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Evaluation of anti-theilerial efficacy of various drugs in crossbred cattle naturally infected with *Theileria annulata*

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Abstract

Theileria annulata is an important tick-borne haemoprotozoan disease affecting dairy animals in tropical and subtropical region of the country and is responsible for causing huge economic losses to the dairy farmers. The present study was undertaken to evaluate the efficacy of buparvaquone, chloroquine, α , β -artether and diminazine aceturate along with oxytetracycline in cattle naturally infected with *T. annulata*. Animals of GI, GII, GIII and GIV of 10 animals each were treated with single dose of buparvaquone @ 2.5mg/kg body weight, chloroquine @ 1ml/30kg body weight, α , β -artether @ 1ml/30kg body weight and diminazine aceturate @ 3.5mg/kg body weight, respectively along with intravenous injection of oxytetracycline @ 20ml/kg body weight for 5 days. The reduction in intensity of infection from ++++ to +, ++++ to ++, ++++ to ++ and ++++ to ++ was recorded in animals of GI, GII, GIII and GIV, respectively. Thus, buparvaquone showed better efficacy than other drugs used in the present study.

Keywords: efficacy, drugs, *Theileria annulata*, cattle

1. Introduction

India is an agriculture based country where about more than 65% population resides in rural areas. Livestock farming by the poor and marginal farmers is an integral component of the Indian economy which provides a regular source of income and gives immense opportunities of employment for the livelihood of the rural people. Bovine Tropical Theileriosis (BTT) caused by *Theileria annulata* is an important tick-borne haemoprotozoan disease affecting dairy animals in tropical and subtropical regions of the country and it causes huge economic losses in terms of high mortality rate due to lack of early diagnosis and therapeutic management. The prevalence of *T. annulata* has been reported from different geographical regions of India including Uttarakhand by the several workers [1, 2, 4, 16]. The treatment of theileriosis is practiced using three effective drugs viz. parvaquone, buparvaquone and halofuginone lactate that are commercially available worldwide [11]. The efficacy of these drugs has been evaluated by several workers and observed that buparvaquone, a second-generation hydroxynaphthoquinone, is more effective as the drug eliminates various developmental stages of *T. annulata* from both erythrocytes and lymphocytes and helps in improvement of the animals from clinical form of the disease and thus stabilizing it as drug of choice [3, 12]. Therefore, treatment of theileriosis with buparvaquone is commonly practiced by the veterinarians under field conditions worldwide [6, 10]. The therapeutic management of theileriosis includes single dose of buparvaquone @ 2.5mg/kg b. wt., oxytetracycline @ 20 mg/kg b. wt. and that costs Rs. 3,000/animal. The treatment cost of BTT is very high and also the indiscriminate use of buparvaquone results in drug resistance [5]. Therefore, several workers have switched over to finding other drugs as an alternative treatment of BTT. Keeping in view the high treatment cost with buparvaquone, the present study was conducted to evaluate the anti-theilerial efficacy of various drugs in cattle naturally infected with *T. annulata*.

2. Materials and Methods

Selection and grouping of animals

The evaluation of anti-theilerial efficacy of various chemical compounds was carried out in cattle in and around Pantnagar. A total of 40 cattle found naturally infected with *Theileria annulata* were selected on the basis of symptoms followed by microscopic examination of

Giemsa stained thin blood smear found positive for the organism. The organisms were identified as per their morphological features [16]. All 40 animals found positive for *T. annulata* were randomly divided into 4 different groups viz. GI, GII, GIII and GIV of 10 animals each.

Drugs used against *T. annulata*

Four chemical compounds viz. buparvaquone, chloroquine, α , β -artether and diminazine acetate along with oxytetracycline were selected for evaluation of their efficacy against *T. annulata*. Animals of GI, GII, GIII and GIV were treated with single dose of buparvaquone @ 2.5mg/kg body weight, chloroquine @ 1ml/30kg body weight, α , β -artether @ 1ml/30kg body weight and diminazine acetate @ 3.5mg/kg body weight intramuscular (IM), respectively along with intravenous (IV) injection of oxytetracycline @ 20ml/kg body weight for 5 days. Blood samples were collected from

each animal on 7, 14, 21 and 28 days post treatment for confirmation of presence or absence of parasites by microscopic examination of thin blood smear using Giemsa stain. The parasitemia percentage in the blood smear was classified as + (scanty < 2%), ++ (mild 2 - 10%), +++ (moderate 10 - 40%), ++++ (high 40 - 60%), +++++ (very high >60%) after seeing 300 erythrocytes from 5-10 different microscopic fields under 100x oil immersion lens [8].

3. Results and Discussion

The anti-theilerial efficacy of various combinations of chemical drugs (buparvaquone + oxytetracycline, chloroquine + oxytetracycline, α , β -artether + oxytetracycline and diminazine acetate + oxytetracycline) was estimated in cattle naturally infected with *T. annulata*. The details of efficacy of various combinations of chemical drugs against *T. annulata* are presented in Table-1.

Table 1: Evaluation of efficacy of drugs against *Theileria annulata* in cattle

Groups	Drug used	Pre-treatment intensity of infection	Intensity of infection post-treatment (days)				Clinical signs	Interpretation of result
			7 DPT	14 DPT	21 DPT	28 DPT		
GI	Buparvaquone + Oxytetracycline	++++	+++	+++	++	+	Disappeared	Cured
GII	Chloroquine + Oxytetracycline	++++	+++	+++	+++	++	Persistent	Not Cured
GIII	α , β -artether + Oxytetracycline	++++	+++	+++	+++	++	Persistent	Not Cured
GIV	Diminazine acetate + Oxytetracycline	++++	+++	+++	+++	++	Reappeared after 2-3 days	Partially improved

The animals of GI were treated with single dose of buparvaquone @ 2.5mg/kg IM and oxytetracycline @ 20 mg/kg IV with 1 lit DNS once in a day for 5 days. The intensity of infection decreased from ++++ to + 28 days post treatment (DPT). The clinical signs disappeared 28DPT and animals returned back to normal health status. It was observed that animals feed intake slightly improved and temperature came into normal range. As time passed, all cattle showed complete recovery and milk yield also increased. The efficacy of buparvaquone @ 2.5 mg/kg body weight against theileriosis has been evaluated by several workers and they have found varied range of (86.66 - 100%) efficacy. Now a days, buparvaquone and oxytetracycline are the most commonly used anti-theilerial drugs in cattle around the world [12]. The early treatment with buparvaquone is 100% effective in eliminating the *T. annulata* from the blood and lymphocytes whereas in the later stages of the disease, it was unable to eliminate the parasites from blood and lymphocyte as well and partially failed to improve the clinical condition of the animal [17].

The animals of GII and GIII were treated with a combination of single dose of chloroquine @ 1 ml/30 kg b. wt. IM along with oxytetracycline @ 20 mg/kg IV with 1 lit DNS once in a day for 5 days and single dose of α , β -artether @ 1 ml/30 kg b. wt. IM along with oxytetracycline @ 20 mg/kg IV with 1 lit DNS once in a day for 5 days, respectively. The intensity of infection decreased from ++++ to ++ in both GII and GIII 28DPT, respectively. Following treatment it was observed that clinical signs were present in the animals at the end of experiment and animals were not cured. The decreased in intensity of infection could be due to the use of oxytetracycline and chloroquine drug was totally non effective for controlling theileriosis [15]. The effectiveness of oxytetracycline against *T. annulata* has also been reported [7, 15]. α , β -artether was used against *T. annulata* for the first time. It is an ethyl ether derivative of artemisinin commonly

used as an anti-malarial drug especially for *Plasmodium falciparum* and is schizonticidal [16, 19]. The lower efficacy of the drug in the present study may be due to the animals being infected with piroplasm of *T. annulata*. It could be used in animals infected with schizont stage of the parasite i.e., in early stage of infection of *Theileria*.

The animals of group IV were treated with single dose of diminazine acetate @ 3.5 mg/kg b. wt. IM and oxytetracycline @ 20 mg/kg IV with 1 lit DNS once in a day for 5 days. The intensity of infection decreased from ++++ to ++ 28DPT as in group II and III but clinical signs disappeared after 28DPT for short duration of time and again reappeared within 2-3 days. Thus, the formulation was found partially effective. The improvement in animals infected with theileriosis following treatment with diminazine acetate with oxytetracycline has been reported [15]. Furthermore, they stated that some more treatment is essentially required in later stage of infection for complete recovery. Oxytetracycline and diminazine acetate is effective against chronic theileriosis [9]. Oxytetracycline @ 20mg/kg is less efficient in theileriosis and recommended buparvaquone @ 2.5 mg/kg deep IM, with supportive treatment [13].

4. Conclusion

The results of the present study clearly indicated that buparvaquone had better efficacy than diminazine acetate, chloroquine and alpha, beta-artether when used in combination with oxytetracycline. Buparvaquone @ 2.5mg/kg body wt. reduces the intensity of infection of *T. annulata* from ++++ to + as compared to other drugs but unable to eliminate the parasite from blood of infected animals. It is suggested that single dose of buparvaquone is not sufficient enough to eliminate *T. annulata* from the blood of infected animals. Hence, two doses of the drug are essentially required for complete elimination of the parasite. It has also been practically observed that several cases of Bovine Tropical

Theileriosis have been cured after two treatments of bupravquone at recommended dose and at 48 hr interval.

5. Author's declaration

Author declared no conflicts of interest.

6. Acknowledgment

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