B000 Cyfluthrin

OPP OFFICIAL RECORD
HEALTH EFFECTS DIVISION
SCIENTIFIC DATA REVIEWS
EPA SERIES 361

PC 128831 3

Special Subacute Inhalation Study

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Secondary Reviewer Toxicologist: John Whalan John Whalan, Date 2-13-01
Registration Action Branch 2 (7509C)

DATA EVALUATION RECORD

STUDY TYPE: 7-day Inhalation Toxicity Study with Spontaneous Motor Activity Measurements After a 14 Week Recovery Period in Mouse Dams and Pups

DP BARCODE:
P.C. CODE:128831

SUBMISSION CODE:
TOX. CHEM. NO.: 266E

TEST MATERIAL (PURITY): Cyfluthrin technical (96.8% pure)

SYNONYMS: FCR 1272, Baythroid, Tempo

CITATION: Jekat, F.W., J. Pauluhn, A. Popp and G. Schmuck (1997) Motor activity measurements in male and female mice postnatally exposed to FCR 1272 (cyfluthrin) by inhalation. Bayer AG Department of Toxicology, Germany. Report No. 26484, Study No. T7060263, July 23, 1997. MRID 44373401. Unpublished.

SPONSOR: Bayer AG, Friedrich-Ebert-Strasse 217-333, D-42096 Wuppertal, Germany

EXECUTIVE SUMMARY:

In a 7-day inhalation toxicity study (MRID 44373401) technical cyfluthrin (96.8%) was administered to groups of SPF-bred NMRI mice, 5 dams/concentration with 8 pups each (4 males and 4 females, 10 days old). The dams were exposed together with their offspring in a dynamic whole-body chamber at concentrations of 0, 0.006, 0.015 or 0.058 mg/L for 6 hours per day for 7 consecutive days. The MMAD ± GSD was 1.6 - 1.8 μm ± 1.8. During the recovery period, the dams were maintained and housed with their pups until weaning, at which time the offspring were housed with their same sex litter mates (4/cage) for 14 weeks. At week 15, the offspring were tested for spontaneous motor activity and hematological and selected clinical chemistry parameters were examined. Brains from the offspring were either processed for microscopic examination or prepared for determination of muscarinic receptors in different brain regions.

At 0.006 mg/L, no effects were observed in either the pups or the dams when compared to the control group. At 0.015 mg/L , clinical signs of toxicity were observed in both sexes of pups during the period right after exposure (exposure

days 0-6; decreased motility, poor general condition, tonic seizures and temporary scratching (direct sensory irritation)). No clinical signs were observed in the dams. Adult female offspring showed higher scores for horizontal and vertical activity, total distance and movement time when compared to the control group. At 0.058 mg/L, all pups except one died during the first exposure period. The one surviving pup was killed in extremis. No deaths or clinical signs were observed in any of the dams. No biologically significant effects were observed in body weights, hematology, clinical chemistry, gross or microscopic pathology. In addition, cyfluthrin had no effect on the muscarinic acetylcholine receptor in the cortex of adult mice in either sex.

The LOAEL for pups is 0.015 mg/L based on clinical signs of toxicity and increased spontaneous motor activity in females 4 months after exposure. The NOAEL for pups is 0.006 mg/L .

The parental LOAEL (dams) is greater than 0.058 mg/L (HDT). No effects were observed at any dose level. The parental NOAEL is 0.058 mg/L (HDT).

This 7-day inhalation study is acceptable nonguideline.

COMPLIANCE: Signed and dated GLP, Quality Assurance, Data Confidentiality, and Flagging statements were provided. The histopathological investigations were not performed according to the GLP standards and the computer software used for the motor activity measurements was tested with positive control compounds but not formally validated according to GLP requirements.

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test Material: FCR 1272 (Cyfluthrin) technical grade
   Description: brown, viscous oil partially containing crystals
   Lot/Batch #: 380368010
   Purity: 96.8% a.i.
   Stability of compound: store at room temperature

2. Vehicle and/or positive control: poly ethylene glycol

3. Test animals: Species: mouse
   Strain: SPF-bred NMRI
   Source: Harlan Winkelmann GmbH, Borchen Germany
   Housing: Dams housed individually with pups in type III Makrolon cages until weaning. After weaning, pups housed with litter mates by sex in

same type cages.
Diet: Altromin 1324 ad libitum
Water: tap, ad libitum
Environmental conditions:
Temperature: 22 ± 2° C
Humidity: 40-70%
Photoperiod: 12 hour dark/light cycle
Acclimation period: 6 days

B. STUDY DESIGN:

1. In life dates - start: 11/20/95 end: 3/8/96

TABLE 1: STUDY DESIGN[a]

Test group	Nominal Conc. (mg/L)	Analytical Conc. (mg/L)	MMAD μm	GSD
Control	0.0	0.0	1.8	1.8
Low (LCT)	0.010	0.006	1.7	1.8
Mid (MCT)	0.026	0.015	1.7	1.8
High (HCT)	0.106	0.058	1.6	1.8

2. Generation of the test atmosphere and description of the chamber:

The solid test substance was heated at approximately 50°C for 2 hours and subsequently dissolved in the vehicle polyethylene glycol 400. Atmospheres were generated using a modified six-nozzle BGI collision nebulizer type MRE. Following nebulization using 30 L air/min the atmosphere was fed into the second preseparator/baffle system to prevent larger particles from entering the chamber. While entering the top of the whole-body inhalation chamber the test atmosphere was diluted with approximately 417 L air/min (25 m^3/h). In all inhalation chambers approximately the same concentration of vehicle was present. All air flows were monitored and adjusted continuously by means of calibrated and computer controlled flow-controllers and an electronic control and data acquisition system provided for constant and reproducible exposure conditions. The air exchange rate was approximately 11 times/hour.

The animals were exposed in wire-mesh cages. Samples for analysis of the particle-size distribution were taken in the vicinity of the animals. The nominal concentrations were calculated each time the generator content was exchanged from the cumulative total air flow through the chamber during the period of exposure. In the test atmosphere, the pyrethroid active ingredient FCR 1272 was determined by HPLC.

C. METHODS:

1. Clinical Signs of Toxicity and Mortality

Experimental animals were inspected for signs of toxicity and mortality twice daily (during the exposure period, before and after the inhalation period) and once daily on weekends and holidays. Detailed examinations occurred once/week.

2. Body Weights

Body weights of pups were measured on the first, third and fifth days of exposure and then weekly, starting at day 8 up to week 15.

3. Automated Measurement of Spontaneous Motor Activity

At week 15, 20 offspring/dose/sex were tested for spontaneous motor activity. They were tested in activity monitors [Omnitech digiscan analyzer] for 60 minutes in sample intervals of 10 minutes. Each analyzer is equipped with 16x16 infra-red light beams for the recording of the horizontal activity and additional 16 light beams 8 cm above the cage floor to record the vertical activity. The following parameters were automatically calculated: horizontal activity, number of movements, number of stereotypy (breaking same light beam(s) repeatedly), total distance, vertical activity, vertical time, movement time, average distance/move, average speed, rest time, number of vertical/rearing movements, stereotypy time, clockwise revolutions, anticlockwise revolutions, left front, center front, right front, left center, right center, left rear, center rear and right rear.

4. Hematology:

Prior to necropsy, blood was collected in 8 randomly selected offspring/group/sex and hematological and selected clinical chemistry parameters were examined. The following CHECKED (X) parameters were examined.

X		X	
x	Hematocrit (HCT)*	x	Leukocyte differential count*
x	Hemoglobin (HGB)*	x	Mean corpuscular HGB (MCH)
x	Leukocyte count (WBC)*	x	Mean corpusc. HGB conc.(MCHC)
x	Erythrocyte count (RBC)*	x	Mean corpusc. volume (MCV)
x	Platelet count*	x	Reticulocyte count
x	(Thromboplastin time)		

5. Clinical Chemistry: The following CHECKED (X) parameters were examined.

x	Albumin*
x	Blood creatinine*
x	Blood urea nitrogen*
x	Total Cholesterol
x	Triglycerides

6. Sacrifice and Pathology

The brains from 5 offspring/sex/group were processed for microscopic examination. Microscopic examination was carried out in controls and the groups exposed to 0.006 and 0.015 mg/L, respectively. Six coronal sections through the brain - olphactory region, forebrain, midbrain, cerebellum with rostral pons, cerebellum with caudal pons, medulla oblongata were examined microscopically.

7. Muscarinic Acetylcholine Receptor in the Cortex

The brains for 12 offspring/sex/group were prepared for determination of muscarinic receptors in different brain regions. Receptor binding studies were performed to characterize and quantify the muscarinic acetylcholine receptor (mAChR) density in adult (4 month) mice of both sexes treated with cyfluthrin (0.006 and 0.015 mg/L) during the brain growth spurt (days 10-17). In parallel to these investigations, in vivo motor activity behavior tests were evaluated as well. The mAChR density was quantified by [^{3}H] Quinucludinyl bencylate (QNB). To differentiate between the specific and unspecific binding, a high concentration of Atropine was given to displace QNB from the specific binding site. A concentration dependent displacement curve of the receptor was made with an unspecific ligand of the cholinergic receptor Carbachol.

II. RESULTS:

A. Observations

1. Toxicity - During the period right after exposure (during days 0-6), no clinical signs of toxicity were observed in either the control group or in the group exposed to 0.006 mg/L. Pups of both sexes exposed to 0.015 mg/L were observed to have decreased motility, poor general condition, tonic seizures and temporary scratching (direct sensory irritation). No clinical signs of toxicity were reported for any of the dams.

Clinical Signs of Toxicity in Pups (# animals affected)*			
Dose (mg/L) Clinical Sign	0	0.006	0.015
Males			
Temporary scratching	0/20	0/20	12/20
Tonic seizures	0/20	0/20	4/20
Decreased motility	0/20	0/20	4/20
Wet fur	0/20	0/20	4/20
Poor general condition	0/20	0/20	0/20
Females			
Temporary scratching	0/20	0/20	8/20
Tonic seizures	0/20	0/20	4/20
Decreased motility	0/20	0/20	4/20
Wet fur	0/20	0/20	0/20
Poor general condition	0/20	0/20	0/20
Head tilted	0/20	0/20	1/20

*At 0.058 mg/L all the pups died. No clinical signs were observed.

2. Mortality - At 0.058 mg/L, all pups died during the first exposure period except one, which was killed in a moribund state. No deaths were observed in any of the dams or in any of the pups in the other dose groups.

B. Body weights

At 0.015 mg/L, mean body weights when compared to controls during exposure was decreased in both sexes of pups (day 1 of exposure (10 days old: 94%), day 3 (92-93%) and day 5 (87-90%)). There was an even slighter decrease in female pups at 0.006 mg/L. Due to the small numerical decrease in body weights, the decreases are not considered to be biologically relevant.

Mean Body Weights of Pups From Age 10 to 14 Days (g)				
Dose mg/L	Sex	10 days	12 days	14 days
0	Male	7.0	7.5	7.9
0	Female	7.0	7.5	7.9
0.006	Male	6.8	7.3	7.6
0.006	Female	6.8	7.3	7.5
0.015	Male	6.6	6.9	6.9
0.015	Female	6.6	7.0	7.1

C. Spontaneous Motor Activity

At 0.015 mg/L, females showed higher scores for horizontal ($p < 0.05$) and vertical activity ($p < 0.05$), total distance ($p < 0.05$) and movement time ($p < 0.05$) when compared to the control group. No other groups showed any treatment-related changes. Supporting figures and tables taken directly from the report are attached.

D. Clinical Pathology

1. Hematology - No treatment-related effects were observed.

2. Clinical chemistry - No treatment-related effects were observed.

E. Sacrifice and pathology

1. Gross pathology - No gross findings were observed during necropsy.

2. Microscopic pathology - No treatment-related microscopic findings were observed. Focal gliosis was found in the cerebrum of 1 control female. No other microscopic findings were observed in any other animals.

F. Muscarinic Acetylcholine Receptor in the Cortex - Cyfluthrin had no effect on mAChR in the cortex of adult mice in both sexes. In addition, mAChR receptor density was not changed in the investigated brain region.

III. DISCUSSION

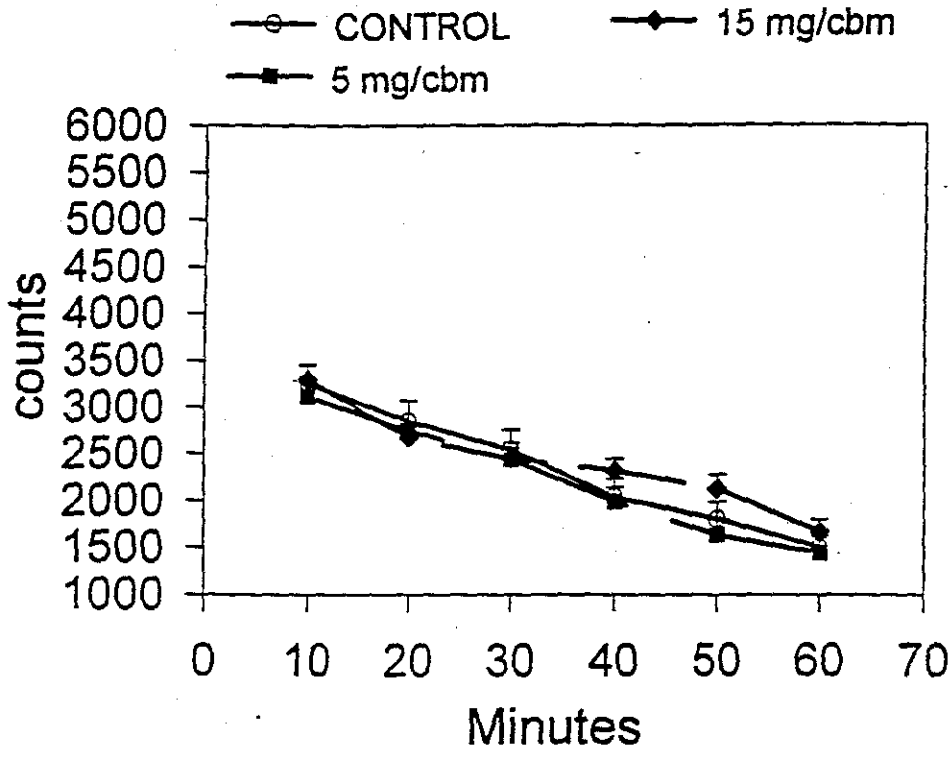
The report states that, "the low sensitivity of mammals against pyrethroids is mostly related to their higher metabolic capacity to hydrolyze these compounds in contrast to insects. Direct injections in the CNS resulted in effects comparable to those of the insect nervous system. An additional resistance of homeothermic organisms against pyrethroids is attributed to the negative temperature coefficient of the channels."

Although the report did not discuss the possible reasons for the differences in sensitivity between the pups and the dams, based on the statements above, it is possible that the pups have not yet developed the metabolic capability to hydrolyze the compounds as well as the adults.

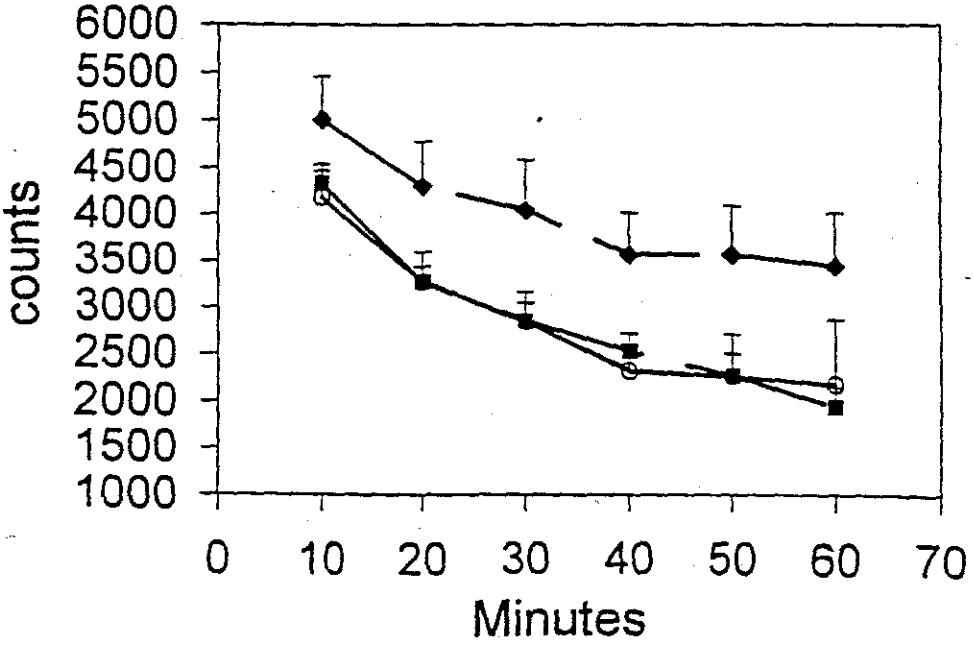
8 FIGURES

T7060263 4 Months Figure 2

HORIZONTAL ACTIVITY MALE

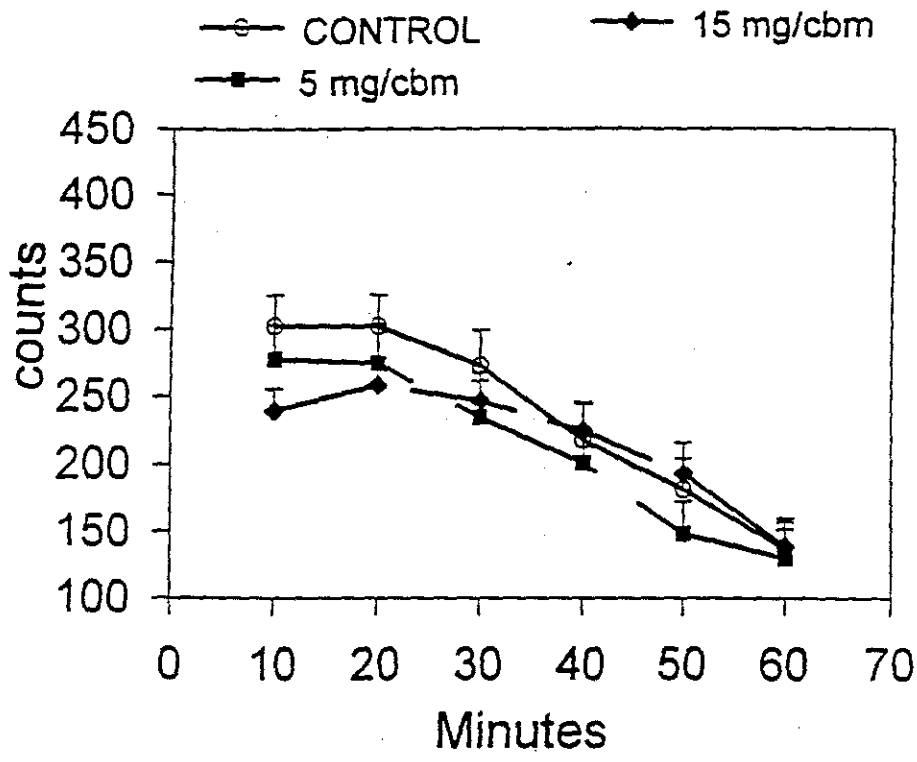


HORIZONTAL ACTIVITY FEMALE

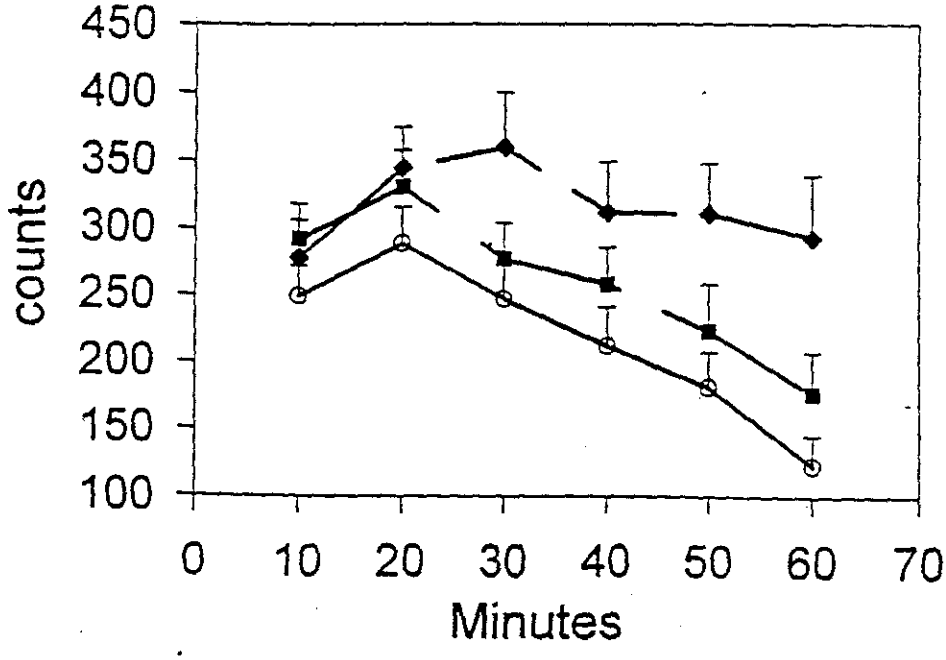


T7060263 4 Months Figure 3

VERTICAL ACTIVITY MALE



VERTICAL ACTIVITY FEMALE



T7060263 FCR 1272 (Cyfluthrin) 4 Months MA

	HA Male		MT Male		NM Male	
Control	Mean	SE	Mean	SE	Mean	SE
Sample 1	3260	192	196	9	149	2
Sample 2	2853	221	158	11	144	5
Sample 3	2541	223	145	11	135	5
Sample 4	2037	191	115	10	115	6
Sample 5	1801	176	98	9	109	8
Sample 6	1503	179	79	10	88	9
5 mg/cbm						
Sample 1	3117	216	169	11	151	4
Sample 2	2756	230	142	13	139	6
Sample 3	2434	183	128	13	128	6
Sample 4	1987	151	106	11	116	7
Sample 5	1631	168	84	12	90	9
Sample 6	1445	169	78	12	84	10
15 mg/cbm						
Sample 1	3299	164	187	9	146	3
Sample 2	2677	121	142	9	135	6
Sample 3	2445	121	128	9	129	7
Sample 4	2316	129	122	8	131	6
Sample 5	2123	147	108	10	118	10
Sample 6	1668	129	83	8	98	8

HA = Horizontal Activity
MT = Movement Time
NM = Number of Movements

T7060263 FCR 1272 (Cyfluthrin) 4 Months MA

	NS Male		ST Male		TD Male	
Control	Mean	SE	Mean	SE	Mean	SE
Sample 1	99	3	145	12	1781	97
Sample 2	93	3	126	13	1471	117
Sample 3	88	4	114	13	1327	100
Sample 4	80	4	93	10	1086	100
Sample 5	74	5	79	10	923	85
Sample 6	66	5	77	9	719	97
5 mg/cbm						
Sample 1	102	2	155	13	1539	121
Sample 2	96	3	148	14	1229	126
Sample 3	93	3	125	9	1135	131
Sample 4	85	3	106	7	948	109
Sample 5	75	5	92	7	751	112
Sample 6	66	5	81	9	691	116
15 mg/cbm						
Sample 1	103	2	150	11	1677	117
Sample 2	96	2	130	8	1246	105
Sample 3	91	3	120	7	1163	101
Sample 4	88	4	103	8	1134	95
Sample 5	79	4	106	9	988	93
Sample 6	71	4	80	7	752	72

NS = Number of Stereotypy
ST = Stereotypy Time
TD = Total Distance

T7060263 FCR 1272 (Cyfluthrin) 4 Months MA

	VA Male		VT Male	
Control	Mean	SE	Mean	SE
Sample 1	303	23	147	8
Sample 2	303	23	159	10
Sample 3	273	27	143	10
Sample 4	218	28	121	11
Sample 5	181	23	104	12
Sample 6	137	23	82	12
5 mg/cbm				
Sample 1	278	26	139	10
Sample 2	275	30	133	10
Sample 3	235	27	121	11
Sample 4	201	25	108	11
Sample 5	148	24	82	12
Sample 6	130	22	73	11
15 mg/cbm				
Sample 1	240	16	132	7
Sample 2	259	20	142	10
Sample 3	247	21	147	11
Sample 4	225	20	146	13
Sample 5	193	23	123	14
Sample 6	139	18	100	12

VA = Vertical Activity
VT = Vertical Time

T7060263 FCR 1272 (Cyfluthrin) 4 Months MA

	HA Female		MT Female		NM Female	
Control	Mean	SE	Mean	SE	Mean	SE
Sample 1	4182	287	225	13	148	3
Sample 2	3274	327	188	15	135	4
Sample 3	2850	324	172	16	132	4
Sample 4	2327	273	139	15	119	5
Sample 5	2263	448	129	19	106	7
Sample 6	2185	692	113	27	83	9
5 mg/cbm						
Sample 1	4338	199	224	13	151	3
Sample 2	3275	167	175	12	145	4
Sample 3	2860	191	155	13	133	6
Sample 4	2533	191	135	12	125	7
Sample 5	2273	232	126	15	110	9
Sample 6	1952	206	109	14	101	9
15 mg/cbm						
Sample 1	5012	452	262	20	141	7
Sample 2	4305	477	242	24	132	6
Sample 3	4052	533	238	28	121	7
Sample 4	3572	452	216	27	120	6
Sample 5	3570	528	209	31	112	7
Sample 6	3453	569	203	33	105	7

HA = Horizontal Activity
MT = Movement Time
NM = Number of Movements

T7060263 FCR 1272 (Cyfluthrin) 4 Months MA

	NS Female		ST Female		TD Female	
Control	Mean	SE	Mean	SE	Mean	SE
Sample 1	103	2	153	10	2397	256
Sample 2	88	4	107	12	2079	325
Sample 3	80	4	87	9	1966	328
Sample 4	73	4	76	10	1592	363
Sample 5	67	3	71	8	1751	574
Sample 6	65	4	67	7	1883	944
5 mg/cbm						
Sample 1	105	2	176	11	2358	208
Sample 2	97	2	127	10	1916	186
Sample 3	86	3	106	9	1689	205
Sample 4	82	4	98	9	1473	169
Sample 5	74	6	84	9	1359	190
Sample 6	71	5	75	9	1166	197
15 mg/cbm						
Sample 1	100	2	176	17	3147	472
Sample 2	93	3	129	13	3307	587
Sample 3	86	4	105	11	3415	646
Sample 4	80	5	96	12	2903	495
Sample 5	79	4	87	11	3206	720
Sample 6	73	6	85	12	3150	754

NS = Number of Stereotypy
ST = Stereotypy Time
TD = Total Distance

T7060263 FCR 1272 (Cyfluthrin) 4 Months MA

	VA Female		VT Female	
Control	Mean	SE	Mean	SE
Sample 1	249	23	140	9
Sample 2	289	27	164	12
Sample 3	247	25	143	10
Sample 4	212	29	134	11
Sample 5	181	26	122	13
Sample 6	123	22	86	13
5 mg/cbm				
Sample 1	292	26	150	10
Sample 2	331	27	167	11
Sample 3	277	27	148	12
Sample 4	258	28	138	11
Sample 5	223	35	122	14
Sample 6	176	31	105	12
15 mg/cbm				
Sample 1	278	28	132	10
Sample 2	345	30	159	10
Sample 3	360	40	163	11
Sample 4	312	37	153	10
Sample 5	311	37	150	12
Sample 6	293	46	139	13

VA = Vertical Activity
VT = Vertical Time

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R058486

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Accession Number:	**412-04-0046**

HED Records Reference Center
03/25/2004