neonate or, rarely, adult cattle. Akabane viruses are known to be widespread in temperate and tropical regions of Australia, Southeast Asia, the Middle East and some African countries. Disease is infrequently observed in regions where this virus is endemic and the presence of the virus remains unrecognised in the absence of serological surveillance. In some Asian countries, vaccines are used to minimise the occurrence of disease.

Keywords

Akabane virus – Arthrogryposis – Congenital defect – *Culicoides* – Hydranencephaly – Orthobunyavirus.

The virus

Akabane virus is an arbovirus that was first isolated in Japan in 1959 (1, 2). Taxonomically this virus is classified in the genus *Orthobunyavirus* in the family *Bunyaviridae*. Historically, Akabane virus was grouped in the Simbu serogroup based on serological relationships with other orthobunyaviruses (3). This serological classification remains useful as it reliably describes closely related orthobunyaviruses and often provides an insight into their *in vivo* biological characteristics. For example, the most recently discovered virus in this group, Schmallerberg (4), has a number of biological properties that are similar to those of Akabane virus. The term 'Akabane disease' has been used to describe the clinical syndrome resulting from *in utero* infection with Akabane virus but this can be misleading because some or all of the elements of the syndrome, e.g. congenital arthrogryposis and hydranencephaly, can be caused by other viruses, especially other orthobunyaviruses. The tripartite segmented genome of the bunyaviruses facilitates the development of reassortants, with segments of Akabane virus being found in closely related Simbu viruses such as Tinaroo (5), though this virus is not recognised as being pathogenic in nature.

Arthropod vectors

The principal vectors of Akabane virus are small biting midges (or gnats) belonging to the genus *Culicoides* (6, 7). Other species within this genus are vectors of bluetongue virus, and some species are competent vectors of both viruses. Multiple *Culicoides* species are often present in the same place at the same time, but while the occurrence of vectors of Akabane virus in a region increases the likelihood that there might be a competent vector(s) of bluetongue virus, this cannot be assumed. There is also a high probability that vectors of bluetongue virus will be competent vectors of a Simbu virus. For example, in Europe, following the emergence of Schmallerberg virus, the proven vectors of bluetongue viruses have also been shown to be efficient vectors of Schmallerberg virus. From an epidemiological perspective, an important consideration is the vector competence and infection rates of *Culicoides* with Akabane virus. Compared to bluetongue viruses, the Simbu viruses are transmitted to mammalian hosts with a high level of efficiency, in part due to the high virus infection rates detected in *Culicoides* and the very large number of insects that may attack an animal. The distribution of Akabane virus antibodies in cattle can be considered a

very sensitive indicator of the presence of a competent population of *Culicoides*, even if the numbers in a region are small. Although Akabane virus has been isolated from mosquitoes (1, 8), these are not considered to be true vectors of the virus.

Geographical and seasonal distribution

Akabane virus has been reported in a number of countries on the African continent, the Middle East, Southeast Asia and Australia (9). It is considered likely that Akabane virus or other Simbu viruses are also present in neighbouring countries in these regions. Its presence is suspected in many other countries in the tropical and temperate zones, especially those where bluetongue is reported. Within a country, the distribution of the virus is absolutely restricted to that of the insect vector (10). In countries with a temperate climate, there is also a distinct seasonal pattern of virus transmission, coinciding with warm, moist summer and autumn months. This seasonal pattern is also a consequence of the abundance of the insect vector. There is a critical population density required before virus spread can occur. Vector numbers begin to increase in the late spring and early summer, usually peaking in early autumn. Even in tropical and subtropical regions, there is a tendency towards seasonal transmission, with the highest infection rates in the summer months. In temperate regions, transmission ceases with the onset of very low temperatures and the first frosts, while in tropical regions transmission rates decline with the onset of the periods of lower rainfall.

Mammalian host range

Akabane virus infects a wide range of domesticated ruminants and wildlife species, especially bovids (10, 11, 12, 13). In endemic areas there is a high prevalence of antibodies in cattle, buffalo, sheep, goats and also horses. In Chinese Taipei, a high prevalence of Akabane virus infection has been reported in pigs held outdoors (14). However, it is not clear whether pigs play a role in the maintenance of Akabane virus in nature and no disease has been described. Human infection has not been reported. In Australia, even in areas where there is frequent virus transmission, infection of marsupials has not been detected.

Pathogenesis

The onset of viraemia with Akabane virus generally occurs one to six days after infection and may last four to six days

before antibodies to the virus are detected and the virus is cleared. Antibodies are detectable by serological tests from about 14 days after infection. The virus may persist for a considerably longer period in the developing fetus and clinical signs are usually not observed for months until an affected fetus is aborted or reaches term.

The outcome of infection of a susceptible mammalian host is determined almost exclusively by its age and reproductive status. In endemic areas, there is often annual transmission, and young animals become infected in their first year of life soon after maternally derived antibodies decline. Postnatal infection with most strains of Akabane virus is asymptomatic.

Akabane virus is a potent teratogen and almost exclusively affects the developing fetus. Infection of the female during pregnancy can result in a range of severe fetal defects affecting the limbs (particularly arthrogryposis) and the central nervous system (CNS). These have been described in several reviews (9, 15, 16). Defects in the brain range from small cystic defects (porencephaly) to almost complete absence of the cerebral hemispheres, with replacement by fluid-filled sacs (hydranencephaly). Infection of calves around the time of, or soon after, birth may cause encephalitis.

The type of abnormality, and also the incidence, is influenced by the stage of gestation at which the dam is infected. Soon after being bitten by the insect, the adult develops a viraemia (7) and the virus crosses the placenta to infect the developing fetus. In most species, the impact of the virus is greatest in mid-gestation. There is no documented evidence of damage to the conceptus following infection in the very early stages of pregnancy (the first three weeks in sheep and goats; the first two months in cattle). The most susceptible stages of gestation in small ruminants range from 28 to 56 days (especially 28 to 36 days) (17, 18, 19, 20, 21) and in cattle from three to six months (22, 23). In the latter stages of gestation (after 60 days in small ruminants and the last two months in cattle), the incidence of abnormalities declines to a very low level.

In cattle, the major defects involve the brain and spinal cord; the effects on skeletal muscle are mainly secondary, although a primary viral myositis sometimes occurs. The damage is evident mainly as hydranencephaly, porencephaly and arthrogryposis. In cattle, there is a successive progression of different defects as a result of the long gestation period (23). Infection of the fetus between approximately 80 and 105 days of gestation results almost exclusively in the development of hydranencephaly and porencephaly. The lesions are markedly more severe in fetuses infected early on in this period and gradually decline in severity until abnormalities are no longer grossly apparent. Arthrogryposis occurs following infection between approximately 105 and

170 days of gestation. There may be a small number of calves born with both mild hydranencephaly and arthrogryposis due to infection around 100 to 120 days of gestation. Calves with arthrogryposis as a result of infection between 100 and 150 days of gestation are more severely affected, with abnormalities involving multiple joints on several, or even all four, limbs. Defects are less severe following infection later in gestation and may involve a single joint on one limb. In sheep and goats, due to the shorter gestation and shorter period of susceptibility, there is usually a combination of severe defects of the limbs with gross CNS lesions (15, 17, 19, 20, 21). In addition to lesions in the CNS, in sheep and goats there may be an impact on the development of other organs, such as pulmonary hypoplasia (21). When calves are infected close to term, encephalitis can occur and may be clinically apparent at birth (23).

Clinical signs

Although Akabane virus infects most ruminant species, disease outbreaks of significance are mainly seen in cattle. Sheep and goats are usually raised in areas where the vector is either absent or uncommon. Further, small ruminants are usually bred at the end of or after the vector season, so there is little risk of fetal infection.

Akabane infection of adult animals is usually asymptomatic (7). Some strains of Akabane virus have been associated with encephalitis in newborn calves (24), but this is generally uncommon. The Iriki strain of Akabane virus in Japan and Korea has been associated with cases of encephalitis in adult cattle (25, 26, 27).

The greatest impact of the virus is on the developing fetus. The range of defects observed will vary depending on herd management and the time of virus transmission. In year-round calving herds (such as some temperate and subtropical dairy herds), the full range of congenital defects may be seen, while in a seasonal calving herd with a very restricted mating period, only one type of abnormality may be noticed. There have been a number of detailed descriptions of the range of calf defects that may occur following infection with Akabane virus (22, 23, 28, 29, 30). The descriptions that follow relate to a herd with a prolonged (or year-round) calving period.

The first indication of an outbreak of Akabane infection is abortion of abnormal fetuses at between four and six months of gestation. Infected fetuses may appear to be grossly normal but careful examination may detect fixation of joints. Removal of the calvarium at post-mortem examination will often reveal severe hydranencephaly.

The first calves that are born during the outbreak will have been infected late in gestation and may show signs

of acute encephalitis, such as flaccid paralysis of the legs, hyperextension of joints and difficulty in standing.

Calves which are infected in the fifth or sixth month of gestation may be born with arthrogryposis and have grossly apparent deformities. Those infected in about the sixth month of gestation may have only one or two joints affected on a single limb, whereas calves that have been infected a little earlier, in the fifth month of gestation, are likely to have more severe lesions, involving multiple joints on all limbs and perhaps abnormalities of the spinal column such as kyphosis or lordosis. Dystokia is common in cows delivering these calves, most of which are stillborn. Many require embryotomy or Caesarian delivery. A small proportion of cows die as a result of obstetric complications, while others suffer permanent infertility. Calves with relatively mild lesions, involving one or two joints on a single limb, are usually born alive and are able to stand.

About six weeks into the outbreak there will be few cases of arthrogryposis but the incidence of calves with hydranencephaly will increase. Those calves that do have arthrogryposis may also have lesions of porencephaly. The severity of hydranencephaly then increases during an outbreak, leading to severe hydranencephaly, with virtual absence of the cerebral hemispheres (23). Most calves delivered in the last four to six weeks of an outbreak will have severe hydranencephaly. Some are stillborn but many will be born alive. The lesions in the brain result in a range of behavioural abnormalities. These animals are usually blind, unaware of their surroundings and wander aimlessly. These defects are usually life-threatening and most affected animals die soon after birth if close supervision and care are not provided. However, due to the nature and severity of the abnormalities, most surviving animals are euthanased.

The brain stem appears to be grossly normal, even when there is complete absence of the cerebral hemispheres. In Akabane cases in cattle, the cerebellum is consistently intact and apparently normal. Torticollis, scoliosis, and brachygnathism are sometimes observed but are more frequent in small ruminants.

In lambs and kids born to sheep and goats that are pregnant during the vector season, congenital defects can be observed following infection 28 to 56 days into pregnancy. However, at term, the chronological progression of defects observed in cattle is not usually apparent. Lambs and kids are likely to show a range of defects of both the skeletal and central nervous systems. Severe hydranencephaly and severe arthrogryposis may be seen affecting the same animal. Other developmental abnormalities such as pulmonary and thymic hypoplasia may also occur (21).

The incidence of congenital abnormalities is determined by the stage of gestation at which the fetus is infected and also by the strain of virus. In cattle, the incidence of defects may be as high as 50% (31) if they are infected at the most critical stages of gestation (three to four months). If they are infected in the fourth month, incidence is typically around 25% and this declines to 5% in those infected in the seventh month (23). In sheep, at the most susceptible stages of gestation, the incidence of fetal infection can range from 15% to 80% depending upon the strain of virus (19).

Epidemiology

The occurrence of Akabane disease is determined entirely by the distribution of the insect vector. This is strongly influenced by climatic conditions, especially temperature and, to a lesser extent, rainfall. *Culicoides* species typically have a well-defined geographical distribution (7, 10). Within that range, Akabane virus transmission occurs frequently and usually each year. Consequently, there is a high level of population immunity and most animals are immune before reaching breeding age. There is also a distinct seasonal pattern of spread. Within an endemic area, midge numbers increase during the late spring/early summer and peak in autumn. There is typically a lag phase between the first occurrence of the midge and virus transmission, which commences once insects become abundant. Conversely, there is a rapid decline in transmission with the onset of cold weather and a cessation with the first frost.

Outbreaks of Akabane disease occur after environmental conditions that are markedly different from usual patterns. For example, prolonged mild moist conditions in autumn can result in spread of the midge well beyond its usual range into areas where there are large populations of susceptible livestock (30). Even a limited period of transmission can result in a significant disease outbreak. Adverse climatic conditions can also result in reduced midge activity and virus transmission within an endemic area. This results in a reduced level of population immunity and the opportunity for an increase in the number of susceptible animals that will reach breeding age before the next or even a subsequent vector season (32). Inevitably, normal transmission patterns return, with the concomitant birth of deformed calves delivered by heifers or young cows (23).

As a result of the interaction between vector and climatic factors, outbreaks of Akabane disease usually follow a well-defined distribution, occurring in areas adjoining regions where vectors are endemic. Exceptions to this pattern occur when pregnant cows are either permanently moved into a vector area or are held temporarily in a vector area and then returned to their home property (31). Because of the distinct seasonal pattern of virus transmission, there is

also a clear pattern of seasonal occurrence of disease. In most areas where outbreaks occur, clinical cases in cattle are first observed in early winter and reach a peak in the early spring months (30).

Pathology

Akabane infection may be suspected from the seasonal clustering of the birth of large numbers of calves with congenital defects and with highly suggestive gross pathology and histopathology. There may also be an increase in cases of neurological disease in newborn calves. Calves with flaccid paralysis as a result of infection late in gestation have histological lesions of a non-suppurative polio-encephalomyelitis (22, 23).

In calves with arthrogryposis, apart from the fixation or severely restricted range of movement of joints, there are few other grossly detectable changes. There are, however, microscopically detectable severe degenerative changes in the motor horns of the spinal cord (22, 23). In some cases, degenerative changes are also apparent in the skeletal muscle.

When calves are stillborn or show behavioural changes, grossly apparent defects are likely to be apparent in the brain. These can vary from small cystic lesions to the virtual absence of the cerebral hemispheres and replacement with fluid-filled meningeal sacs (22, 23, 30, 33). Histopathology on Akabane cases with severe hydranencephaly is unrewarding and of minimal diagnostic value. There will be an absence of large areas of brain surrounded by tissues with relatively normal architecture. In cattle, the cerebellum is rarely, if ever, affected, a useful differential feature to distinguish Akabane virus from other congenital infections such as bovine viral diarrhoea virus. However, gross cerebellar lesions may be detected in calves that have been infected with other orthobunyaviruses such as Aino and Schmollenberg.

If a fresh aborted fetus is found, depending on the age of the fetus and the time since it was infected, gross lesions may not be apparent. A range of acute, necrotic, degenerative changes may be detected and perhaps also a mild to moderate non-suppurative encephalomyelitis suggestive of a viral infection. The lesions can be detected in all parts of the CNS, with perivascular cuffing, neuronal degeneration and cavitation of the brain, and neuronal degeneration in the motor neurones of the spinal cord. Muscular dystrophy may also be observed.

Diagnosis

Akabane disease should be considered when there is an outbreak of congenital defects in cattle, sheep or goats

commencing in the winter months and extending to spring. The gross pathology should provide a strong index of suspicion. An aetiological diagnosis and confirmation of Akabane infection depends on the detection of specific antibody in blood or fluids of fetuses and affected neonates that have been deprived of colostrum. Most stillborn or aborted fetuses and calves that are born at term mount a specific antibody response to the virus (28, 34). Testing can be carried out systematically by initially examining fetal fluids or pre-colostral serum to determine the IgG levels. An elevated IgG level will incriminate an infectious agent (35, 36) and Akabane virus-specific serology can then be undertaken. A range of serological tests have been used to detect antibodies to Akabane virus but virus neutralisation tests and enzyme-linked immunosorbent assays are most frequently used.

Virus detection by virus isolation (37) or polymerase chain reaction (PCR) (38) may be considered if a fetus has been aborted in the early stages of pregnancy. Using real-time PCR, it may be possible to detect residual RNA in affected tissues of neonates (38). Testing of swabs taken from the surface of cotyledons of the placenta may also give positive results. Maternal serology is of value only in regions where the virus is not endemic. In these situations, positive maternal serology will raise the index of suspicion, while a negative result will convincingly exclude Akabane virus as the aetiological agent.

Germplasm: semen and embryos

Virus has not been detected in the semen of bulls experimentally infected with Akabane virus (39). There is no evidence that Akabane virus can infect the developing embryo and washing techniques are considered to be a safe approach to ensuring that Akabane virus is not inadvertently transmitted by embryo transfer (40).

Control

The impact of Akabane virus is best controlled by strategic vaccination of susceptible animals prior to the time of potential exposure to vector activity. A live attenuated vaccine has been used in Japan (41) and inactivated vaccines have been used in Australia, Japan and Korea (42, 43, 44). Inactivated vaccines have the advantage of being suitable for the emergency vaccination of pregnant animals. Alterations to herd or flock management, such as delaying mating or changing the calving period from spring to autumn, can be used to prevent outbreaks if there is warning of impending vector activity. Vector control measures, such as covering breeding sites and using insect repellents and insecticide treatments may be effective for short periods, but are usually ineffective in preventing fetal infection over a period of more than a few days.

Infection par le virus Akabane

P.D. Kirkland

Résumé

Le virus Akabane est un orthobunyavirus transmis par les moucheron piqueurs du genre *Culicoides* ; il est tératogène pour les fœtus de bovins, de caprins et d'ovins. Il peut causer un certain nombre de malformations congénitales dont l'apparition dépend du stade de gestation au moment de l'infection et de la durée de la gestation de l'espèce mammifère hôte. C'est généralement le système central nerveux en développement qui est le plus gravement atteint, une hydranencéphalie et une arthrogrypose étant les affections les plus fréquentes. De manière moins fréquente, certaines souches du virus Akabane sont responsables d'encéphalite chez le veau nouveau-né ou plus rarement chez le bovin adulte. Les virus Akabane sont très présents dans les régions tempérées et tropicales de l'Australie, de l'Asie du Sud-Est, du Moyen-Orient et de certains pays d'Afrique. Les maladies dues à ce virus étant rarement observées dans les régions où celui-ci est endémique, sa présence peut passer longtemps inaperçue si aucune surveillance sérologique n'est exercée. Certains pays asiatiques pratiquent la vaccination pour minimiser l'incidence de l'infection.

Mots-clés

Akabane – Arthrogrypose – *Culicoides* – Hydranencéphalie – Malformation congénitale – Orthobunyavirus.

La infección por el virus de Akabane

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Resumen

El virus de Akabane es un orthobunyavirus transmitido por *Culicoides* que tiene efectos teratógenos en los fetos del ganado bovino y de pequeños rumiantes. Dependiendo del estadio de gestación en el que se produzca la infección y de la duración del embarazo en el mamífero hospedador se podrán observar diversas anomalías congénitas. Lo que en general resulta más gravemente afectado es el desarrollo del sistema nervioso central, y las anomalías más comunes son la hidranencefalia y la artrogriposis. A veces, con menos frecuencia, algunas cepas del virus causan encefalitis en el vacuno neonato o, más rara vez, en el adulto. Se sabe que los virus de Akabane están muy extendidos en regiones templadas y tropicales de Australia, Asia Sudoriental, Oriente Medio y algunos países africanos. Resulta infrecuente observar la enfermedad en regiones donde el virus es endémico, por lo que en ausencia de vigilancia serológica su presencia pasa desapercibida. En algunos países asiáticos se utilizan vacunas para reducir al mínimo los casos de enfermedad.

Palabras clave

Akabane – Anomalía congénita – Artrogriposis – *Culicoides* – Hidranencefalia – Orthobunyavirus.



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