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Effects of moxidectin on coprophagous insects in cattle dung pats in Japan

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Abstract

Effects of the antiparasitic drug moxidectin were studied in laboratory and field experiments in Hokkaido, Japan by pour-on administrations (500 μg/kg) on a target pest *Haematobia irritans* Linnaeus, nontarget coprophagous flies represented by *Neomyia cornicina* (Fabricius), and the dung beetle *Caccobius jessoensis* Harold. The concentration of moxidectin excreted into cattle dung was maximum at 3 days post-treatment both in the first and second trials, and then it diminished. No moxidectin was detected on or after day 21 post-treatment in the first trial, and on or after day 28 post-treatment in the second trial. Larval development of *H. irritans* was hampered from 1 to 7 days post-treatment. No *N. cornicina* pupated in dung at days 1 and 3 post-treatment, and pupation and emergence rates were reduced in the dung until 7 days post-treatment. There were no significant differences in numbers and weight of brood balls constructed by *Caccobius jessoensis* in dung from treated and control cattle. Adult emergence rates of *C. jessoensis* on days 1, 3, 7, 14 post-treatments were not significantly different between control and treated groups, and more than 90% of adult emergence rates were demonstrated in both groups. In the field study using emergence traps, 3,433 (18 families) flies emerged from dung from untreated control cattle and 1,667 (16 families) flies emerged from dung from treated cattle. Notably, the number of Sciaridae spp. (first Experiment) and *Sepsis latiforceps* Duda and Sphaeroceridae spp. (second Experiment) significantly decreased in dung pats of treated cattle. From 48 dung pats in the field experiments, total dry weight of major coprophagous flies that emerged was 1,741.8 mg in dung from control cattle and 1,170.0 mg in dung from treated cattle, showing 32.8% reduction in treated dung. Emergence rates of *C. jessoensis* from brood balls recovered from soil beneath dung pats in the field experiments were not significantly different between dung from control and treated cattle on each sampling day (1, 7, 14, and 21 days) post-treatment.

Key words: Moxidectin; *Haematobia irritans*; *Neomyia cornicina*; *Caccobius jessoensis*; Japan

INTRODUCTION

Currently available endectocides include avermectins (ivermectin, doramectin, and eprinomectin) and milbemycins (moxidectin). It is well-known that these parasiticides are effective against ectoparasitic pests as well as endoparasites. In particular, ivermectin and moxidectin have been widely used in the world because of a convenient "pour-on" application method. It has been reported, however, that ivermectin persists in dung of cattle and inhibits the growth of coprophagous dung degrading insects, involving delays in dung degradation (Wall and Strong, 1987; Madsen et al., 1990; Fincher, 1992; Sommer et al., 1992; Strong, 1992, 1993; Strong and Wall, 1994; Floate, 1998; Floate et al., 2002, 2005). This phenomenon will increase the fouled grassland with reduction of available forage and reduce incorporation back into the soil by dung degrading insects, and consequently could lead to an adverse effect on the pasture ecosystem.

On the other hand, moxidectin has been reported as having a less significant effect on dung beetles and other dung-inhabiting insects (Fincher and Wang, 1992; Strong and Wall, 1994; Wardhaugh et al., 1996; Floate et al., 2001, 2002). However, the effects of moxidectin on nontarget dung-degrading insects have been studied in limited countries and areas.

In Japan, ivermectin is now widely used to control parasites in cattle, and its effects on non-target coprophagous insects have been investigated (Iwasa et al., 2005a, b, 2007). Although the consumption of moxidectin also has increased, no

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study has been performed to assess the effect of moxidectin on Japanese dung-degrading insects. The purpose of the present study was to estimate the effects of pour-on application of moxidectin on a target ectoparasitic pest, *Hematobia irritans* Linnaeus, and nontarget coprophagous flies represented by *Neomyia cornicina* (Fabricius), and on the reproduction and survival of the dung beetle *Caccobius jessoensis* Harold, which is an important dung-decomposing species in Hokkaido, Japan.

MATERIALS AND METHODS

Cattle used and dung collection. The experiments were conducted at the pasture of Obihiro University of Agriculture and Veterinary Medicine in Hokkaido, Japan. A 10% moxidectin preparation (Cydectin® Pour-on: Fort Dodge Animal Health, Overland Park, Kansas, USA) was applied to cattle at a dose of 500 μg/kg body weight by pour-on formulation. Treatments were made twice using a specialized container to two separate cattle groups.

First treatment. Treatment group: Five Japanese black cattle of 30 to 50 months of age weighing an average of 642±109.9 kg (moxidectin treatment on 28 May 2005). Control group: Five Japanese black cattle of 30 to 50 months of age weighing an average of 558±97.5 kg (no treatment). Both groups were raised in paddocks and fed with hay made of orchard grass, meadow fescue, and white clover.

Second treatment. Treatment group: Four Holstein cattle of 30 to 50 months of age and one Japanese black cow weighing an average of 638±94.0 kg (moxidectin treatment on 2 July 2005). Control group: Four Holstein-Friesian heifers/dry cattle and one Japanese black cow of 30 to 50 month of age weighing an average of 618±24.5 kg (no treatment). Both groups were raised on grazing land mostly with orchard grass, meadow fescue, and white clover.

Fresh dung samples were collected from treated and control groups on the preceding day of treatment and at 1, 3, 7, 14, 21, 28, and 35 days after treatments. In each collection, dung pats from 5 cows in each group were mixed and frozen at −20°C until used.

Determination of moxidectin concentrations. Determination of moxidectin in feces was performed by high-performance liquid chromatography (HPLC) following the method of Payne et al. (1995) with some modifications: 5 g of dung on each date after treatment were analyzed, and the extraction was carried out with a homogenizer (PHYSCOTRON, Microtec Co., Ltd.) at 10,000 rpm for one min instead of sonication, and the concentration was calculated from a calibration curve of standard solutions.

Laboratory bioassays.

Effects of moxidectin on pupation and adult emergence of Haematobia irritans Linnaeus and Neomyia cornicina (Fabricius). *Haematobia irritans* is a target species which is a blood-sucking pest fly of cattle, and its larvae inhabit dung pats in pastures in Hokkaido. Adult flies collected from cattle bodies or freshly voided dung in pastureland were allowed to lay eggs inside an aspirator or plastic tube (1.2 cm diameter×7.5 cm length) and then eggs obtained were placed on 10 g feces in plastic cups (8 cm diameter×4 cm depth) for hatching. Using the hair tip of a writing brush, we inoculated 30 hatched larvae of *H. irritans* onto 40 g of thawed dung in a plastic cup (5 cm diameter×3 cm depth), which was placed inside a larger plastic cup (8 cm diameter×4 cm depth) containing sawdust for pupation. These cups were maintained at 25°C (16L : 8D) and rates of pupation and adult emergence were recorded. Inoculations were replicated 4 times for each collection date of dung from control and treated cattle in the second treatment.

Neomyia cornicina (Fabricius) was chosen as a nontarget beneficial coprophagous species which is also commonly found in dung pats in Hokkaido. Eggs of *N. cornicina* were collected from the surface of fresh dung pats voided within 1 or 2 days in pastures, and they were placed on 10 g feces in a plastic cup (8 cm diameter×4 cm depth) for hatching. Using the hair tip of a writing brush, we inoculated 30 hatched onto 60 g of thawed dung in a plastic cup (5 cm diameter×4 cm depth), which was placed in a larger plastic cup (10 cm diameter×5.5 cm depth) containing sawdust for pupation. These cups were maintained at 25°C (16L : 8D), and the number of pupae were counted. The pupae obtained were transferred to another plastic cup (8 cm diameter×4 cm depth) for adult emergence. Inoculations were replicated 5 times for each collection date after treatment in Experiment 2.

Effects of moxidectin on reproduction and emer-

gence of the dung beetle Caccobius jessoensis Harold. Adults of *C. jessoensis* were collected in a pasture at Obihiro University and were brought to the laboratory. A 30 g fecal sample from treatment or control was placed on volcanic ash-soil (15 cm depth) in a glass container (12 cm diameter×18 cm depth). Three male and female pairs were placed on feces in each container which was covered with gauze, and maintained at 22°C, 16L : 8D. Fresh feces was replaced once a week for 4 (2nd Exp.) to 5 weeks (1st Exp.), and at that time volcanic ash-soil from each container was sieved to collect brood balls produced, and the number and weight of brood balls were recorded. Then these brood balls were placed in a plastic cup (5 cm diameter×3 cm depth) containing andosol, and kept at 22°C (16L : 8D) to record the adult emergence rate.

Field experiment.

Effects of moxidectin on fly emergence. Twenty-four artificial dung pats (700 g, three pats each per collection date for treated and control cattle) of 1, 7, 14, and 21 days after treatment were placed on a 18 cm layer of andosol in a plastic box (33×33×19 cm). Boxes with these dung pats were then placed in a field adjacent to the pasture in early July (Experiment 1) and early August (Experiment 2) for exposure to insect activity for 7 days. After exposure, these boxes were brought to a grassland on the University campus and covered with emergence trap nets attached with cups for collection of emerged flies. Traps were checked daily, and emerged flies from dung pats were collected, counted, and identified to species (Anthomyiidae, Muscidae, Sarcophagidae and Sepsidae) or families. These specimens were dried indoors for more than two months, and their dry weights measured with an electric balance (Sartorius BJ1500; Göttingen, Germany) as an indicator of assessment for degradation of dung pats.

Effects of moxidectin on brood ball construction and adult emergence rate of dung beetle. After 1 month, when fly emergence came to an end, the brood balls constructed by dung beetles were collected from soil beneath dung pats. Brood balls were kept at 22°C (16L : 8D) until adult emergence.

Data analysis. Differences between the control and treatment groups were analyzed by the non-parametric Mann-Whitney *U* test.

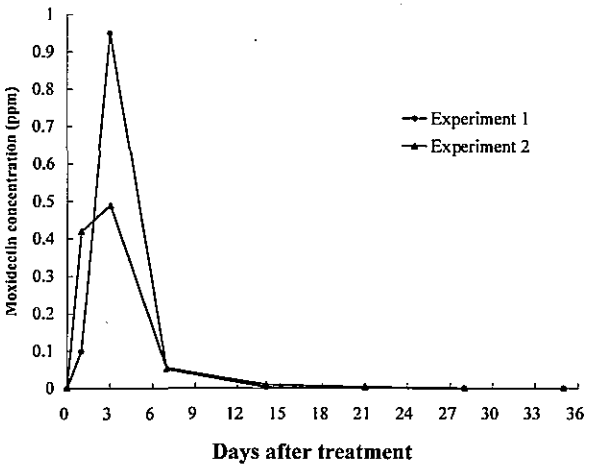

Fig. 1. Moxidectin concentration in dung (ppm of wet weight) after treatments (500 μg/kg).

RESULTS

Moxidectin concentrations in cattle dung

Concentrations of moxidectin in the feces reached a maximum level at 3 days after treatment in 2 trials, and samples in both trials showed a marked decline by 7 days (Fig. 1). In the first trial, the maximum concentration found on day 3 was approximately double the level of the same day in the second trial. The detected level on day 1 of the first trial was 0.099 ppm, which was approximately one-quarter that of day 1 in the second trial. No moxidectin was detected at 21 days or later in the first trial and at 28 days or later in the second trial.

Laboratory bioassays

Effects of moxidectin on pupation and adult emergence of Haematobia irritans Linnaeus and Neomyia cornicina Fabricius

Results of pupation rates and adult emergence rates of *H. irritans* and *N. cornicina* in dung from control and treated groups are presented in the second Experiment (Table 1). Because the dung pats used in the first trial were crusty and hard throughout, larvae did not develop normally. Pupation rates of *H. irritans* in dung from the control group ranged from 59.2 to 95.8% at 1 to 35 days after treatment, whereas in dung from treated cattle they were 0.8% at 1 day, 0% at 3 days, and 25% at 7 days, showing marked difference by day 7. There were no significant differences in pupation rates between control and treated cattle from days 14 to

35 post-treatment. Adult emergence rates showed a similar pattern to that obtained with pupation rates. Pupation rates of *N. cornicina* in dung of control cattle were high, ranging from 81.3 to 98.7% on all days post-treatment. However, no pupation was noted at 1 and 3 days post-treatment and a significant reduction was found in the treatment at 7 days post-treatment compared to control. There were no significant differences in the pupation rates from day 14 to 35 post-treatment between control and treated cattle. Adult emergence rates also showed a similar trend to that of pupation rates. In dung of treated cattle no great difference between pupation and emergence rates in *H. irritans* and *N. cornicina*

Table 1. Larval and pupal survival of *H. irritans* and *N. cornicina* in dung from treated and control cattle (Experiment 2)

Days after treatment	*Haematobia irritans*				*Neomyia cornicina*			
	% Pupation (Mean±SE)[a]		% Emergence (Mean±SE)		% Pupation (Mean±SE)[b]		% Emergence (Mean±SE)	
	Control	Treatment	Control	Treatment	Control	Treatment	Control	Treatment
1	81.7±7.9	0.8±1.7*	73.3±11.2	0.8±1.7*	90.7±6.4	0±0*	88.7±9.0	0±0*
3	59.2±5.7	0±0*	56.7±2.7	0±0*	81.3±7.3	0±0*	73.3±11.3	0±0*
7	82.5±12.0	25.0±19.9*	75.0±13.7	20.0±17.9*	98.0±1.8	68.7±10.7*	80.8±12.0	50.7±12.3*
14	79.2±5.7	65.8±12.0	65.0±4.3	58.3±9.6	94.7±3.8	98.0±1.8	89.4±4.6	94.1±4.3
21	95.8±5.0	95.8±5.0	89.2±6.9	80.8±16.6	84.7±15.0	96.0±4.4	80.0±16.3	91.3±3.0
28	55.0±42.0	60.8±28.9	53.3±40.4	55.0±25.8	98.7±1.8	88.7±1.6	97.3±2.8	84.0±19.2
35	63.3±21.1	71.7±20.6	58.3±16.7	62.6±16.2	92.0±3.8	92.7±8.3	82.7±8.6	83.3±16.2

[a] Each value is a mean of four replicates each 30 larvae.
[b] Each value is a mean of five replicates each 30 larvae.
* Significantly different between control and treatment in pupation and emergence rates, respectively ($p<0.01$: Mann-Whitney U test).

Table 2. Numbers and weights of brood balls, and emergence rates in *Caccobius jessoensis* reared in dung from treated and control cattle in Experiments 1 and 2

Days after treatment	Experiment 1		Experiment 2	
	Control	Treatment	Control	Treatment
	No. of brood balls/female±SE			
1	1.60±1.53 (24)[a]	2.47±1.54 (37)	1.67±1.03 (25)	3.07±1.98 (46)
3	2.40±1.50 (36)	3.87±2.85 (58)	3.93±2.07 (59)	3.93±1.38 (59)
7	3.60±0.77 (54)	2.00±1.83 (30)	1.33±0.82 (20)	1.67±0.91 (25)
14	0.60±0.83 (9)	1.87±1.28 (28)	2.40±0.72 (36)	2.87±1.88 (43)
	Weight of brood balls (g)±SE			
1	1.3±0.14	1.6±0.34	1.2±0.18	1.2±0.35
3	1.5±0.09	1.6±0.11	1.2±0.10	1.1±0.20
7	1.6±0.28	1.6±0.31	1.2±0.22	1.0±0.21
14	1.5±0.20	1.4±0.09	1.3±0.19	1.2±0.07
	Emergence rate (%)±SE			
1	91.7±16.67	97.30±3.44	92.0±12.16	100±0
3	97.2±6.39	98.28±2.50	94.92±3.84	94.92±14.9
7	94.2±7.21	96.67±3.57	95.0±11.18	92.00±9.08
14	77.8±19.3	82.14±11.9	97.22±6.39	95.35±6.84

[a] Values in parentheses are total numbers of brood balls.
No significant difference between control and treatment within the lines in each experiment ($p>0.05$: Mann-Whitney U test).

shows that moxidectin have larvicidal activity against the two species, not emergence inhibition.

Effects of moxidectin on brood ball construction, adult survival, and emergence rate of Caccobius jessoensis Harold

Average numbers, weights, and emergence rates of brood balls per female *Caccobius jessoensis* Harold recorded at 1, 3, 7, and 14 days post-treatment in the control and treated groups are presented in Table 2. There were no significant differences in numbers of brood balls per pair and average weights of brood balls on any day post-treatment between control and treated cattle in two experiments. Emergence rates were more than 90% on all days except at days 14 of control and treated cattle in experiment 1, and no significant difference was noted between control and treated cattle throughout experiment.

Field experiment

Effects of moxidectin on fly emergence and brood ball construction of dung beetles

In field experiments using the emergence traps, a total of 5,100 flies from 18 families emerged from 48 dung pats (700 g per pat) exposed to insect activity for a week—3,433 flies in dung of control cattle and 1,667 flies in dung of treated cattle (Table 3). Those identified to species were *Paregle*

Table 3. Numbers of flies (±SE) emerged from 3 dung pats (1 pat, 700 g) of control and treated cattle in exposure for 1 wk in the field

Families and species	Experiment 1		Experiment 2	
	Control	Treatment	Control	Treatment
Anthomyiidae				
Paregle cinerella (Fallén)	1.3±0.9	6±6.0	3.7±3.4	2.7±1.8
Emmesomyia sp.	0±0	0±0	0±0	0.3±0.2
Muscidae				
Musca bezzi Patton et Cragg	2.8±1.8	0.8±0.4	0.3±0.2	1.3±0.4
Neomyia cornicina (Fabricius)	0±0	0±0	41.2±41.2	34.8±30.8
Myospila meditabunda (Fabricius)	1.3±0.7	0.5±0.3	1±0.4	1±1.0
Hydrotaea albipuncta (Zetterstedt)	1.5±1.5	1.5±1.2	0.3±0.2	10.3±0.2
Coenosia sp.	0.5±0.5	0±0	1.3±0.7	1±0.4
Sarcophagidae				
Ravinia striata (Fabricius)	0±0	0.8±0.7	2.5±2.5	4.5±2.6
Sepsidae				
Sepsis latiforceps Duda	37.5±23.4	34±18.5	119.2±32.6	22±9.2*
Sepsis thoracica (Robineau-Desvoidy)	0±0	0±0	4.3±1.8	2.3±1.3
Sepsis duplicata Haliday	0.8±0.7	0±0	0±0	0.3±0.2
Sepsis cynipsea (Linnaeus)	0±0	0±0	4.5±3.8	0.3±0.25
Saltella sphondylii (Schrank)	0±0	0±0	0.5±0.5	0±0
Sphaeroceridae spp.	11.5±0.6	6.5±2.9	199±76.5	18.5±12.3*
Drosophilidae spp.	0.8±0.4	0.3±0.2	2.3±0.7	1.3±0.9
Milichiidae spp.	0±0	0±0	0.3±0.2	0±0
Chloropidae spp.	0±0	0.5±0.3	1±0.4	10.3±0.2
Ephydridae spp.	0±0	0±0	5.5±4.2	0±0
Phoridae spp.	5±0.8	3.5±1.5	6.3±2.9	3.8±1.1
Dolichopodidae spp.	0.5±0.3	0.3±0.2	0.8±0.3	0.8±0.4
Phoridae spp.	5±0.8	3.5±1.5	6.3±2.9	3.8±1.1
Empididare spp.	1.5±0.8	1.7±0.7	1.8±0.4	1.3±0.4
Chironomidae spp.	0±0	0±0	1±1.0	0.5±0.5
Ceratopogonidae spp.	2±2	0±0	50.2±20.9	47.2±27.1
Sciaridae spp.	267.7±21.4	169.7±21.5*	59.2±10.8	44.7±12.9
Mycetophilidae spp.	0±0	0±0	0.5±0.5	0.3±0.2
Psychodidae spp.	0±0	0.3±0.2	11±7.7	0±0
Cecidomyiidae spp.	2.2±0.6	1.3±0.6	4±2.0	1±0.4
Total (12 dung pats)	1,347	907	2,086	760

* Significantly different versus control ($p<0.05$: Mann-Whitney U test).

cinerella Fallén of Anthomyiidae; *Musca bezzii* Patton et Cragg, *Neomyia cornicina* Fabricius and *Myospila meditabunda* Fabricius of Muscidae; *Ravinia striata* Fabricius of Sacrophagide; and *Sepsis latiforceps* Duda, *Sepsis thoracica* Robineau-Desvoidy, *Sepsis duplicata* Haliday, *Sepsis cynipsea* Linnaeus, and *Saltella sphondylii* Schrank of Sepsidae. The predominant families and species in abundance were Sciaridae (2,166), followed by Sphaeroceridae spp. (942), *Sepsis latiforceps* (851), and Ceratopogonidae spp. (390). Adult emergences of Diptera were comparatively small as a whole, and no significant difference in numbers between control and treated groups was recorded in many species and families, but emergences of Sciaridae spp. in the first experiment and *Sepsis latiforceps*, Sphaeroceridae spp. in the second experiment were significantly reduced in dung from treated cattle.

Total dry weights of main coprophagous flies that emerged in the 2 trials were 1,741.8 mg in dung of control cattle and 1,170.0 mg in dung of treated cattle; that is, the weights of flies from dung of treated cattle were reduced to 67.2% of those of the control cattle (Table 4). Dry weights of total flies emerged from dung pats voided by treated cattle were significantly smaller than those of dung from control cattle in *Sepsis latiforceps*, Sphaeroceridae spp., and Sciaridae spp.

Brood balls recovered from soil beneath dung pats after termination of fly emergence were those of *Caccobius jessoensis* and *Liatongus minutus*. Only data of *C. jessoensis* were presented in Table 5, because the number of *L. minutus* was so small. Adult emergence rates of *C. jessoensis* were not significantly different on all days (1, 7, 14, and 21) after treatment between control and treated cattle.

Table 4. Dry weights (±SE) of main coprophagous flies emerged per 6 dung pats voided by control and treated cattle

Families and species	Dry weight (mg)	
	Control	Treatment
Anthomyiidae		
Paregle cinerella	2.7±2.3	4.2±3.7
Muscidae		
Musca bezzi	31.6±20.2	21.1±7.4
Neomyia cornicina	168.3±168	141.7±125
Sarcophagidae		
Ravinia striata	19.3±19.3	40.8±23.4
Sepsidae		
Sepsis latiforceps	62.7±8.6	22.4±9.7*
Sepsis thoracica	1.7±0.7	0.9±0.5
Sepsis duplicata	0.1±0.1	0.03±0.04
Sepsis cynipsea	1.8±1.5	0.1±0.1
Saltella sphondylii	0.2±0.2	0±0
Sphaerocedae spp.	65.2±23.9	7.7±3.8*
Sciaridae spp.	81.7±7.5	53.6±8.0*
Total (24 dung pats)	1,741.8	1,170.0

* Significantly different versus control ($p<0.05$: Mann-Whitney U test).

Table 5. Emergence rates of *Caccobius jessoensis* from brood balls recovered from soil beneath dung pats of control and treated cattle after 7 days exposure in the field

Days after treatment	% Emergence±SE	
	Control	Treatment
1	71.4±12.5 (14)	60.0±16.3 (10)
7	61.5±14.0 (13)	80.0±20.0 (5)
14	45.5±15.7 (11)	50.0±50.0 (2)
21	16.7±16.7 (6)	38.5±14.0 (13)

Values in parentheses are total numbers of brood balls recovered.
No significant difference between control and treatment within the lines in each experiment ($p>0.05$: Mann-Whitney U test).

DISCUSSION

Zulalian et al. (1994) reported that a peak concentration of moxidectin in the dung of cattle was detected within 3 days after subcutaneous injection at 0.2 mg/kg. It may be conceivable that a pour-on formulation also causes approximately similar results as the highest concentration was reached at day 3 post-treatment and subsequently decreased in this trial. This result is different from those where residual concentration of ivermectin in dung by pour-on had a peak at 1 day post-treatment (Sommer et al., 1992; Strong and James, 1992; Iwasa et al., 2005a, b, 2007). Maximum detectable levels of moxidectin in the dung were 0.95 ppm on day 3 in the first trial and 0.49 ppm on day 3 in the second trial of the present study. These levels were much higher than those obtained from trials carried out in Obihiro over the past 3 years with ivermectin pour-on formulations (Iwasa et al., 2005a, b, 2007). It is not surprising that the timing and concentration of fecal excretion of drugs would be different

between moxidectin and ivermectin in cattle. Cook et al. (1996) reported that concentration of residual ivermectin in the dung of cattle can be affected by the diet of cattle. The marked difference observed in maximum fecal residual levels of moxidectin at day 3 post-treatment in the first and second trials could be attributable to the difference in diets even in our study; cattle were mainly fed with orchard grass hay in the first trial, while cattle were fed with green pasture grass in the second trial. The difference of drug concentration in fecal residues could also be associated with the difference in fecal moisture contents between the two trials. Average moisture contents of cattle dung pats were 83.6% in the first trial versus 86.9% in the second trial.

Results of the laboratory bioassay using *Haematobia irritans* in which 0.8% (only 1 out of 120 larvae) pupated on day 1 post-treatment in the treated group suggested that a critical concentration of moxidectin (to allow pupation of fly larvae) would exist at a level between 0.42 ppm and 0.49 ppm. In dung of cattle treated by pour-on formulation of ivermectin, larval survival of *H. irritans* was reduced for 5–6 weeks (Fincher, 1996), 4–8 weeks (Floate et al., 2001), and 3 weeks (Iwasa et al., 2005a). Floate et al. (2001) also reported that the pour-on treatment of moxidectin suppressed development of *H. irritans* for a week. So, the inhibitory effects of moxidectin on the pupation and emergence of *H. irritans* were at least 2 or 3 weeks shorter when compared to those of ivermectin. In the present study, normal pupation and emergence which were equivalent to the levels of the control group were restored by day 14 post-treatment. Floate et al. (2001) further reported that the pour-on administration of moxidectin did not affect the adult emergence of *Musca domestica* Linnaeus, but ivermectin inhibited the adult emergence over a period of 5 weeks. According to Wardhaugh et al. (1996), no inhibitory effects on the adult emergence of *M. domestica* were observed from subcutaneous injection of moxidectin except on day 14 in the results of an experiment using dung pats collected on days 3, 7, 14, 28, and 35 post-treatment. On the other hand, no adult emergence was observed from day 3 through day 14, and normal pupation and emergence which were equivalent to the levels of control group were not restored until day 35 after treatment with ivermectin. It is fair to say from these reports that moxidectin was significantly less efficacy for control of target flies of Muscidae, and it has a shorter duration of activity in comparison with ivermectin.

It has been reported that ivermectin severely inhibits larval development of nontarget dung-breeding flies (Schmidt, 1983; Wall and Strong, 1987; Floate, 1998; Floate et al., 2002; Iwasa, 2005a, b). *Neomyia cornicina* can be considered be beneficial because of the role it plays in dung degradation in Europe and also Japan. Mortality of *Neomyia cornicina* larvae was 100% up to 32 days post-treatment by injection (Wardhaugh and Rodriguez-Menendez, 1988) and 97% up to 14 days post-treatment by pour-on in dung of ivermectin-treated cattle (Sommer et al., 1992). Iwasa et al. (2005a) reported that no pupation or adult emergence of *N. cornicina* was observed by day 14; inhibitory effects of medication were observed even on day 21, and normal growth was restored by day 28 post-treatment in the result of a laboratory test using dung of ivermectin-treated cattle. Also, the inhibitory effects of moxidectin on the pupation and emergence of *N. cornicina* were 2 weeks shorter as compared to those of ivermectin.

Strong and Wall (1994) reported that no Cyclorrhapha larva was observed on days 2, 7, or 14 post-treatment in dung of ivermectin-treated cattle, and the number of larvae found on day 21 was much fewer than those of the control and moxidectin-treated groups. They also showed that the number of larvae found on any observation day post-treatment in the moxidectin group was not significantly different from that of the control group. In the present study on fly species that emerged from dung pats in the field, no significant difference in numbers of emergence between control and treated groups was seen in many species and families, showing that moxidectin is less toxic also against many other nontarget coprophagous flies. However, the number of Sciaridae spp. in the first Experiment and *Sepsis latiforceps* and Sphaeroceridae spp. in the second Experiment significantly decreased in dung of treated cattle, so perhaps they were influenced by high residual levels in the dung after moxidectin treatment.

Nematocera flies have been shown to be unaffected by ivermectin in some reports (Schmidt, 1983; Madsen et al., 1990; Sommer et al., 1992; Strong, 1992), but Iwasa et al. (2005a, b) showed

emergence of some families of Nematocera has increased in dung of treated cattle. Iwasa et al. (2005b) also suggested a possibility for increased emergence of some Nematocera in the treated group through the growth inhibition of other fly larvae produced by ivermectin residues in cattle dung. However, there may be no such observation in the Nematocera in the dung of cattle treated with moxidectin.

Papp (1970) investigated the level of productivity of Muscidae, Sepsidae, and Sphaeroceridae and specifically reported on the high dung-decomposing ability of Muscidae having a larger body size. Dry body weights of major coprophagous flies in dung of treated cattle were reduced to 5.9% (Iwasa et al., 2005a) and to 21.7% (Iwasa et al., 2005b) of those of the control cattle in field trials using ivermectin. The present dry weights of major coprophagous flies in dung of treated cattle were reduced to 67.2% of those of the control cattle, and the effect of moxidectin against flies taking an active part as dung decomposers in the field is fairly small in comparison with that of ivermectin.

Larval survivals of dung beetles *Onthophagus*, *Euoniticellus*, *Aphodius*, *Onitis*, and *Copris* are known to decrease in dung from cattle treated with ivermectin or abamectin (Ridsdill-Smith, 1988; Wardhaugh and Rodriguez-Menendez, 1988; Roncalli, 1989; Madsen et al., 1990; Fincher, 1992, 1996; Sommer and Nielsen, 1992; Lumaret et al., 1993; Krüger and Scholtz, 1997; Dadour et al., 2000; Errouissi et al., 2001; Wardhaugh et al., 2001). On the other hand, Fincher and Wang (1992) reported that the dung of cattle injected subcutaneously with moxidectin at 0.2 mg/kg did not have any effect on the mortality rate or emergence rate of *Euoniticellus intermedius* (Reiche) and *Onthophagus gazella* (Linnaeus). Strong and Wall (1994) also didn't detect inhibitory effects on the development of *Aphodius* spp. in dung from moxidectin-treated cattle. When Doherty et al. (1994) investigated the effects of ivermectin and moxidectin directly added to the dung of cattle on the growth of *O. gazella* larvae, ivermectin at 0.008 ppm inhibited adult emergence by 95% and at 0.016 ppm by 100%, while, moxidectin at 0.512, 0.256, 0.008, and 0.016 ppm inhibited adult emergence by 93, 39, 19, and 11%, respectively. Since no effect of moxidectin was observed in adult emergence rates of *Caccobius jessoensis* (in control and treated groups on any date of the first or second trial in the laboratory), and since fecal moxidectin residue reached a level of 0.95 ppm, perhaps the tolerance of *C. jessoensis* to moxidectin is equivalent to or larger than that of *O. gazella*. Iwasa et al. (2007) tested effects of ivermectin by pour-on treatment using *C. jessoensis* and reported that adult emergence rates were 0% at a concentration of 0.15 ppm, 24% at a concentration of 0.13 ppm and 77.8% at a concentration 0.085 ppm. These results indicate that the toxicity of moxidectin to *C. jessoensis* is much lower than that of ivermectin. Further, a recommended dose rate of moxidectin of 500 μg/kg did not produce any effect on *C. jessoensis*, since there were no significant differences in average number and weight of brood balls between control and moxidectin treated groups. These results agreed with those of emergence rates of *C. jessoensis* in the field experiment (Table 5). However, further work is necessary to understand the subsequent effects on adult dung-beetles emerged from dung pats of cattle medicated with moxidectin.

Moxidectin has been reported to be the least toxic compound for dung-inhabiting insects such as flies and dung-beetles as compared to other endoectocides such as doramectin, ivermectin, and eprinomectin (Doherty et al., 1994; Wardhaugh et al., 1996, 2001; Floate et al., 2001, 2002). The present study demonstrated that moxidectin produced no or at most the least effect against nontarget coprophagous insects commencing with local varieties of *C. jessoensis* and *N. cornicina*. Moxidectin is used to treat on cattle 2–3 times from spring to autumn in Japan; this frequency in treatments probably at least have no short-term effects on many coprophagous insects in dung pats. The present results may have importance to livestock pest management in Japan. Further work will be necessary to select an appropriate endectocide for many more coprophagous insects and set up an application regimen toward environmental conservation in cattle grazing pasture.

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