Introduction

Prenatal viral infections have been identified as teratogens in cattle. Cerebellar hypoplasia is caused by prenatal infection with bovine viral diarrhea-mucosal disease (BVD-MD) virus (KAHRS et al., 1970); hydranencephaly may be linked to prenatal infection of bluetongue virus (OSBURN et al., 1971).

Since the big outbreaks of Akabane disease in 1972, moderate outbreaks of central nervous system (CNS) defects in calves have occurred several more times (HAMANA et al., 1992). Kagoshima has a big cattle industry and because it is located in the southern part of Japan, calves suffer from these arbovirus-mediated CNS defects. We have collected more than 2,500 cases of calves with congenital defects and estimate that more than 60% of them can be classified as defects caused by Akabane, Chuzan and Aino viruses. Fortunately, we were able to investigate the first case of each defect during outbreaks.

An outbreak of hydranencephaly and cerebellar hypoplasia affecting over 2,000 calves was observed elsewhere in Kyushu, during November 1985 and June 1986. The calves were generally weak and paretic at birth and could not suck milk by themselves. Nervous signs were found in many cases. Chuzan virus was confirmed to be the causative agent.

During November 1995 and March 1996, an outbreak in over 1,500 calves featured cerebellar hypoplasia and torticollis. Most of them showed nervous signs, including ataxia, tetany, swimming movement, opisthotonus or circular walking. Aino virus was strongly suspected serologically to be a causative agent, and this was confirmed later.

The origin of these arboviruses is said to be the tropical jungle. Therefore, the possibility of their occurrence is high in tropical areas including Yap.

Key words: arboviruses, calves, congenital defects, hydranencephaly

Katsumi HAMANA

Abstract

Repeated outbreaks of central nervous system (CNS) defects with or without arthrogryposis in calves have occurred in Kagoshima, Japan. They were caused by arboviruses, especially Akabane, Chuzan and Aino virus.

The outbreaks of abortion, premature birth, stillbirth and congenital arthrogryposis-hydranencephaly syndrome in Japan during 1972-1973 and 1973-1974 affected over 40,000 fetuses and calves. In 1975, Akabane virus was confirmed to be a causative agent. Akabane disease has occurred several times thereafter in Kagoshima.

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Since the big outbreaks of Akabane disease in 1972, moderate outbreaks of central nervous system (CNS) defects in calves have occurred several more times (HAMANA et al., 1992). Kagoshima has a big cattle industry and because it is located in the southern part of Japan, calves suffer from these arbovirus-mediated CNS defects. We have collected more than 2,500 cases of calves with congenital defects and estimate that more than 60% of them can be classified as defects caused by Akabane, Chuzan and Aino viruses. Fortunately, we were able to investigate the first case of each defect during outbreaks.

The outbreaks of abortion, premature birth, stillbirth and congenital arthrogryposis-hydranencephaly syndrome in Japan during the summer through winter of 1972-1973 and 1973-1974 affected over 40,000 fetuses and calves. In 1975, Akabane virus was confirmed to be a causative agent (KUROGI et al., 1975).
Over 2,000 calves with congenital hydranencephaly-cerebellar hypoplasia were born in several parts of Kyushu during November 1985 and June 1986 (HAMANA & TAURA, 1988). Chuzan virus was confirmed as the cause.

During November 1995 and March 1996, an outbreak in over 1,500 calves featured cerebellar hypoplasia and torticollis was investigated and Aino virus was found as the cause (HAMANA et al., 1997).

**Materials and Methods**

Cases of calves with congenital defects have been continuously collected in the Kagoshima area. For the defective calves, the breed, sex, date of birth, condition at birth, clinical findings and prognosis were investigated. At necropsy, pathological examination was performed. For the dams, feeding and management practice and vaccination by Akabane virus were investigated. The serums obtained from dams and calves were frozen and later virologically analyzed.

**Results and Discussion**

**Akabane virus**

During the outbreak of Akabane disease in 1972-1973, 48 defective calves were examined (HAMANA et al., 1973). Japanese Black beef cattle and Holstein cattle of both sexes were included. The parity of dams was first to fourth. Usually the dam had no clinical sign during pregnancy except for some cases of dropsy in fetal membranes. Recurrence of estrus and conception were generally normal in spite of the delivery of defective calves, except in the case of dystocia and retained placenta. Dystocia and stillbirth were often encountered in the case of arthrogrypotic calves. Small size of calves was generally observed.

Most of the arthrogrypotic calves were unable to stand, but some could stand and even walk when helped. Arthrogryposis was found in one, two, three or four limbs and was often associated with single or multiple defects such as torticollis, scoliosis or hydranencephaly.

Complete or partial hydranencephaly with or without arthrogryposis was observed at necropsy (HAMANA & LEIPOLD, 1980). Various ocular anomalies were commonly found in these cases, including blindness, corneal opacity, inflammation and disappearance of eye reflexes. Insufficiently calcified incisors were often associated with gingivitis. Difficult breathing was another clinical feature of hydranencephaly.

The influence of arthrogryposis-hydranencephaly syndrome on the growth of 21 calves (two to ten months old) was investigated. In the cases of arthrogryposis, only calves with mild to moderate defects were nursed for longer periods. Inability to stand or difficulty in sucking was observed in the newborn stage. However, these conditions gradually improved and the calves began to be able to support themselves in spite of the apparent defects, although their growth was retarded.

The calves with hydranencephaly showed the common symptoms at the newborn stage, such as inability to stand, difficult sucking, ocular anomalies, abnormal eruption of incisors and difficult breathing. Most of the calves were slow and showed dull activity, but some were very strong and moved abruptly. In spite of the complete absence of the cerebral hemispheres, these calves could be kept alive as long as the owner would take care of them.

**Chuzan virus**

Among 1,677 cases found in Kagoshima, we investigated 211. Almost all the cases were
found in Japanese Black beef cattle, except two cases in Holstein cattle. There was no significant
difference in the sex of the calves; 56% were male. As to monthly incidence, the first case was
found in late November 1985. January saw the highest incidence and most of the cases were
found by March 1986. There were no significant differences in the parity of dams which were
distributed between the 1st and 13th parturition. Most of the calves were born within the normal
range of the gestation period.

Clinically, the defective calves were generally weak and paretic at birth; they could not suck
milk by themselves. Most calves could stand several hours later, but none of them achieved self-
suck. It was necessary to nurse them by bottle at first, and then take them to the mother's teat
later. Only a few cases showed self-suck after several days of nursing, but even then it usually
was not enough, causing marked retarded growth. Some showed persistent paresis. Nervous
signs such as opisthotonus, spasms and nystagmus were found in many cases. Even if the af-
pected calves could walk, they only walked aimlessly. We thus suspected disturbance of vision.

At necropsy, hydranencephaly was found in most of the cases, with a few having severe in-
ternal hydrocephalus. The cerebellum showed moderate to severe hypoplasia. It always accom-
panied the cerebral defects.

Akabane disease was determined not to be responsible for these defects because 66 dams
(33.8%) had a history of Akabane vaccination, and all five cases of colostrum-deprived calf se-
rums showed negative antibody titers against Akabane virus.

After many attempts at surveying serum antibody against several arboviruses which were
performed by Goto and Miura at the Kyushu branch of National Institute of Animal Health, one
orbi virus, belonging to the Reoviridae, showed strong positive reactions against all 187 serums
of dams and 184(98.4%) serums of the defective calves. In the colostrum-deprived calf serums,
the positive rate was 96.6%(29/30).

As a result, this virus was strongly suspected to be the causative agent and Goto and Miura
confirmed it finally in 1987 by the successful induction of the same defect following the inocu-
lation of the virus into an early pregnant cow. This virus was newly discovered and was named
Chuzan virus. It was first isolated from the blood of a Japanese Black beef cow and Culicoides

Though clinical cases of these defects were not recognized before November 1985, the ret-
rospective study of serum antibody titer against Chuzan virus using defective calves and their
dams showed 37.5%(27/72) positive in the serum collected from September 1984 to November
1985. However, no positive cases were found in 61 serums from September 1983 to September
1984.

Thereafter, no clinical case of Chuzan disease was confirmed although positive serum anti-
body titers against Chuzan virus were found at a low rate, and Chuzan virus was occasionally re-
covered from the blood of a cow and Culicoides oxystoma. Recently, however, some cases of
Chuzan disease were been suspected in an outbreak during January-April 1998, twelve years
after the previous outbreak.

**Aino virus**

During November 1995 to March 1996, 58 calves with CNS defects were studied. Both
Japanese Black beef cattle and Holstein cattle were included. There were no sexual differences
in occurrence. The parity of dams varied widely up to 12th parturition and dystocia was found
in 20 cases (34%) including five cases of caesarean section. Premature birth was common ~ 43%
of the cases.

Seasonal incidence was characteristic. Fifteen calves (26%) were born in November, 15 (26%) in December, 21 (36%) in January, 3 (5%) in February and 4 (7%) in March. No cases were found before October or after April.

Clinically, most of the calves (46; 79%) showed paresis with or without nervous signs. Torticollis was another feature, found in 47% of calves. Moderate arthrogryposis of limbs was found in 9% and severe ones in 17% of the cases. Nervous signs were found in 28 calves (48%), including ataxia, tetany, swimming movement, opisthotonus and circular walking. These nervous signs were often associated with cerebellar defects.

At necropsy, 46 calves (79%) showed either moderate (34%) or severe (45%) cerebellar hypoplasia. Mild internal hydrocephalus was found in five cases and severe in ten cases. In the latter half of the incidence, calves with cerebral cysts increased up ten cases. As for the other cerebral defects, micrencephaly in eight cases, polencephaly in two, partial absence of cerebrum in four and near-hydranencephaly in four was found.

The serum titers of three arboviruses, which are known as causative agents to induce the congenital CNS defects in calves in Japan, were investigated. The results are shown in Table 1.

<table>
<thead>
<tr>
<th></th>
<th>Colostrum-deprived serum</th>
<th>Colostrum-ingested serum</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>dam</td>
<td>calf</td>
<td>dam</td>
</tr>
<tr>
<td>No. of cases</td>
<td>9</td>
<td>15</td>
<td>23</td>
</tr>
<tr>
<td>Akabane virus</td>
<td>8(89)</td>
<td>3(20)</td>
<td>12(52)</td>
</tr>
<tr>
<td>Chuzan virus</td>
<td>0(0)</td>
<td>0(0)</td>
<td>2(9)</td>
</tr>
<tr>
<td>Aino virus</td>
<td>9(100)</td>
<td>15(100)</td>
<td>23(100)</td>
</tr>
</tbody>
</table>

Akabane virus, the most prevalent virus in hydranencephaly in Kagoshima, had moderately high positive rates in dam’s serum (63% in total). However, in the colostrum-deprived calf sera, it decreased to 20% (3 cases). Chuzan virus, which caused the big outbreak in calves of cerebellar hypoplasia and hydranencephaly in 1985-86, had very low positive rates in dams and calves. Especially, there was no positive case in the colostrum-deprived calf sera.

On the other hand, the positive rates against Aino virus were always 100% both in the dam and calf sera and also both in the colostrum-deprived and -ingested calf sera.

As for BVD-MD virus, which is known to induce cerebellar defects in calves, there were no or few positive rates in the defective calves examined by the Local Institute of Animal Health. Bluetongue virus was not investigated because there were no reports of CNS defects in Japan, though some positive serum titers were found sporadically.

From the above-mentioned findings and results, Akabane, Chuzan or BVD-MD virus should not be considered causative agents because of the different form of disease expression observed. Aino virus was strongly suspected and is now confirmed by the reproduction of the same defects through the experimental inoculation of fetuses in 1997 in the Kyushu branch of National Institute of Animal Health.

Both Aino and Akabane viruses belong to the Bunyavirus Simbu group first found in Japan in the 1970’s. Akabane disease has occurred many times since, but Aino virus has only sporadically been found in calves with cerebellar and/or cerebral defects, although positive serum titers
are relatively high in cows in Japan. The question is whether the same Aino virus caused the outbreak in which some 1,500 to 2,000 calves are estimated to have been sacrificed in Kagoshima and the neighbor Miyazaki areas.

Conclusions

Since 1972 when we started the investigation of calves with congenital defects, several outbreaks have occurred, as shown in Table 2, with an interval of one to five years.

<table>
<thead>
<tr>
<th>Year</th>
<th>Month</th>
<th>Features</th>
<th>Virus</th>
</tr>
</thead>
<tbody>
<tr>
<td>1972-73</td>
<td>Aug-May</td>
<td>Abortion, Arthrogryposis, Hydranencephaly</td>
<td>Akabane</td>
</tr>
<tr>
<td>77-78</td>
<td>Dec-Apr</td>
<td>Arthrogryposis, Hydranencephaly</td>
<td>Akabane</td>
</tr>
<tr>
<td>79-80</td>
<td>Oct-May</td>
<td>Arthrogryposis, Hydranencephaly</td>
<td>Akabane</td>
</tr>
<tr>
<td>82-83</td>
<td>Nov-Apr</td>
<td>Arthrogryposis, Hydranencephaly</td>
<td>Akabane</td>
</tr>
<tr>
<td>85-86</td>
<td>Nov-Apr</td>
<td>Hydranencephaly, Cerebellar hypoplasia</td>
<td>Chuzan</td>
</tr>
<tr>
<td>88</td>
<td>Mar-Jun</td>
<td>Hydranencephaly</td>
<td>Akabane</td>
</tr>
<tr>
<td>90-91</td>
<td>Dec-Apr</td>
<td>Hydranencephaly</td>
<td>Akabane</td>
</tr>
<tr>
<td>95-96</td>
<td>Nov-Mar</td>
<td>Torticollis, Cerebellar hypoplasia, etc</td>
<td>Aino</td>
</tr>
<tr>
<td>97-98</td>
<td>Aug-Feb</td>
<td>Abortion, Mummification</td>
<td>Ibaraki</td>
</tr>
</tbody>
</table>

During 26 years of research, we also found several hereditary defects. However, the largest damage to calf production was brought about by arboviruses in Kagoshima. Today, it is possible to differentiate these defects using etiological, clinical and pathological findings. Then, they can be confirmed by serological examination.

The reasons for repeated outbreaks at such a short intervals are suspected to be the short generation change of cattle and some variation of viral components. As the eradication of the insects which mediate the viruses is impossible, a vaccination program become very important for prevention. Monitoring and investigation centers will be needed to control future occurrence.

References