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#### FLOCOUMAFEN -- A NEW ANTICOAGULANT RODENTICIDE

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ABSTRACT: Flocoumafen is a new anticoagulant rodenticide with an acute toxicity between that of bromadiolone and brodifacoum. It has performed well in field tests against house mice and susceptible as well as resistant brown rat populations. Danish lab tests reveal a considerable variation in susceptibility between rodent species and indicate that practical problems in the control of certain Scandinavian bromadiolone-resistant house mouse populations may arise shortly after introduction.

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#### INTRODUCTION

The increasing problems in the control of rodent populations resistant to the anticoagulant rodenticides also have certain positive aspects. Commercial interests in synthesizing, screening and introducing new products on the market of rodent control have been greatly stimulated. First, the more potent hydroxycoumarins difenacoum and bromadiolone became important alternatives to the older anticoagulants in the mid-seventies. Then the completely different, but still slow-acting compounds, the calciferols, were introduced, especially in countries with practical resistance problems. Almost at the same time the highly efficient anticoagulant brodifacoum appeared, and most recently another slowacting rodenticide, bromethalin, as well as two new anticoagulants, difethialone and flocoumafen, have been developed. Flocoumafen (4-hydroxy-3-[1,2,3,4-tetrahydro-3-[4-(4-trifluormethylbenzyloxy)phenyl-l-napthylcoumarin) was synthesized in 1984 by Shell International Chemical Co. Ltd., and it was marketed in several countries under the trade name of STORM, formulated as loose grain bait, small pellets, and wax blocks for rats and mice - all at concentrations of 0.005% flocoumafen.

LABORATORY DATA ON TOXICITY AND EFFICACY The acute oral  $LD_{50}$  is for most species reported to be below 1 mg/kg, but it varies considerably from 0.12 mg/kg in Microtus arvalis to more than lOmg/kginAcomyscahirinus. a species remarkably tolerant to most anticoagulants (Table 1). The acute toxicity of flocoumafen to the three major commensal rodent species is compared with that of difenacoum, bromadiolone, and brodifacoum in Table 2.

Tests with warfarin-resistant strains of rats and mice indicate that flocoumafen will perform well in practice to most populations. The only species in which the resistance factor is significantly increased is the house mouse (Table 3).

### TOXICITY TO NON-TARGET ANIMALS

Flocoumafen is generally less toxic to non-target species than to rats as can be seen from the following figures, where the dog is an important exception from the rule (Johnson et al. 1986): Pig 70 mg/kg; Hen < 100 mg/kg; Quail 100 - > 300 mg/kg; Duck 24-94 mg/kg and Dog 0.075 - 0.25 mg/kg. As

far as the dog is concerned, brodifacoum has been reported to be similarly toxic (Dubock et al. 1980), whereas the acute  $LD_{50}$  of bromadiolone to dogs is above 10 mg/kg (Grand 1976)

Of the three STORM formulations the biconvex wax block is the principal bait formulation selected for the commercialization of flocoumafen. It seems to be attractive to small children, but the addition of a substance tasting highly bitter to humans and not to rodents is an important improvement compared with all other rodenticide formulations. Predators, including dogs, are, however, still at some risk of being poisoned by consuming these blocks, but they have been found unattractive to birds (Garforth et al. 1987). Like the other 'second generation' anticoagulants, flocoumafen has a long biological half-life. This first of all has the consequence that the vitamin Kj treatment of accidentally poisoned animals or humans must be considerably prolonged, e.g., for several weeks (Shell 1987b). Dead rodents collected during field trials have been found to contain 1 -3 mg flocoumafen per kg, on average 25 times less than the bait itself.

#### FIELD TRIALS WITH RATS

In the UK 12 farmsteads with documented rat infestations in the warfarin-resistance area were treated with medium oat meal containing 0.005% flocoumafen (Buckle 1986). On half of the premises the traditional, unrestricted baiting method was applied using 100-g depots at a high number of baiting points and maintaining a surplus throughout the treatment. On the other half a restricted treatment was carried out, starting with only 50 g of bait and replenishing only twice a week until bait take had ceased. Complete control was achieved in both types of treatment, but the amount of bait used was considerably larger in the traditional treatment, whereas the time used to complete control was one week shorter.

In another series of 12 similar UK field trials with flocoumafen wax blocks carried out by the Ministry of Agriculture, Fisheries and Food an average kill of 99.2% was achieved on half of the premises where unrestricted baiting was carried out compared with a 96% reduction in rat numbers, where the baiting was restricted to two instead of

Table 1. The acute oral toxicity of technical flocoumafen to various rodent species and the New Zealand white rabbit. (Shell 1987a).

National Control of the Control of t	Acute oral	Days to	
Species (sex) & strain	LD <sub>50</sub> (mg/kg)	Range	Mean
RAT			
Bandicota indica (M&F)	0.48	122	10
Pracomys natalensis (M&F)	1.3	3-7	5
Rattus argentiventer (M)	0.48	4-18	9
R. argentiventer (M)	0.56	4-19	7
R. norvegious (M&F) F344	0.25	5.7	6
R. norvegicus (M) Wistar	0.46	5-8	6
R. norvegicus (F) Wistar	0.56	5-8	6
R. rattus (M)	1.8	6-11	9
R. rattus (F)	1.0	4-13	8
R. r. diardii (M&F)	0.65	3-20	7
R. tiomanicus (M)	0.28	3-17	5
R. tiomanicus (F)	0.42	3-10	7
Sigmodon hispidus (M)	1.21	6-14	8
MOUSE			
Mus musculus (M) C57BL/10	0.79	4-11	7
M. musculus (F) C57BL/10	1.5	6-10	7
M. musculus (M&F) CF1	2.4	2-10	6
Apodemys flavicollis (M&F)	4.2	4-6	5
Acomys cahirinus (M&F)	>10	÷+	
GERBIL			
Meriones unguiculatus (M)			
Mongolian	0.18	4-7	5
GUINEA PIG			
Cavis porcellus (M)			
Dunkin Hartley	>10	4	4
HAMSTER			
Mesocricetus auratus	>50	9	9
VOLE			
Arvicola terrestris (M&F)	0.2	3-18	7
Clethrionomys glareolus			
(M&F)	0.23	2-6	4
Microtus arvalis (M&F)	0.12	2-8	4
RABBIT			
Oryctolagus cuniculus	0.7	8-14	10

three blocks per baiting point and where the baiting period was reduced from 29 to 21 days (Johnson 1988). In South East Asia oil palm trees are reported to have been protected again st damage from Rattus tiomanicus by baiting with flocoumafen wax blocks, one per tree at 4-day intervals (Shell 1987a). Damage from Rattus argentiventer to tillers in ricefields in Thailand was reduced from 5.05 % to 0.65 % by using 0.005 % flocoumafen rice bait (Shell 1987a).

Table 2. The acute  $LD_{50}$  of four 'new' anticoagulant rodenticides to susceptible strains of the three major commensal rodent species (mg/kg).

ev-=:	Flocoumaten	Difonaccom	Bromadiologe B	eedifacoun
Rattus norvegicus	0.25-0.56	1.8-3.5*	0.65-1.125**	0.22***
Rattos rattus	1.00-1.78	2.5-7.0*	1370	0.65***
Mus domesticus	1.13-2.40	0.80*	0.99-1.750**	0.40***
* Bull 1976 ** Grand 197 *** Dubock :	6; Mechan 197 t al. 1978	8		

Table 3. Acute oral toxicity of technical flocoumaten to resistant strains of rats and mice. (Shell 1987a)

	Acute oral Non-	I.D <sub>50</sub> (mg/kg)	Resistance factor
Species (sex)	resistant	Resistant	(RF)
RAT			
R. norvegicus (M)	0.46	0.46	1.0
R. norvegicus (F)	0.56	0.42	0.8
R. ratuis (M)	1.8	ca. 1.9	ca. 1.1
R. rattus (F)	1.0	ca. 1.4	ca. 1.4
R. tiomatticus (M)	0.28	0.28	1.0
R. tiomanicus (F)	0.42	0.65	1.5
MOUSE			
M. musculus (M&I	F) 1.3	5.3	4.1

#### FIELD AND PEN TRIALS WITH HOUSE MICE

Ten field trials carried out by the Ministry of Agriculture, Fisheries and Food on mouse-infested farms in Sussex, UK, using 0.005 % flocoumafen baits based on oat meal, resulted in complete eradication in seven cases and a reduction in numbers on the other farms ranging from 87.1% to 95.5%. On eight farms the results were achieved in 3-4 weeks and in two cases the treatment period was prolonged to 5 and 6 weeks, respectively. No resistance was detected in survivors trapped and tested in the laboratory (Rowe et al. 1985). Compared with previous field trials conducted in a similar way with difenacoum, bromadiolone and brodifacoum, flocoumafen turned out to be just as efficient, or even better, particularly as far as the length of the treatment period is concerned.

Four pen trials with family groups of house mice from a warfarin-resistant strain resulted in a complete kill when baiting with 0.005 % flocoumafen in pinhead oatmeal together with plain food. Only 5 mice out of 68 survived for more than seven days. The results are comparable with, or

slightly better than those obtained with other anticoagulants in similar tests (Rowe et al. 1985). Four similar trials with flocoumafen wax blocks also resulted in a 100% mortality in the same strain of house mice (Johnson 1988), but the bait had to be presented for a longer period, e.g., 16 instead of 10 days.

#### LABORATORY TESTS CARRIED OUT WITH FLO-COUMAFEN IN DENMARK

Very recently (January, 1988) flocoumafen has been registered for rat control by the Danish Ministry of the Environment and after efficacy testing approved by the Danish Pest Infestation Laboratory. For the present wax blocks have only passed the registration process, but the pellet formulation has also been approved and may soon appear on the market.

Apart from  $LD_{50}$  tests incorporated into Table 1, feeding tests with singly caged as well as groups of rodents have been carried out since 1984. In tests with singly caged rodents the bait was offered as the only food for a varying number of day s; in the group tests the rodents were offered a nonpoisonous alternative.

The effect on singly caged brown rats of baits containing from 5 ppm to 80 ppm flocoumafen and with feeding periods varying from 24 hours to 6 days is summarized in Table 4. In most cases survival was due to individual rats refusing to

feed. A complete kill of bromadiolone-resistant rats was achieved after a 6-day feeding period using a 40-ppm or 50-ppm flocoumafen bait. The highest dose survived by a rat from the resistance area was 1.1 mg/kg.

House mice in Western Europe have recently been found to belong to two different species, <u>Mus domesticus</u> and M-musculus. Whereas the former species is the only one found in the UK, both species are abundant in Denmark. No difference has previously been detected between the two species as far as susceptibility to warfarin is concerned, but in Table 5 on feeding tests with Danish house mice a tendency can be seen of a higher tolerance to flocoumafen in M. <u>musculus</u> than in M-domesticus. This may explain some of the differences between the figures given for house mice in Table 1 and those appearing from the Danish tests.

The highest dose survived by M-<u>musculus</u> in the Danish tests is 13.5 mg/kg, and that by M-<u>domesticus</u> 5.6 mg/kg. The higher mortality obtained with wax blocks as well as with a cereal bait formulated by Sorex, compared with a broken barley bait, may also indicate that mice are capable of selecting less toxic particles from certain bait types.

Results from tests on house mice from a warfarin and bromadiolone-resistant strain are summarized in Table 6. A complete kill could not be achieved in a 24-hour test, even if the concentration was raised to 320 ppm. Survivors con-

Table 4. The effect of various concentrations and formulations of flocoumafen on singly caged brown rats (Rattus norvegicus')

Test no.	Rat strain	Concen- tration (ppm)	Test period (hr)	Bait type	Mortality	Consumption of non-survivors	a.i. (mg/kg) survivors
1.	Susceptible	50	24	Pellets	4/5	1.80(0.80-2.45)	0
2.	ausceptione	50	48	r eners	4/5	4,30(2.68-5.32)	0
3.	563	50	72	385	5/5	6.33(0.39-9.73)	v.
4	40	50	96	či.	4/5	6.16(3.99-9.33)	0
4. 5.	96	50	24	Wax blocks	10/10	1.73(0.31-4.26)	**
6.	a	50	48	Max Diocks	9/10	5.35(2,07-10.44)	0.25
7.	Resistant (not tested)	5	24	Broken barley	0/5	J.JJ(2,07-10,44)	0.27(0.06-0.49)
8.		10	24	M.	0/5	0.770	0.32(0.07-0.82)
9.	61	20	24	M.	2/5	1.58(0.88-2.27)	0.04(0-0.09)
10.	360	40	24	60	3/5(4)	0.71(0.43-1.08)	0.65
11.	Survivors from 7,8,9,10	20	24	80:	3/5	1.37(1.22-1.65)	0.84(0.58-1.10)
12.	44	40	24	44	5/5	1.89(1.46-2.73)	92
13.		80	24	u	5/5	4.74(3.88-7.65)	39
14.	Resistant to bromadiolone (tested)	40	144	ù	6/6	15,41(7,18-29,94)	ä
15.	Resistant area (not treated)	50	144	Pellets	5/5	8,85(6.60-12.51)	*
	Resistant to brumadiolone (tested)	50	144	Wax blocks	10/10	12,75(10.30-15.12)	2

Table 5.The effect of various concentrations of flocoumafen on singly caged house mice (Mus musculus (m) and Mdomesticus (d)) from warfarin-resistant strains. Varying test periods. No choice. Sorex formulation: cut wheat.

Test	Species	pecies Concen-	Test	Bait type	Mortality	Consumption of	a.i. (mg/kg)
no.	10	(ppm)	period (h)			non-survivors	survivors
ι.	m	5	24	Broken barley	0/5	100	0.73(0.50-0.83)
2.	m	10	24		0/5	· ·	1.52(1.22-2.20)
3.	m	20	24	14	0/4(5)	2550	2.58(1.52-3.69)
4.	m	20	24	Sorex form,	6/20	2.49(2.17-3.17)	2.64(1.44-3.63)
5.	d	20	24	M	5/6	3.19(2.53-3.92)	1.69
6.	m	40	24	Broken bariey	8/20	6.03(2.12-9.23)	4.74(2.00-7.24)
7.	m	40	24	Sorex form.	10/15	5.86(3.28-8.95)	4.40(1.69-5.00)
8.	m	40	24	Flour	8/10	7.26(5.13-10.86)	8.98(8.57-9.39)
9.	d	40	24	Sorex form.	3/4	4.94(4.18-5.38)	5.60
10.	m	50	24	Wax blocks	5/5	17.44(15.31-20.31)	
11.	m.	50	48		5/5	32.88(26.09-38.57)	18
12,	m	50	72	a	5/5	48.07(37.31-55.67)	**
13.	m	50	96	*	5/5	40.76(31.79-53.75)	
14.	m	80	24	Broken barley	8/10	10.50(8.00-12.71)	11.20(8.89-13.50)
15.	m	160	24		5/5	24.23(20.00-20.72)	

Table 6. The effect of various concentrations of flocoumafen on singly caged house mice (Mus musculus) from a warfarin-resistant strain. Feeding period 24 hours. No choice. Sorex formulation: cut wheat.

Test no.	Concen-	Bait type	Mortality	Consumption	of a.i. (mg/kg)
	tration (ppm)			non-survivors	survivors
1.	5	Broken barley	0/5	39	0.67(0.39-1.05)
2.	10	44	0/5	int.	1.66(1.15-2.23)
3.	20	5145	0/5		2.79(1.80-3.85)
4.	20	Sorex form.	0/10	99	2.57(1.14-4.00)
5.	40	Broken barley	1/10	4.33	7.07(3.85-8.92)
6.	40	Sorex form.	2/9	6.94(5.27-8.60)	4.59(3.24-7.43)
7.	80	Broken barley	3/5	11.17(9.36-12.73)	9.97(8.55-11.38)
8.	160	a	1/5	29.30	21.74(17.22-25.95)
9.	320	41	4/5	51,45(36,40-62,30)	51.69

sumed up to 51.69 mg/kg. Survivors have also been seen in this strain in feeding tests with brodifacoum (0.005%) for 24 hours and 48 hours (Lund 1981).

Table 7 represents results obtained with some other rodent species. A complete kill was not obtained with <u>Rattus rattus</u> in a 24-hour test, even at concentration s up to 160 ppm. This was partly due to bait refusal as the maximum amount of a.i. consumed by a survivor was 2.3 mg/kg.

As can be seen in Table 1 on the LD50 values for various rodent species, the European <u>Apodemus</u> species seem to be relatively tolerant to flocoumafen. This is confirmed in Table 7 showing that a few A. <u>flavicollis</u> survived doses up to 16.39 mg/kg.

A certain tolerance and individual variation in susceptibility is also seen in the short-tailed field vole (Microtus

agrestis) where five individuals survived doses ranging from 10.2 to 21.2 mg/kg. The multimammate rat (Mastomys (Praomys) natalensis\*) was as susceptible to flocoumafen as to bromadiolone (Lund et al. 1986), a single individual surviving, however, a 48-hour test after having consumed 7.3 mg/kg. The acceptance of various STORM products studied in choice tests with a non-poisonous cereal as the alternative, was satisfactory, especially as far as the wax blocks are concerned (Table 8). This is remarkable, as this product contains not only an insecticide and a fungicide, but also the previously mentioned bitter substance intended to increase safety by being a taste deterrent to man but not to rodents. A tendency was seen in the palatability tests of the rodents transporting the blocks from the baiting site to a shelter, where they were consumed, not stored. If the blocks were

Table 7. The effect of various concentrations and formulations of flocoumafen on singly caged rodents. Varying test periods. No choice. Sorex formulation: cut wheat.

Test	Species	Concen-	Test	Bait type	Morta-	Consumption 0	f a.i.(mg/kg)
no.		tration (ppm)	period (h)		lity	non-survivors	survivors
1. E	Rattus fattus	20	24	Broken barley	0/5		0.33(0.009-0.64)
2.	E4	20	24	Sorex form.	0/5		0.54(0.21-0.94)
3.	ii .	40	24	Broken barley	1/5	1.67	0.92(0.67-1.25)
4.	RC.	40	24	Sorex form.	2/5	1.39(1.33-1.49)	0.72(0.61-0.85)
5.	16	40	48	Broken barley	12/14	3.90(2.47-5.78)	0.40(0.22-0.58)
6.	16	80	24		1/5	3.10	1.49(0.37-2.30)
7.		160	24	14	3/5	2.21(2.02-2.50)	0,55(0.49-0.60)
8. /	Apodemus flavicollus	40	24		4/10	5.24(4.00-6.75)	5.29(3.77-7.48)
9.		50	24	Wax blocks	4/5	9.46(3.37-13.82)	16.39
10.	46	50	48	16	4/5	20.5(17.02-27.65)	13,09
11.	44	50	72	**	5/5	30.81(27.22-35.91)	
12.	0	50	96	16	5/5	20.32(9.07-26.54)	<sup>(2)</sup>
13. M	Microtus agrestis	40	48	Sorex form.	14/19	14.3(11.8-21.0)	14.2(10.2-21.2)
	Microtus arvalis	40	24	Broken barley	10/10	6.52(5,24-9.33)	
15.	ći .	40	72	12	9/9	13.53(8.17-19.71)	
16. &	Arvicola terrestris	40	24	Sorex form.+ alt.food	8/9	2,56(1,15-3,52)	1.05
17.	₩	40	48	Broken barley + alt.food	5/5	6,15(5.01-8.48)	**
18. N	Mastomys natalensis	40	48	Broken barley	24/25	10.1(4.5-14.8)	7.3

crushed, transporting ceased and palatability was improved. Experience from field trials on wax blocks also indicates that transporting is of minor practical importance, if the blocks are placed in rat holes or in bait boxes (Garforth et al. 1987).

#### **TESTS WITH PIGEONS**

To investigate the risk of flocoumafen to a non-target species often feeding at rat infested sites, an 0.005% flocoumafen cereal bait was offered to singly caged pigeons as the only food for four days. As can be seen from Table 9 one pigeon succumbed after an intake of 5.87 mg/kg. Two others were bleeding shortly after having consumed only 0.74 mg/kg and 4.7 mg/kg, respectively. Seven pigeons did not show

any symptoms of anticoagulant poisoning, presumably because they refused to consume more than tiny amounts of the bait. This preliminary study seems to indicate that flocoumafen is somewhat more toxic to this bird species than to hens.

#### CONCLUSIONS

Flocoumafen is a promising new anticoagulant rodenticide with an acute potency between that of bromadiolone and brodifacoum, but with considerable variation between species. Practical control problems with certain resistant house mouse populations in Scandinavia are likely to occur in the not-too- distant future.

Table 8. The effect of 0.005% flocoumafen baits on groups of rodents with non-poisonous choice. Test period: 4 days.

Species	Test bait	Alternative food	Mortality	Acceptance test bait/total (%)	Origin of test animals
Rattus norvegicus	Wax blocks	Rolled oats	8/10	23.6	Warfarin-susceptible strain
44	Wax blocks (crushed)	Rolled oats	7/7	33.7	Trapped on warfarin- resistant sites.
	Pellets	Rolled oats	10/10	18.5	Warfarin susceptible strain
Mus musculus	Pellets	Whole wheat	22/22	76.7	44
Apodemus flavicollis	Wax blocks	Whole wheat	25/25	55.0	Wild trapped

Table 9. The effect of 0.005% flocoumafen on cut wheat on singly caged pigeons (Columba livia) offered the bait as the only food for four days.

Pigeon		ount consum	Action to the second of the se
<u>no.</u>	bait (g)	a.i. (mg/kg	<u> </u>
1.	1.1	0.14	Surviving. No symptoms
2.	41.7	5.87	Bleeding on day 7.
			Dead on day 8. Bleedings
3.	5.3	0.74	Surviving.
			Bleeding on day 7.
4.	9.4	1.34	Surviving, No symptoms,
5.	4,2	0.51	Surviving. No symptoms.
6.	18.7	2.34	Surviving, No symptoms,
7.	<30*	<4.70*	Surviving.
			Bleeding on day 7.
8.	24.8	3.32	Surviving, No symptoms,
9.	0.9	0.11	Surviving, No symptoms,
10.	1.7	0.23	Surviving, No symptoms,

<sup>\*</sup> fouled, difficult to measure.

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