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Efficacy and Nontarget Effects of Larvadex® as a Feed Additive for Controlling House Flies in Caged-Layer Poultry Manure[1]

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ABSTRACT The insect growth regulator N-cyclopropyl-1,3,5-triazine-2,4,6 triamine (Larvadex®, CGA 72662, cyromazine) was provided as a feed additive (.3% Premix per ton of feed) to caged laying hens under field conditions in high rise, wide span and narrow poultry houses. The chemical effectively controlled house flies *(Musca domestica)* and soldier flies *(Hermetia illucens)*. The feed additive had no adverse effect on the populations of manure-inhabiting mites (Macrochelidae and Uropodidae) and histerid beetles *(Carcinops pumulio)*, which prey on fly eggs and larvae. Satisfactory fly control was demonstrated by use of the additive 50% of the time when the interval without the additive in the feed was 4 days but not when the interval was 7 days. Use of a fly monitoring program to time the use of the feed additive is advocated.

(*Key words:* insect growth regulator, Larvadex®, cyromazine, flies, feed additive, mites, *Carcinops* beetles)

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INTRODUCTION

House flies *(Musca domestica* L.) breed prolifically in poultry manure, especially in the accumulations under caged laying hens. Fly control in caged layer operations is difficult and requires an integrated approach using proper manure management, enhancement of populations of biological control agents (fly predators and parasites), and selective use of insecticides (Axtell, 1970a, 1981). If the caged layer production system does not provide for complete manure removal every few days, then the accumulated manure should be as dry as possible to discourage fly production and to provide a physical environment conductive to the survival and multiplication of manure-inhabiting mites and beetles, which prey upon fly eggs or first-instar larvae. The major predacious mites in poultry manure are *Macrocheles muscaedomesticae* (family Macrochelidae) and *Fuscuropoda vegetans* (family Uropodidae) (Willis and Axtell, 1968). The major beetle is *Carcinops pumulio* (family Histeridae) (Pfeiffer and Axtell, 1980). Selective applications of insecticides, such as residual applications to building surfaces and toxicant bait stations, directed against only the adult stage of the fly, and the avoidance of extensive larviciding of the manure have been recommended in order to minimize any adverse effects of the insecticides on the manure-inhabiting, beneficial mites and beetles. Generally, the insecticides used for fly control are toxic to predacious arthropods (Axtell, 1966, 1968, 1970b).

If a feed additive is used for the control of fly breeding in poultry manure, the possibility of adverse effects on manure-inhabiting, predacious mites and beetles should be considered. Therefore, field experiments were conducted to determine the efficacy for house fly control and the effects on manure mites and beetles of the feed additive N-cyclopropyl-1,3,5-triazine-2,4,6-triamine, which is also known as CGA-72662, cyromazine, and Larvadex® (Ciba-Geigy Corp., Greensboro, NC). This chemical has been shown to be effective against the house fly, although the mechanism of toxicity is uncertain (Hall and Foehse, 1980; Williams and Berry, 1980; Miller and Corley, 1980; Miller *et al.*, 1981). The chemical interferes with the normal development of fly larvae and is therefore broadly classified as an insect growth regulator. Its biological activity is thought to be limited mainly to dipterous insects. Preliminary tests (Axtell, unpublished) by spraying onto the manure under caged laying hens indicated no apparent effects on free living mites and beetles.

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MATERIALS AMD METHODS

The Premix A of Larvadex (.3% active ingredient) was furnished by Ciba-Geigy Corp., Greensboro, NC. Tests were conducted on farms having feed mills, and the premix was added according to label instructions (1 lb premix per ton feed). All farms had caged White Leghorn laying hens. The feed containing the premix was fed continuously during the periods of the experiments unless specified otherwise.

Experiment 1. The feed additive was provided continuously from August 2 to October 6, 1980 to *ca.* 30,000 laying hens in a high-rise ("deep pit") type of house in Davidson County, NC (Sink Farm). At the beginning of the experiment there was a high population of house flies, and a piped-in pyrethrin aerosol system was being used several times a week for temporary adult fly control. The manure had accumulated for *ca.* a year and was very wet, flattened, and heavily infested with soldier fly larvae, *Hermetia illucens* (L.).

Evaluation of the abundance of house flies was by baited jug traps and spot cards. The trap was a 1-gal plastic jug containing 25 g of Improved Golden Malrin® fly bait (Zoecon Corp.) and with four openings (3 cm diameter) around the upper third circumference of the jug. There were six baited jug traps hung above the top cage level (three at each end of the house). The number of flies in the trap were counted weekly, the traps cleaned, and fresh bait added. The spot cards (to measure fly activity) were white 3 × 5 paper file cards attached to the rafters. The fecal and regurgitation spots left by the flies were counted weekly and the cards replaced.

Experiment 2. The feed additive was provided continuously from August 2 to October 6, 1980 to *ca.* 30,000 caged laying hens contained in seven adjacent narrow ("California") type houses in Davidson County, NC (Sink Farm). These houses had a center concrete aisle with two-tiered cages on each side and open sides. The manure accumulated on the soil beneath the cages. All the manure was removed a few days prior to beginning the use of the feed additive. Until that time the houses had a very high population of house flies, and pyrethrin was misted several times a week. Also, there was a very high population of soldier fly larvae in the manure, and the manure was wet and spilling onto the walkways.

House fly abundance was evaluated by seven baited jug traps (one in the entrance way of each house). After 52 and 59 days of feed additive use, manure fauna was sampled. This was done by taking a composite 1-gal sample of manure from 10 locations in each house and extracting the mites and insects by means of Tulgren funnels, which subjected the manure to slow drying under a 60-W incandescent bulb in order to drive the organisms downward into 70% alcohol in a collecting jar.

Experiment 3. The feed additive was provided continuously from June 5 to October 12, 1981 at a farm (Jones) in Alamance County, NC. There were about 40,000 caged laying hens contained in two narrow houses and two widespan houses. The narrow houses had a single center aisle with two-tiered cages on each side while the widespan houses had four aisles with two-tiered cages on each side. All houses had plastic side curtains. The four houses were adjacent and *ca.* 10 m apart.

Evaluation of adult fly populations was by three baited jug traps (one in each narrow house and one in a wide-span house). Manure samples were collected at *ca.* 2-week intervals and the manure-inhabiting fauna extracted by Tulgren funnels. At the treated farm a composite sample of *ca.* 2 liters of manure was taken from 10 locations on each side of the houses (total of six samples). In order to have an untreated comparison, manure samples were taken from another untreated farm with narrow cage houses (8 composite samples at 2-week intervals within 2 or 3 days of the corresponding samples taken from the Jones farm). Data on adult fly numbers were not obtained at the untreated farm.

The data were transformed to log (x + 1) and analyzed using the general linear model and Duncan's multiple range procedure to determine significant differences (Barr *et al.*, 1979).

Experiment 4. The feed additive was provided in a replicated experiment in a part of a narrow caged layer curtain-sided house on the NCSU Research farm, Wake County, NC. The house had a center aisle with two-tiered step cages on each side. The portion of the house used in this experiment was divided by partitions into four sections, each containing 120 birds (60 on each side of the aisle). The feed containing the additive was provided to one side in each section and the opposite side received feed with no additive. The positions of the treated and untreated were alternated in the four sections.

Adult fly populations could not be evaluated because only a part of the poultry house was included in this experiment, and the experiment contained untreated as well as treated birds. Evaluation was by extraction of the fauna from weekly composite manure samples (2 liters per replicate) extracted with Tulgren funnels. The data were combined for a 2-week period so there were 8 manure samples for the treated and the untreated for each 2-week interval pre and posttreatment.

The data were transformed to log (x + 1) and analyzed using the general linear model and Duncan's multiple range procedure to determine significant differences (Barr *et al.*, 1979).

Experiment 5. The use of the feed additive began June 7, 1982 after the fly population had reached a high level at houses described in Experiment 3. The feed additive was provided initially for 7 days alternated with 7 days of no use. Later this was changed to use of the additive for four days alternated with four days of no use. Monitoring of the fly population was by baited jug traps (eight traps, two per house) and spot cards (eight cards, two per house) as previously described.

TABLE 1. *Effect of feeding caged-layer hens Larvadex® continuously in the feed on the abundance of house flies in a high-rise house (30,000 birds). Feed additive used August 2 to October 8, 1980. (Experiment 1)*

No. days fed chemical	Mean no. flies per trap per day[1]	Mean no. spots per card per day[1]
4	151.1	9.0
11	143.3	4.0
17	20.4	1.4
24	22.4	.8
31	27.4	1.3
37	34.8	1.0
44	33.7	1.1
52	45.1	2.0
59	19.4	1.1
64	14.0	.4

[1] n = 6.

TABLE 2. *Effect of feeding caged-layer hens Larvadex® continuously in the feed on the abundance of house flies in 7 narrow open-sided houses (30,000 birds). Feed additive used August 28 to October 8, 1980. (Experiment 2)*

No. days fed chemical	Mean no. flies per trap per day[1]	Mean no. spots per card per day[1]
4	6.8	.2
11	5.0	.3
17	5.1	.2
24	6.1	0
31	4.7	.1
37	6.7	0
44	6.4	.1
52	7.6	.2
59	3.1	.2
64	1.7	.2

[1] n = 7.

RESULTS

Experiment 1. Following the continuous feeding of Larvadex in the laying hen ration in the high rise poultry house, the house fly population declined rapidly. After 11 to 17 days of using the feed additive, the flies were significantly reduced, based on trap and spot counts (Table 1). A slight rise in flies in September (37 to 52 days of feeding) occurred, which reflects the usual increases in house flies during that month in this region of North Carolina. Concurrently with the reduced numbers of house flies in this experiment, it was observed that the soldier fly larvae *(Hermetia illucens)* in the manure became sluggish and began to die slowly. As the numbers of soldier fly larvae declined, the manure dried more effectively and began to accumulate in piles under the cage rows rather than being wet and flattened.

Experiment 2. A few days before beginning the use of the feed additive the manure in the seven narrow houses was removed due to its wet condition and large population of fly larvae. With the continuous use of the feed additive for the remainder of the fly season the fly population was kept low (Table 2). Most trap counts were below 7 flies per day and spot counts were less than 1 per day. Fly control was greater in these narrow houses than in the high rise house because the manure was removed from the narrow houses at the begin-

TABLE 3. *Effects of using the poultry feed additive Larvadex® on abundance of adult house flies and manure-inhabiting house fly larvae and beneficial mites (Macrocheles muscaedomesticae and Furcuropoda vegetans) and beetles (Carcinops pumulio) in caged layer houses. Feed additive used June 5 to October 1, 1981. (Experiment 3)*

Organism[1] and treatment	No. days fed chemical									
	0	5	21	35	47	61	75	87	101	115
Adult flies										
Treated	150.0	283.3	76.2	16.0	14.6	15.9	9.4	8.2	.9	.9
Fly larvae										
Treated	89.2*	22.0	76.5	30.0	4.5*	0*	0*	.2*	0*	10.3*
Untreated	32.6	32.1	99.4	14.0	385.7	535.0	85.0	359.4	531.2	394.7
M. muscaedomesticae										
Treated	32.0	113.2	94.5	223.8*	544.3*	857.0*	247.3*	291.0*	724.3*	119.0
Untreated	36.0	73.6	79.5	20.4	239.0	71.2	67.0	39.7	91.6	79.5
F. vegetans										
Treated	0	146.5	18.2*	457.2	1413.7	1720.3*	1951.0*	1272.7*	1537.0*	237.8
Untreated	5.0	34.9	143.6	265.2	898.2	677.7	660.2	168.2	332.7	255.0
C. pumulio										
Treated	.2*	7.3*	30.8	11.5	7.5*	20.0	30.5	35.2*	23.0*	6.8
Untreated	4.9	31.2	37.3	12.4	28.9	28.7	26.6	3.7	5.1	3.5

[1] Data for adult flies are mean number per trap per day; for others data are mean number recovered from 2 liters of manure.

*Means for the treated and untreated for that organism on the given sampling day are significantly different ($P \leq .05$).

ning of the experiment but not from the high rise.

During this experiment the manure was examined for soldier fly larvae and very few were found. The manure accumulated in piles under the cage rows and effectively dried on the surface.

Samples of the manure were extracted by Tulgren funnels to determine the presence of fly larvae and predacious mites and beetles. Seven 4-liter composite manure samples (none from each house) were taken after 52 days of using the feed additive and the same number of samples after 59 days. No larvae of the house fly were recovered. There were very few soldier fly larvae ($\overline{X}$ = 4.8 ± 5.1 per sample in first set and 2.7 ± 2.4 per sample in the second set of samples). The mean number of the macrochelid mite, *Macrocheles muscaedomesticae,* was 86.4 ± 52.0 in the first set and 88.8 ± 60.1 in the second set. The mean number of the uropodid mite, *Leiodinychus krameri,* was 357.8 ± 252.3 in the first set and 434.3 ± 134.7 in the second. The mean number of histerid beetle, *Carcinops pumulio,* was 22.7 ± 15.6 in the first set and 16.4 ± 13.1 in the second.

Experiment 3. With the use of the feed additive at the treated farm, the numbers of adult flies trapped decreased rapidly from a high of 283 per trap per day to 16 or less during the remainder of the fly season after 35 days of using the additive (Table 3). Concurrently, fly larvae recovered from the manure samples declined to extremely low levels (10 or less per 2 liter manure sample) for the remainder of the fly season after 47 days of using the additive and were significantly ($P \leq .05$) less than in the untreated comparison farm where numbers of fly larvae remained high throughout the season.

The predacious mites and beetles were abundant in the manure samples throughout the period of the experiment in both the treated and untreated houses. At most sampling times, the number of mites was actually significantly greater in the manure in the treated houses after 35 days of using the feed additive than in the untreated houses. The beetles were significantly more abundant in the manure in the treated houses at 3 sampling times after 47 days of using the feed additive. The fewer mites and beetles in the untreated manure than in the treated was apparently due to the physical state of the manure, i.e., the treated manure was drier and less flattened than the untreated and therefore more suitable for the survival and reproduction of mites and beetles.

Experiment 4. There were few or no house fly larvae in the manure from birds given the feed additive after 56 days of feeding while the

TABLE 4. *Effects of using the poultry feed additive Larvadex® on the abundance of manure-inhabiting house fly larvae and beneficial mites (Macrocheles muscaedomesticae and Fuscuropoda vegetans) and beetles (Carcinops pumulio) in a replicated experiment in a narrow caged layer house. Feed additive used June 19 to October 1, 1981 (Experiment 4)*

Organism[1] treatment	No. days fed							
	0	14	28	42	56	77	91	105
Fly larvae								
Treated	33.1	81.2	24.2	131.3	4.8*	0*	0.2*	2.7*
Untreated	74.6	90.2	25.3	99.2	236.8	225.1	145.1	298.3
M. muscaedomesticae								
Treated	61.5	24.3	181.1	206.0	315.8	567.0*	517.2*	719.0*
Untreated	65.1	22.8	176.7	229.2	146.2	58.6	93.7	89.6
F. vegetans								
Treated	92.0	34.1	189.2	1068.7	1041.2	1893.5*	1888.7*	1644.3*
Untreated	80.1	35.7	209.7	938.0	1050.5	402.8	301.2	243.0
C. pumulio								
Treated	29.3	20.7	14.2	22.5	58.9	79.4*	31.6*	25.1*
Untreated	23.2	25.2	12.7	25.3	40.3	10.7	7.6	2.9

[1] Mean number recovered from 2 liters of manure.

*Means for the treated and untreated for that organism on the given sampling day are significantly different ($P \leq .05$).

numbers of fly larvae continued to be high in the manure from untreated birds (Table 4). Mites and beetles in the manure increased during the experiment. There were no significant differences in the numbers of mites and beetles in the manure through the sampling on day 56 of using the feed additive. In the sampling on Day 77 and after, there were significantly higher numbers of mites and beetles in the manure under the birds given the feed additive than in the manure under the untreated birds. As in Experiment 3, the drier manure under the treated birds provided a more suitable habitat for the mites and beetles.

Experiment 5. As shown in Table 5, the adult house fly population in the four caged layer houses reached a nuisance level by Weeks 6 and 7 (last half of May). The use of the feed additive during alternate Weeks 8, 10, 12, and 14 resulted in a gradual reduction in the numbers of adult flies after an initial delay. By Weeks 12 to 14 the fly numbers based on trap counts (32 to 38 flies per trap per day) and spot counts (3 to 5 spots per card per day) was moderately low but not low enough to be an acceptable level of control. With the use of the feed additive on an alternate schedule of 4 days with and 4 days without (weeks 15 to 26) the fly population was reduced to a satisfactory level of control (5 to 18 flies per trap per day, 1 to 4 spots per card per day). After Week 26, although no feed additive was used, the fly population remained low, largely due to the advent of cool weather.

TABLE 5. *Seasonal abundance of adult house flies and effects of alternate use of the poultry feed additive Larvadex® on the abundance of flies in caged layer houses. April 6 to November 8, 1982. (Experiment 5)*

Treatment and date		Week no.	Mean no. flies per trap per day[1]	Mean no. spots per card per day[1]
No feed additive				
Apr	12	1	5.0	0
	26	2	4.7	.8
May	3	3	6.5	1.1
	10	4	14.9	3.8
	17	5	13.1	4.4
	24	6	34.6	4.3
	31	7	73.8	6.8
Feed additive alternately 7 days on, 7 days off				
June	7	8	140.1	18.3
	14	9	190.2	15.5
	21	10	97.3	10.1
	28	11	59.1	4.6
July	6	12	37.0	4.1
	12	13	32.7	3.3
	19	14	38.7	5.1
Feed additive alternately 4 days on, 4 days off				
	26	15	15.9	3.5
Aug	2	16	15.9	4.7
	9	17	17.1	3.5
	16	18	12.9	3.3
	23	19	18.7	4.4
	30	20	10.6	3.7
Sept	6	21	5.2	2.9
	13	22	13.4	3.8
	20	23	7.0	3.3
	27	24	7.0	1.4
Oct	4	25	8.9	3.2
	11	26	12.3	4.0
No feed additive				
	18	27	7.8	1.6
	25	28	6.8	.8
Nov	1	29	4.8	.8
	8	30	2.3	.5

[1] n = 8.

DISCUSSION

The data from these experiments under field conditions and using large numbers of caged laying hens showed that Larvadex effectively controlled house flies while not adversely affecting the manure-inhabiting predacious mites and beetles. Experiment 1 involved starting the use of the feed additive in the middle of the summer in a high-rise house after a large fly population had developed. Normally the additive would be used earlier in the fly season. Nevertheless, reasonable fly control was achieved after 17 days of feeding the additive. In the narrow houses (Experiment 2) the additive was used after the manure was removed in the middle of the summer and satisfactory fly control was achieved for the balance of the season. These two experiments demonstrated that practical fly control can be achieved even when use of the additive is initiated in the middle of the season of fly activity.

Experiments 3 and 4 demonstrated that the use of the feed additive continuously during most of the fly season had no adverse effects on the populations of predacious mites and beetles in the manure. In addition, the increased drying of the manure with the use of the additive tended to promote higher numbers of mites and beetles as compared to the wetter manure under birds not given the feed additive. In both experiments, fly populations were high at the beginning of the experiments and some time was required to produce significant reductions in the numbers of fly larvae in the manure as a result of using the feed additive (47 days in Experiment 3 and 56 days in Experiment 4). However, in Experiment 3 the numbers of adult flies were reduced to below the nuisance level after 35 days of using the feed additive continuously.

Experiment 5 was an attempt to reduce the cost of using the feed additive by not using it continuously. With the high population of flies at the beginning of the feeding the degree of fly control was not satisfactory when the additive was used alternately for 7 days and omitted for 7 days. An obvious decline in the numbers of adult flies occurred, however. With alternate feeding on a 4-day schedule (with additive for 4 days, without for 4 days), fly control was excellent. Thus, it is possible to achieve a high level of fly control using the additive 50% of the time when the interval without use of the additive is 4 days but not when the interval is 7 days.

These data were obtained under field conditions and are directly applicable to the conditions encountered by poultry producers using similar types of caged layer housing. From these experiments and others, the baited jug traps have been an effective means to monitor the fly population. A mean trap count of less than 20 flies per day represents excellent fly control with few adult flies observed in the houses. For the most effective and economical fly control, it would be advisable to monitor the fly population and initiate use of the feed additive when the fly population reaches 20 or more per trap per day. If conditions or seasonal effects naturally cause the fly numbers to be below that level there would be no need to use the feed additive.

The lack of adverse effects of the feed additive on the manure-inhabiting predacious mites and beetles is an important attribute of the chemical. The macrochelid and uropidid mites as well as the histerid beetles feed on fly eggs and first-instar fly larvae. Therefore, the mites and beetles contribute substantially to reducing fly numbers. Predation by these mites and beetles as well as their reproduction is enhanced by dry manure while the use of the feed additive encourages manure drying. Thus, the feed additive and the predacious mites and beetles are not only compatible but complement each other in reducing fly numbers.

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