

Trypanosoma evansi Infection in Livestock in Thailand

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ABSTRACT

Trypanosoma evansi infection in livestock in Thailand has been reported in various kinds of animals for over 80 years. Clinical manifestations observed vary from asymptom to abortion and dead. Development of diagnostic methods parasitologically and serologically reveal more severe economic losses due to trypanosomiasis outbreaks. Whole herd treatment is costly while preventive control measures are not effectively implemented, especially in reservoir herds. Research is being undertaken in the development of efficient method for field diagnosis.

INTRODUCTION

Trypanosomosis or trypanosomiasis or surra caused by *Trypanosoma evansi*, has become a well-known disease to veterinarians all over the world for more than 100 years. It can be found in camel, elephant, horse, donkey, mule, cattle, buffalo, sheep, goat, pig, dog, cat, deer, and some wild animals. The geographical distribution of the disease is extensive in the New and Old Worlds, in Central and South America, Africa, Asia, Far East (Stephen, 1986).

Griffith Evans, an English veterinarian, who was working as Inspecting Veterinary Surgeon with the British Army at Punjab Frontier Force, India, first found the "eel-like" body moving actively in wet blood preparations of sick horses and camels in September and October 1880 (Evans 1880). The sickness was called by local people as "surra", denoting anything rotten. It was recognized later for Evan's contribution when he was over 90 (Stephen 1986). John Henry Steel made similar observations during an outbreak of a disease in transport mules in Burma (now Myanmar), in 1885 (Mahmoud and Gray 1980).

In Thailand, the first presumably outbreak in the troop of 13 imported mules from Algeria in 1916 was later reported by Colonel Luangsananruksat in 1949. Twelve of them died of the clinical signs similar to surra. During those days, surra was found mostly in horses, donkeys and mules which were used in the army as the means of transportation in wild and mountainous area. The reports revealed many losses of those animals during World War II, due to the lack of laboratory diagnostic facilities and effective drugs (Luangsananruksat 1949; Komutbut 1950 and 1956).

Due to the severity of surra in equidae and wide distribution in other animals, it was listed as one of the notifiable diseases in Thailand. According to the Animal Epidemic Act B.E. 2499 (A.D. 1957), it is compulsory for the farmers (animal owner) to notify the incidence or outbreak of the disease, or the death of his animals caused by the disease, to the local veterinarians or official-in-charge of the Department of Livestock Development within 24 hours. Sick or dead animals are subjected to move from the area before an official inspection. Dead animals must be burned, buried, or destroyed by other means.

LIVESTOCK HOST RANGE

Trypanosoma evansi infection has been found in livestock and domestic animals in every part of the country, however, some occurrences may not be reported.

Surra in buffaloes in Thailand was first reported by Kengkrasat (1929), but no details were described. Mathias and Muangyai (1980) first reported that a 24-day-old buffalo calf at the Veterinary Training Center of Chulalongkorn University in Nakhonpathom province was accidentally found infected with *T. evansi*, in a blood smear performing differential w.b.c. count. But the calf was not clinically ill. Monthly hemogram performed from the age of one month up to 5 months showed no abnormalities. Blood smears were found positive until the calf was 80 days old, while mouse inoculation was negative at the age of 136 days. Neramitmansook et al. (1982) made a survey of 85 healthy-looking buffaloes in Pitsanuloke province and found 22 animals were positive with *T. evansi* by mouse inoculation. Field investigations of 1,396 swamp buffaloes in 15 provinces in the north-eastern part of the country between January and December 1981 by Regional Veterinary Research and Diagnostic Center, Khon Kaen Province, alarmed all concerns that the infection rate of *T. evansi* by parasitological examination and complement fixation test was as high as about 20%, with a distinct peak of acute infection during the rainy season when fly activity was high (Loehr et al. 1985). The investigators made the notice that stress factors such as fascioliasis combined with insufficient nutrition during the dry season enhanced clinical trypanosomiasis. Observations of late abortion and stillbirth in buffaloes infected with *T. evansi* were reported (Timsad et al. 1985; Loehr et al. 1986). During 1984 and 1989, Kasemsant et al. (1989) conducted epidemiological survey of *T. evansi* infection in various animals in the north-eastern part of the country and found 92 outbreaks in 9 provinces. Infection rate in cattle, buffaloes, horses and dogs were 13%, 20%, 57% and 100% respectively. There was no incidence of *T. evansi* infection in pigs in this region.

Trisanarom (1981) reported the incidence of trypanosomiasis in crossbred dairy cattle in Chiangnai, the northern province. And in 1987, Trisanarom et al. reported a serious outbreak of trypanosomiasis in dairy cattle farm in Chiangnai, with abortion and stillbirth. Individual treatment with diminazene aceturate was not effective to control the outbreak, but finally, whole herd treatment using the same drug was. Sarataphan et al. (1989) reported an outbreak of trypanosomiasis in dairy cattle farm in Nakhonpathom province, the central part.

Native cattle (*Bos indicus*) are rather resistant to *T. evansi* infection comparing with other animals. However, there have been reports made by Chaichanapunpol et al. (1985) of a severely ill native cattle in Lampang province, and Tuntasuvan et al. (1997) of an outbreak in 13 farms in Petchaboon province. Both cases occurred in the northern part of the country. Affected animals showed nervous signs. The maledies were controlled by diminazene aceturate.

Nishikawa et al. (1990) conducted sero-epidemiological survey of *T. evansi* infection in 428 native cattle and 531 swamp buffaloes from 12 and 16 provinces, using IFAT, and found antibody titers to *T. evansi* 50% and 38.6% Of sera collected respectively. Prevalence rate in buffaloes was found highest in the North (57.4%) and lowest in the South (28.7%), while in cattle, the highest rate was found in the Central (64.7%) and the lowest in the Northeast (28.9%).

Pigs were also affected and reported in the North, in Pitsanuloke province (Teeraprasert, et al. 1984 a), the Central, in Nakhonpathom province (Thepsuthanone et al. 1984) and the West, in Suphanburi province (Siriwan et al. 1987). In the South, where climate is humid and

warm because there are only 2 seasons a year - rainy season and summer, which should be suitable for vectors development; but trypanosomiasis is rare. Recently, through personal communication with the staff of the Southern Veterinary Research and Diagnostic Center, there were outbreaks during August 1997 - June 1998 in 2 farms, one located in Nakhonsithamarat province, another one in Suratthani province. Sows had abortion and skin rashes. *Trypanosoma evansi* was examined from buffy coat.

Indrakamhang et al. (1996) reported the occurrence of *Trypanosoma* sp. in 3 native Sambar deer (*Cervus unicolor*) and 2 imported Rusa deer (*C. timorensis*) raised in a deer farm in Ayudhya, a province in the Central . Many of them gradually died between January 1 1995 and March 1996 with nervous signs and convulsion as the terminal stage. Early field diagnosis made through clinical signs and post-mortem observations had led to suspected hemorrhagic septicemia. Antibiotics treatment have been tried, but unsuccessfully. Whole blood samples in EDTA of 5 deer were sent to Parasitology lab., National Institute of Animal Health. Blood smears were all negative while mouse inoculation were all positive. PCV values were below average. There were 2 distinct forms in size and shape of *Trypanosoma* found, which were then named slender form and large form by the authors. The slender form had no statistically difference in size to *T. evansi*, but the large form needs further study to identify the species. Mimapan et al. (1998) detected *T. evansi* in fresh blood, cerebrospinal fluid, impression smear of cerebrum, and by immunohistochemistry (SAB) of 6 hog deer (*C. porcinus*) who were sent dying after showing nervous signs, and 1 carcass of dead animal. Antibody detection by ELISA technic were also positive (Tuntasuvan et al. 1998).

SEASONAL OCCURRENCE

Most of the outbreaks of surra occurred between the late rainy season and the early winter, which in Thailand last from August until February. Boonyawong et al. (1975) reported surra in crossbred thoroughbred mares in the Military Unit, in December 1974; Chaichanapunpol et al. (1985) found *T. evansi* in native cattle in October 1983; Timsad et al. (1985) observed abortion in swamp buffalo herd between August 1983 and July 1984; Trisanarom et al. (1987) reported an outbreak of trypanosomiasis in dairy cattle during June and September 1986; Teeraprasert et al. (1984a) reported *T. evansi* in pigs during June and August 1984; Siriwan et al. (1987) reported an outbreak in pig farm which occurred between June and November 1986; Indrakamhang et al. (1996) found *Trypanosoma* sp. in Sambar and Rusa deer from a deer farm where many deer became sick and died since December 1995 until late March 1996. Mimapan et al. (1998) detected *T. evansi* in hog deer from a farm where some animals became ill and died between September 1997 - February 1998. However, detection of *T. evansi* in animals blood in summer is not unusual, though much less occurrence (Patchimasiri et al. 1983).

TRANSMISSION

The climatic condition of Thailand is suitable for the vectors development. Tabanid flies have been incriminated as being mechanical vectors of trypanosomiasis, therefore an intensive survey of tabanid flies from cattle raising area between July 1994 and June 1996 in 30 provinces to identify the species of the vectors was headed by Dr. Yasuhiro Ito, an entomologist expert. Three genera, *Tabanus*, *Haematopota* and *Chrysops*, and 45 species were identified, with *T. striatus* as the highest amount collected (Ito 1996). Field observations on the seasonal distribution of the flies in a key farm where trypanosomiasis is

endemic, in Pathunrthani province, the central part of the country, made by Boonchit et al. (1996) between June 1994 and July 1995 every 10 days, showed the abundance of the flies was between June and October. This study meaningfully supported the seasonal occurrence of trypanosomiasis. From this report, *T. megalops* was predominant (65.49% of the total number 768 collected), among 29.69% *T. rubidus*, 3.13% *T. rufiscutellatus*, 0.65% *T. striatus* and 0.26% *T. oxybeles*. Pholpark et al. (1996) reported the highest number of tabanid flies in September (16) and October (9), by monthly collection in 2 key farms in Khon Kaen province during July 1995 and April 1996. *Trypanosoma striatus* and *T. rubidus* were identified.

Besides the mechanical transmission by blood sucking flies, transplacental is another means of *T. evansi* (Ogwu and Nuru 1981). In Thailand, there are some supportive evidences made by Loehr et al. (1986) who detected *T. evansi* in the impression smear made from the cotyledon of 12-hour aborted swamp buffalo fetus, and Chaichanapunpol et al. (1987) who found *T. evansi* in thin blood smear from the heart of an aborted fetus of dairy cow.

Trypanosoma evansi infected blood and meat were experimentally fed in dogs and mice by Raina et al. (1985) to prove the possible oral transmission in carnivores although the exact mechanism could not be explained.

Draught, pregnancy, abrupt climatical changes, insufficient feed, immunosuppression by other infections can be stress factors to enhance clinical *T. evansi* infection and flare up of the parasites in reservoirs (Loehr et al. 1985).

CLINICAL SIGNS

Equidae such as horses, mules and donkeys, which are the most susceptible species usually show acute clinical signs: depressed, inappetence, intermittent fever, anemia and icterus. Edema and ecchymotic hemorrhage of some part of bulbar, palpebral conjunctiva and the third eyelid of both eyes. Edema of subcutaneous tissue of brisket, belly and legs (Luangsanaruksat 1949; Boonyawong et al. 1975). Restlessness, dullness, incoordination of the legs, staggering gait and last stage convulsion seemed to be the results of brain tissue disturbance by the parasites (Komutbut 1950). They died shortly after showing some clinical manifestations. Peracute cases usually died quickly without noticeable symptoms, while chronic cases showed edematous swelling of lower part of the body and limbs, anemia, weakness and died later if untreated.

In buffaloes, while asymptomatic *T. evansi* infection is common, but the clinical ones are quite seriously affected. Clinical observations made by Loehr et al. (1985) ranged from high to low frequency as follows: stiffness, conjunctivitis, emaciation, swelling of legs, fever (over 40°C), inappetence, dyspnea, anemia, recumbency, death, diarrhea and abortion.

Dairy cattle were most affected and quickly spread. Trisanarom et al. (1987) reported an outbreak in dairy cattle which caused an enormous economic loss due to milk reduction, abortion and stillbirth as well as retained placenta. In 1989, Sarataphan et al. reported the effect of natural *T. evansi* infection on milk yield of dairy cattle.

In symptomatic beef cattle, besides general manifestations such as fever, emaciated, anemic, jaundice, lethargic, loss of condition, nervous clinical signs observed are shivering, salivation, circling movement, jumping, lateral recumbency and convulsion (Chaichanapunpol et al. 1985; Tuntasuvan et al. 1997). Mostly they are reservoir hosts.

Pigs, especially the breeding pigs and pregnant sows are quite affected. Skin rashes on the ears, under and lateral sides of the body, udder, scrotum are usually observed. Abortion

occurs at any stage of pregnancy, but more frequently at 1-2 months (Teeraprasert et al. 1984a; Thepsumetanone et al. 1984; and Siriwan et al. 1987). Rate of abortion are usually high. Deaths sometimes followed abortion. Experimental infection of sows at 1 1/2 month pregnancy with *T. evansi* resulted in abortion at 100 hrs. postinfection. Parasitemia was detected at 18 hrs. (Teeraprasert et al. 1984b) Nervous signs such as convulsion, circling movement, were observed (Teeraprasert et al. 1984a) and sometimes followed with death. Infected weaning and fattening pigs showed no symptoms (Teeraprasert et al. 1984a; Siriwan et al. 1987).

Trypanosoma sp. naturally infected Rusa and Sambar deer had edema of the neck, rough hair coat and weakness, later showed nervous signs such as standing hair, teeth grinding, foamy salivation, circling movement, bent neck, and last stage convulsion before dead (Indrakamhang et al. 1996). *Trypanosoma evansi* infected hog deer developed similar clinical nervous manifestations (Tuntasuvan et al. 1998).

ECONOMIC LOSS

It is well aware that economic loss is enormous, whenever outbreak occurs in any farm. Loehr et al. (1986) reported losses due to abortion and stillbirth in cow and buffaloes who were found infected with *T. evansi*, in the North-east Thailand. Trisanarom et al. (1987) reported an enormous economic loss due to milk reduction, abortion and stillbirth as well as the cost of control in an outbreak in Chiangmai dairy cattle farm. Siriwan et al. 1989 calculated economic loss due to *T. evansi* infection in an outbreak occurred in the intensive pig farm as high as US \$8,500 (at the exchange rate of US \$ 1 = Thai β 41) when 61 sows were dead, 45 were aborted, and 46 were culled; including feed, labor wage and drug used in the control.

DIAGNOSIS

In early reports of trypanosomiasis in Thailand, diagnosis was based on clinical symptoms and basic parasitological examinations such as wet smear, thin smear, Woo's method and mouse inoculation (Luangsanaruksat 1949; Komutbut 1950, 1956; Mathias and Muangyai 1980; Indrakamhang et al. 1982; Neramitmansook et al. 1982; Patchimasiri et al. 1983). Among those methods, mouse inoculation is the most sensitive (Neramitmansook et al. 1982; Kasemsant et al. 1989). Later, some serological methods such as CFT, IHA, IFA, were developed (Pholpark and Pholpark 1982; Vitoorakul et al. 1986; Nishikawa et al. 1989), which enabled sero-epidemiological survey and revealed more infection rate and distribution of trypanosomiasis in various host animals. Some works compared the sensitivity and specificity of these methods (Loehr et al. 1986; Vitoorakul et al. 1986; Nishikawa et al. 1989). Commercial test kit (Testryp®CATT) was tried in some serological survey with promising results in comparable with IHA (Vitoorakul et al. 1986), and CFT (Bajyana Songa et al. 1987). More advanced methods were also developed: DNA probe by Visenskul et al. (1988), CIEP (counter immunoelectrophoresis) by Vitoorakul et al. (1989), ELISA technic for both antibody detection by Indrakamhang et al. (1991) and Tuntasuvan et al. (1996); and antigen detection by Thammasart and Kashiwazaki (1998) and Kashiwazaki and Thammasart (1998). But DNA probe and CIEP are not used in routine diagnosis any more.

Streptavidin-biotin complex (SAB) technic was recently developed to detect the parasites in tissue section (Mimapan et al. 1998), and was able to detect *T. evansi* in brain tissue of hog deer.

Research is now being undertaken at the National Institute of Animal Health, Bangkok to produce dipstick colloidal dye to be used for field diagnosis.

TREATMENT AND CONTROL

In the old days, the first effective trypanocidal drug was quinapyramine methyl sulphate (Antrycide®). It was widely used in the Army (Komutbut 1950; 1956). Boonyawong et al. (1975) successfully treated 4 severely ill thoroughbred mares with quinapyramine and hematinics. But local reaction from subcutaneous injection, anaphylaxis, and short curative effect led to new drug, sulphonate naphthylamine (Naganol®). It can be administered intravenously and intrathecally in concurrent or separately with little side effect (Komutbut, 1956).

Diminazene aceturate (Berenil®) was introduced into the market later and is the only available trypanocidal drug in Thailand until now. Many reports claimed the successful control of the outbreaks using Berenil® (Loehr et al. 1986; Teeraprasert et al. 1984a; Trisanarom et al. 1987; Siriwan et al. 1987; Tuntasuvan et al. 1997), although the curative effect follow up did not appear. Indrakamhang and Thammasart (1994) conducted a trial using low dose of diminazene aceturate to treat milking cow and found that it was as effective as recommended dose for 4 months. In contrast, Kasemsant et al. (1984) reported the failure of recommended dose in swamp buffaloes but the success of double dose.

Mass or whole herd treatment is needed to get rid of the carriers or reservoirs. There are still a number of endemic farms where annually outbreaks occur eventhough the whole herd were treated. It is quite costly for treatment in a large farm. Preventive control measures are also too expensive for the farmers. At least biannual prevention must be administered before the seasonal occurrence. Last-longer trypanocidal drug such as isometamidium chloride (Samorin®) is better in prevention than treatment (Kasemsant et al. 1989) and safe for the pregnant (Dowler et al. 1989), but it was on the market in Thailand only for a short sale. It can be administered intravenously to avoid depot formation by intramuscular injection (Dowler et al. 1989).

However, the effective control measures still need two more considerations, the vectors which are abundant and difficult to be under control, and the nearby reservoir hosts.

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