REVIEW OF RESEARCH IN VETERINARY PARASITOLOGY IN THE RESEARCH INSTITUTE FOR VETERINARY SCIENCE (RIVS) SINCE 1983

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ABSTRACT

Studies on gastrointestinal parasites and the epidemiology and pathogenesis of blood protozoa in ruminants have been undertaken during the last few years. An abattoir survey for a period of one year showed that most large ruminants slaughtered in the Bogor abattoir were infected with nematodes, about half with trematodes and few with blood parasites. Worm infections were also common in small ruminants but none were infected with blood protozoa. Some field studies on *Fasciola* infection in large ruminants showed that these animals were susceptible to the disease. The seasonal prevalence of fascioliasis is not yet fully determined; management systems and the local geography play an important role. Most anthelmintics were effective against worm infection in large and small ruminants. A new flukicide (triclabendazole) is highly effective against both immature and mature liver flukes. Studies on the epidemiology and pathogenesis of trypanosomiasis in cattle and buffalo have been conducted since 1982. The results indicate that *T. evansi* is prevalent in five regions in Java; the prevalence rates are higher in Sahiwal and Belmont Red than Ongole cattle or buffalo. ELISA values increase with increasing age of the animals. Buffaloes are more susceptible than cattle. Naganol is the drug of choice for control of trypanosomiasis in Indonesia.

INTRODUCTION

Buffalo *(Bubalus bubalis)*, Ongole cattle *(Bos indicus)*, Bali cattle *(Bos javanicus)* and horses are kept under small-holder farming systems in Indonesia. These animals are mainly used as draught animals for traction either for agricultural purposes in the rice fields, or transport; they also provide an important cash income when they are sent for slaughter. Other animals such as sheep and goats also contribute significant income for small holder farmers, while dairy cattle which are mainly located in Java, have an important role in improving the income of small-holder farmers through dairy cooperatives. Attempts have been made by the government to increase the animal population by importing from different countries; imported animals include Sahiwal, Brahman, Belmont Red, Buffaloes and Holstein Friesian cattle.

The tropical conditions of Indonesia enhance parasite development and consequently heavy infections are often found. These may result in losses of production, growth retardation, and death of animals. Several parasitic diseases are of considerable economic importance; fascioliasis in cattle,

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buffaloes, sheep and goats; trypanosomiasis in cattle, buffaloes and horses; and scabies and nematodiasis in sheep and goats. Research has been conducted during the last five years on the epidemiology, pathogenesis, diagnosis and control of some important parasitic diseases with the aim of improving the small holder farmer's income and animal draught power facilities.

FASCIOLIASIS

Fascioliasis is a parasitic disease caused by helminth trematodes from the genus *Fasciola*. Only one species, *Fasciola gigantica*, occurs in Indonesia. In its life cycle the parasite needs a fresh water snail, *Lymnaea rubiginosa*, as the intermediate host. The disease is particularly important in ruminants. Chronic fascioliasis usually occurs in large ruminants and acute cases, in small ruminants. This disease can cause growth retardation and lowered milk production, reproductive efficiency and food conversion. In sheep an infection with 45 flukes can cause a significant decrease of weight gain.

Epidemiology : An abattoir survey in Bogor 1982/1983, based on the presence of eggs in the faeces, showed prevalences of fascioliasis in cattle and buffalo of 20% and 14% respectively. In 1984/1985 *Fasciola* infection was examined at the same abattoir by recovering the adult fluke from the liver. The results were as follows : in cattle and buffaloes the prevalence was 61% and 31% respectively. Adult fluke measured from 24.8 to 56 mm in length and from 6.3 to 14.5 mm in width, while the egg measurement was (128-171) x (75-107) µm. *Fasciola* collected from buffaloes were significantly smaller than those from other hosts but all flukes were identified as *F. gigantica*. It is suspected that there are variations in resistance between the host species.

Over a four year period, liver samples from large ruminants, were examined for pathological changes by Tarmudji *et al* (1984)[1]. The highest prevalence of fascioliasis was in 1984 (34%), the lowest in 1981 (13.7 %) and in the other years it was about the same (27%). Compared to other districts, Malang had the highest prevalence of fascioliasis (74.6 %) in 2-4 year old animals.

Suhardono *et al*[2] (1988) have investigated fascioliasis in large animals in some districts of the provinces of Central Java and D.I. Yogyakarta. They found differences in the prevalence of the disease with different farming or agricultural systems. Dairy cattle kept in the high land such as the northern district of Klaten, where crop plantation is mainly grain plants had an extremely low incidence. However, the same breed of cattle raised on the low land in the southern district of Bantul, they had an incidence of 50%. These dairy cattle ate grass which was an agricultural by-product. Fascioliasis in beef cattle and buffalo in this area was also high (10% - 95%).

The prevalence of fascioliasis in sheep and goats is high and from the abattoir survey in Bogor in 1984, based on the presence of adult fluke in the liver (Suhardono, unpublished data) was about 21% and 20% respectively. The prevalence of fascioliasis in goats in three districts of East Java, based on egg count, was 7% (highest proportion in district Ponorogo but none from Malang).

Suhardono *et al*[2] (1988) found o.62 % of *Lymnaea rubiginosa* infected with developing stage of *Fasciola* (range of 0.05% - 1.49%). The village of Negla had the highest infection rate of three villages in subdistrict Surade. It was also found that fascioliasis in

the lymnaeid snail fluctuated during the year; this may coincide with fluctuations in the climate. Snails collected between December and May had a higher infection rate than those collected in other months. Developing trematodes of other species were found more commonly than those of *Fasciola*; they differed in their morphology, activity and numbers. Based on the morphology of these cercaria, it is thought that the adults parasitize birds, reptiles, rodents or other wild mammals. The low incidence of fascioliasis in the snail showed that the snail has a great capacity in spreading fascioliasis, by means of cercarial production, in livestock.

Pathogenesis : Basically two main changes associated with stages in fluke infection of young sheep were found by Wiedosari (unpublished data). The most severe and acute damage to the liver was caused at the beginning of infection by migrating flukes which caused mechanical damage to the liver parenchyma and also ingested liver cells. The result was thrombosis of veins, haemorrhage, necrosis and infiltration by inflamatory exudate and cells. At the beginning the predominant inflammatory cells were eosinophils, which were gradually replaced by neutrophils, which in turn were replaced by mononuclear cells. The second form of lesion started to develop at the time when the flukes entered the bile ducts 12-14 weeks after infection. There was progressive biliary and portal cirrhosis with enlargement of bile ducts and thickening of their walls.

Diagnosis : The usual method of diagnosis for fascioliasis in livestock held at RIVS is by detection of eggs in the faeces using a sedimentation method. Another method, an enzymatic test of GLDH and GGT, has also been done to evaluate acute and chronic liver damage. Laboratory studies using artificially infected calves (350 metacercariae) and young buffalo (650 metacercariae) of *F. gigantica* by Suhardono *et al*[2] (1988) found that there was no significant difference between treated and control groups in the levels of enzyme GGT and GLDH. Plasma enzyme levels show greater variation in heavily infected animals. The low infection rates in these experiments may have influenced the observed results for this parameter. However, in a field trial conducted over a period of one year (treatments every eight weeks), the values of GGT and GLDH were significantly different between treated and control animals from the time of the first treatment.

Drug trials : Laboratory tests of triclabendazole on calves artificially infected with metacercariae of *F. gigantica*[2] demonstrated a high level of efficacy of triclabendazole against early immature and adult *F. gigantica* from 2-10 weeks after infection. Triclabendazole was less effective in buffaloes. There was a very low efficacy against 2 week old flukes and efficacy against more mature flukes was also reduced. An explanation for the difference in efficacy of this drug in cattle compared to buffalo must lie with the pharmacokinetics of the drug in the two host species. The dose rates used in the experiments with buffaloes are those recommended for use in cattle.

TRYPANOSOMIASIS

Trypanosomiasis caused by *Trypanosoma evansi* is widely distributed in most islands of Indonesia. Cattle, buffalo, and the horse are the natural hosts; deaths in these animals were recently reported in Aceh (Payne *et al*, unpublished data), in Madura (Sukanto *et al*, unpublished data), and in buffalo imported from Australia (Payne *et al*, unpublished data). Research in trypanosomiasis has been conducted on diagnostic

techniques, epidemiology, pathogenesis and treatment.

Clinical signs: Cattle and buffaloes. infected with *T. evansi* at a dose rate of 10^8 parasites showed chronic clinical signs such as weakness, roughness of the skin and hair coat; young animals were more severely affected than adults. Infection resulted in a reduction of body weight. Under field conditions, only some infected animals showed chronic clinical signs, Growth retardation was found in buffalo calves in Blora (Central Java) and Tuban (East Java).

Diagnostic techniques: Several techniques have been used to detect active infection of *T. evansi*. These include fresh blood examination, blood smears, mouse inoculation (MIC) and the haematocrit centrifugation technique (HCT). Fresh blood examinations and blood smear have a low accuracy in detecting active infections. A study on the use of MIC & HCT indicates that MIC is the most reliable test with a positive detection rate of 78%[3]. However, the need for experimental mice and the time consumed are considered to be limiting factors for diagnosis in the field; therefore HCT with a reasonably high positive detection rate of 46% is recommended for rapid diagnosis in the field.

***T. evansi* Prevalence Rate** (TPR) : A field study on the epidemiology of *T. evansi* in buffaloes, Ongole, Brahman, Sahiwal, and Belmont Red in West, Central and East Java was conducted from 1982 to 1986. The results indicated that TPR was o% in Brahman, 1.58% in Ongole, 5.84% in Buffaloes, 6.45% in Sahiwal and 7.89% in Belmont Red. TPR increased with increase in age to maturity but it declined from the mature to the adult stage. Medium rainfall was associated with TPR. A low PCV was associated with infection. Exotic breeds (Sahiwal and Belmont Red with the exception of Brahman) are more susceptible than Ongole cattle or buffalo.

Pathogenesis: Experimental studies on the pathogenesis of *T. evansi* in buffaloes, Ongole and Holstein Friesian calves and adults indicated that *T evansi* caused greater weight loss over the first 12 weeks after infection. This disease is more pathogenic in calves than adults as judged by a higher mortality rate in calves. Poor nutrition may have an indirect effect on pathogenicity through increased susceptibility to intercurrent disease as a result of reduced immunocompetence. Field studies indicate that *T. evansi* and *Anaplasma marginale* were the cause of death of cattle in Aceh (Sukanto, unpublished data), and imported buffaloes in West Java (Payne *et al*, Unpublished data). During an outbreak of trypanosomiasis in cattle, buffaloes and horses in Madura, *T evansi* was the cause of death and it was detected by HCT in 50% of buffaloes and 13% of cattle examined (Sukanto *et al*, unpublished data).

Serology : The indirect ELISA test was developed and used for seroepidemiology of *T. evansi* in Indonesia; this demonstrated antibodies to *T. evansi* in 54% of buffaloes and 45% of cattle tested. Experiments in buffaloes and cattle in the field and laboratory indicated that ELISA detected infection in week-1 after infection. ELISA prevalence rates and ELISA titres increased with increase in age, but the ELISA prevalence rate decreased from mature to the adult in buffalo[3]. Indirect ELISA detects antibody, therefore this test is applicable for serodiagnosis. The direct ELISA to detect active infection is more useful; this test is now being developed in collaboration with CTVM, University of Edinburgh.

Factors which may affect the pathogenesis in trypanosomiasis are antigenic variations and strain differences. Work in this

area indicated for the first time in Indonesia that antigenic variation in *T. evansi* occurs in Ongole, Holstein Friesian and buffalo and that during the early infection a new variation occurs at least every 7 days. This work has subsequently been expanded by Jones[4] (1987) using a more sophisticated technique. Using a cloned population of *T. evansi* to prepare specific antisera to variable antigen type (VAT), he demonstrated that calves in the Jonggol area had maternally derived VAT-specific antibody which persisted for up to three months of age.

Immunosuppression in swamp buffalo maintained on high or low planes of nutrition and infected with *T. evansi* was investigated. Responses to dinitrochlorobenzone (DNCB) and phytohaemagglutinin (PHA) were used as an indicator of cell mediated immunity (CMI), and homologous passive cutaneous anaphylaxin (HPCA) to egg albumin and serological responses to chicken and sheep red blood cells (CRBC; SRBC) as indicators of humoral immune competence. There was evidence of suppression of CMI to the DNCB during the first week after infection but not at week 5 whereas the CMI response to PHA infected animals remained suppressed throughout the 14 weeks of observation. No satisfactory result was achieved with HPCA to egg albumin using protocols which are successful in cattle. However, the serological response to CRBC and SRBC was suppressed in infected animals and also in non-infected animals on a low plane of nutrition.

Treatment trials : To date naganol is the drug of choice for *T. evansi* in Indonesia. This drug was still effective against most isolations of *T. evansi* in mice at a dose rate of 3-10 mg/kg BW, and against one isolate naganol was still effective at the lowest dose of 1.25 mg/kg BW[5,6,7]. Other results showed that naganol was effective against *T. evansi* in mice, cattle and buffaloes, but some isolates were relatively resistant to naganol[8,9]. Serial experiments were carried out to test the susceptibility of *T. evansi* against berenil. In mice infected with *T. evansi*, berenil at a dose rate of 3.5 - 7 mg/kg BW gave only variably therapeutic effects[5,6,7,8,9]. The results of drug trials in mice indicated that trypamedium killed only some isolates of *T. evansi* in mice, while in cattle and buffaloes this drug only gave temporary cures[8,9].

NEMATODIASIS IN SHEEP AND GOAT

Epidemiology and Pathogenesis : Endoparasitic infestation of sheep in the village of Ciawi, West Java has been reported by Beriajaya[10] (1984). He recorded at least 6 nematode genera based on larval identifications. These are *Haemonchus, Trichostrongylus, Oesophagostomum, Bunostomum, Cooperia, Strongyloides*. Eggs of *Capillaria, Trichuris, Fasciola* and paramphistomes were found in faeces. The trematodes and *Capillaria, Trichuris & Moniezia* were less common than the other listed nematodes. Nematodiasis is more common in young animals than in adults except for paramphistome infection which is more common in adults. The importance of endoparasite infection in goats kept under traditional farming methods in villages in East Java has been studied by Beriajaya (unpublished). Based on the mean egg count in the faeces, the infection rate was very light. Very few goats had egg count greater than 1000 epg. Chronic infections caused a decrease in body weight gain and lowered the packed cell volume.

Immunology : Research on the immunity of sheep to nematodes has been conducted by Beriajaya (1987, unpublished); 5000 third stage larvae of *Haemonchus con-*

tortus irradiated with Co^{60} 500 Gy, were administered orally as a live vaccine, either once or twice. The group receiving a single vaccination had higher faecal egg counts than those which were vaccinated twice. A double vaccination with irradiated larvae may stimulate an immune response.

Anthelmintic trials : Some anthelmintic tests have been done by Beriajaya[11] (1986). One group of naturally infested sheep *(Haemonchus, Oesophagostomum & Trichostrongylus)* were treated with closantel at 7.5 mg/kg BW once every 6 weeks for a period of 18 weeks; another group was treated once only at the beginning of the trial. Two weeks after the first treatment both groups showed a significant decrease in their mean egg count. The group which received three treatments maintained a slightly lower nematode egg count than the other group. Good control was achieved of *Haemonchus* but not of the other worms. However there was a significant difference in liveweight gain between treated and control animals.

Another study was conducted in which sheep were treated with levamisole at 8.9 mg/kg BW every 4 weeks for 16 weeks. By the fourth week after each treatment most sheep were again excreting nematode eggs, the majority being *Haemonchus*. A significance difference in liveweight gain was seen after the third treatment had been given.

ECTOPARASITIC DISEASES OF GOAT, RABBIT AND BUFFALO

Sarcoptic mange (*Sarcoptes scabiei* infestation) in goats, *S. scabiei* and *Psoroptes sp.* in buffaloes, and *Notoedres cati* in rabbits are important parasites in Indonesia. Trials were conducted to test the efficacy of ivermectin. Ivermectin (200 µg/kg BW) treatment caused a reduction in mite numbers and an increase of body weight in goats, buffaloes and rabbits[12,13,14].

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QUESTIONS AND ANSWERS :

1. Question: You said that only *Fasciola gigantica* is in Indonesia. Is there no Fasciola hepatica at all in Indonesia ?

   Answer : Only one species that can survive because the other *(F.hepatica)* there is no intermediate host for it.

2. Question: What is the role of ivermectin in the treatment of helminths in animals ?

   Answer : May be it is now the best nematocide in livestock by blocking the GABA enzyme of the parasite.