Research Note

EFFICACY OF MK-401 (4-AMINO-6 TRICHLOROETHENYL-1,3 BENZENEDISULFONMIDE) AGAINST FASCIOLA HEPATICA *IN ARTIFICIALLY INFECTED CALVES*[1,2]

Fascioliasis is one of the important parasitic diseases affecting cattle in Puerto Rico[3].

Control of fascioliasis is directed towards the destruction of the snail intermediate host, *Lymnaea cubensis*, and its habitat[4]. In Puerto Rico, areas of flat and poorly drained pasturelands where water accumulates during the rainy season, and high ambient temperatures combine with high rain-fall and humidity to make control of fascioliasis in grazing cattle extremely difficult are common. In addition, the ability of the snails to undergo cryptobiosis or aestivation presents a difficult problem.

MK-401 (4-amino-6 trichloroethenyl-1, 3 benzenedisulfonamide), a recently developed benzenedisulfonamide, is reported to be effective against mature and immature *F. hepatica* worms, while having a wide margin of safety[5]. De León and Quiñones[6] reported that MK-401 at the rates of 6 and 12 mg/kg body weight had a 100% efficacy against 8-week old *F. hepatica*. The present study was undertaken to further evaluate MK-401 at the dosage rate of 7 mg/kg body weight.

Twenty-one Holstein-Friesian/Brown Swiss male calves (approximately 3 months old) from the Lajas Experiment Station herd, were brought to the Gurabo Experiment Station. Each calf was identified by a numbered ear tag and a colored neck chain.

The animals were examined for ectoparasites and treated orally with thiabendazole for gastrointestinal parasites. Twenty calves (10 replicates of 2) used in this study were kept indoors in individual raised cages

[1] Manuscript submitted to Editorial Board November 21, 1983.

[2] Thanks are expressed to R. A. Barrick, M. A., for the statistical analysis.

[3] De León, D. D., Ritchie, L. and Chiriboga, J., 1971. Programa de *Fasciola hepatica* en Puerto Rico. First Report, January 1970 to April 1971, PRNC-148.

[4] Chiriboga, J., De León, D. and Rodríguez-Frías, J., 1980. Epidemiology of *Fasciola hepatica* infestation in dairy cattle at Dorado, Puerto Rico, J. Agric. Univ. P. R. 64 (1): 93–106.

[5] Mrozik, H., Bochis, R. J., Eskola, P., Matzuk, A., Waksmunski, F. S., Olen, L. E., Schwartz-Kopf, G., Grodski, A., Linn, B. O., Lusi, A., Wu, M. T., Shunk, C. H., Peterson, L. H., Milkowski, J. D., Hoff, D. R. Kulsa, P., Ostlind, D. A., Campbell, W. C., Riek, F. R., and Harmon, R. E. New fasciolicides with benzenesulfonamide structures, Abstr. of Papers 173rd Nat. Am. Chem. Soc., New Orleans, March 20–25, 1977, Meet. MEDI-15.

[6] De León, D. and Quiñones, R., 1983. The efficacy of MK-401 (4-amino-6 trichloroethenyl-1, 3-benzenedisulfonamide) against *Fasciola hepatica* in cattle under Puerto Rican conditions, J. Agric. Univ. P. R. 67 (1): 53–5.

during the preliminary period and until the second week following treatment. The large size of the calves at this time made it necessary to place them in a *F. hepatica*-free pen for the final 7 days prior to slaughter.

The calves were fed hay and concentrates free choice during the entire period of observation. Water was continuously available in buckets and was changed twice daily.

On March 9, 1982, 10 weeks before treatment, each calf was given orally, via balling gun, a sealed gelatin capsule that contained about 500 *F. hepatica* metacercariae.

Ten weeks after infection, 1 day before treatment, the calves were weighed and grouped by weight into pairs. The two calves of each pair were arbitrarily allocated to either the MK-401 or the placebo treatment, these being designated as the red and green groups, respectively. The pens and neck chains were identified with these colors. The two treatments were as follows:

1. A single dose of L-631,529-09N-A03, Vehicle, .083 ml/kg body weight orally (green group).
2. A single dose of L-631,529-13E01-MK-401, 7.0 mg/kg body weight orally (red group).

The treatments were administered May 18, 1982, 10 weeks after artificial infection.

On June 1, 1982, 2 weeks post-treatment and 12 weeks post-metacercariae infection, the calves were slaughtered. The livers were removed and identified and the bile conducts opened and examined for liver flukes.

The livers were then cut into small pieces of about 7.5 to 10 cm dimensions and placed in warm physiological saline solution. After 30 minutes the liver tissue was removed from the container and discarded. The physiological saline solution was filtered through a 40 mesh sieve and the fluke and fluke fragments (heads and tails) were collected and counted.

Fluke counts were obtained by adding the number of heads or tails, whichever was bigger, to the number of whole flukes found.

The section of duodenum into which the bile duct enters was removed, the ends tied and the contents examined for adult *F. hepatica.*

The carcass remains were saturated with creolin and buried.

Total fluke counts per animal were transformed to the natural logarithm of (count + 1) for calculation of geometric means and percent efficacy. The two groups were compared with the Wilcoxon rank-sum statistic in a randomization procedure which leads to an exact probability of finding a test statistic as extreme as or more extreme than the one observed. A two-sided significance level of .05 was used.

Fasciola hepatica eggs were detected in the feces of some of the calves

at 5 weeks after infection with metacercariae, however, treatment was delayed until 10 weeks post infection, at which time all the animals had a significant fecal egg populations. Fourteen days after treatment, *F. hepatica* eggs were detected in the feces of only 11% (1/9) of the MK-401 treated animals, compared to 70% (7/10) of the vehicle treated controls. At necropsy, moderate to severe *F. hepatica* infestation was observed in 100% (10/10) of the livers from the control group, while 90% (9/10) of the livers from the MK-401 treated group (7.0 mg/kg body weight) were parasite-free. Two flukes were found in the liver of one of the latter animals. The diaphragmatic lobe of the liver from another MK-401 treated animal contained an abscess approximately 5.0 cm in diameter, but there is no reason to incriminate the drug in the pathogenesis of this abscess.

Significantly ($P = .01$) fewer flukes were recovered from MK-401 treated animals than from the controls. The percent efficacy of this drug was 99.2.

No adverse clinical signs were observed in either group of animals.

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