THE PROBLEM OF ANTICOAGULANT RODENTICIDE RESISTANCE IN THE UNITED STATES

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Resistance of commensal rodents to anticoagulant rodenticides is not a new phenomenon. Its confirmed presence in several areas of northern Europe is well-documented (Jackson 1969, 1972; Bentley 1969; Lund 1969). Not until 1971 was a similar situation with the Norway rat (Rattus norvegicus) to be demonstrated in the United States (Jackson et al. 1971). Because it represents an initial occurrence, the site and background observations will be described in some detail.

The rural area involved around Cleveland School in Johnson County is 25 miles SE of Raleigh, N. C. and about five miles in diameter (fig. 1). The typical farm is small (20-25A) and produces tobacco, corn, and cotton. Animal sheds (some left from days of mule power), small barns, tobacco sheds, and granaries are characteristic. Dirt floors and perforated foundation walls are common. Cleanliness is not a prime requisite, and considerable harborage (farm machinery and parts, lumber piles, tall weeds, junk, old cars) exists. Stored grains are easily accessible, as are dry foods, animal feed, and special supplements (table 1).

Table 1. Summary of Cleveland School area (Johnson County, North Carolina) premises reporting resistant rats.

		Farm Building Characteristics					Control History	
Map No.	Name	machine sheds, farm machinery	granary stored feed	tobacco storage, drying	junk	animals*	Use of anti-coagulants	Resistance noted
1	Allen #2	+	+	+	+		10 years	1970
4	Allen #1**	+	+	+		H	10 years	1/71
2	Milton Johnson**	+	+	+	+	C,D	"for years"	Fall 1970
3	Charley Barefoot		+		+	D	"quite a while"	?
5	D. R. Wells Emporium		+		+		"some time"	?
6	Mathews Grocery**	+				C	6-7 years	3-4 years
7	Tom Coats	+		+		H	6 years	4 years
8	Ben Masengil	+				D	3-4 years	1970

*C - Chickens, D - Dogs, H - Hogs
**Rats trapped on these premises included in laboratory tests

We cannot determine the detailed history of rodenticide usage in this area, but in at least several cases anticoagulants were in use for more than two decades. All farmers doing their own baiting used d-Con®. The Flowers Exterminating Company has monthly service contracts with some owners, but the present serviceman (Henry Creech) was not assigned to this area until 1968. At that time rats were present, the control difficulties of the former serviceman being attributed to poor technique. By 1970 control was difficult on all the premises, yet servicemen in adjacent territories were experiencing no difficulty in obtaining rat kills. Typically a corn-meal horse-feed bait with warfarin was used.

The efforts of the Flowers Exterminating Company met with increasing frustration. By 1971 some service contracts were being cancelled. Diphacinone was alternated with warfarin

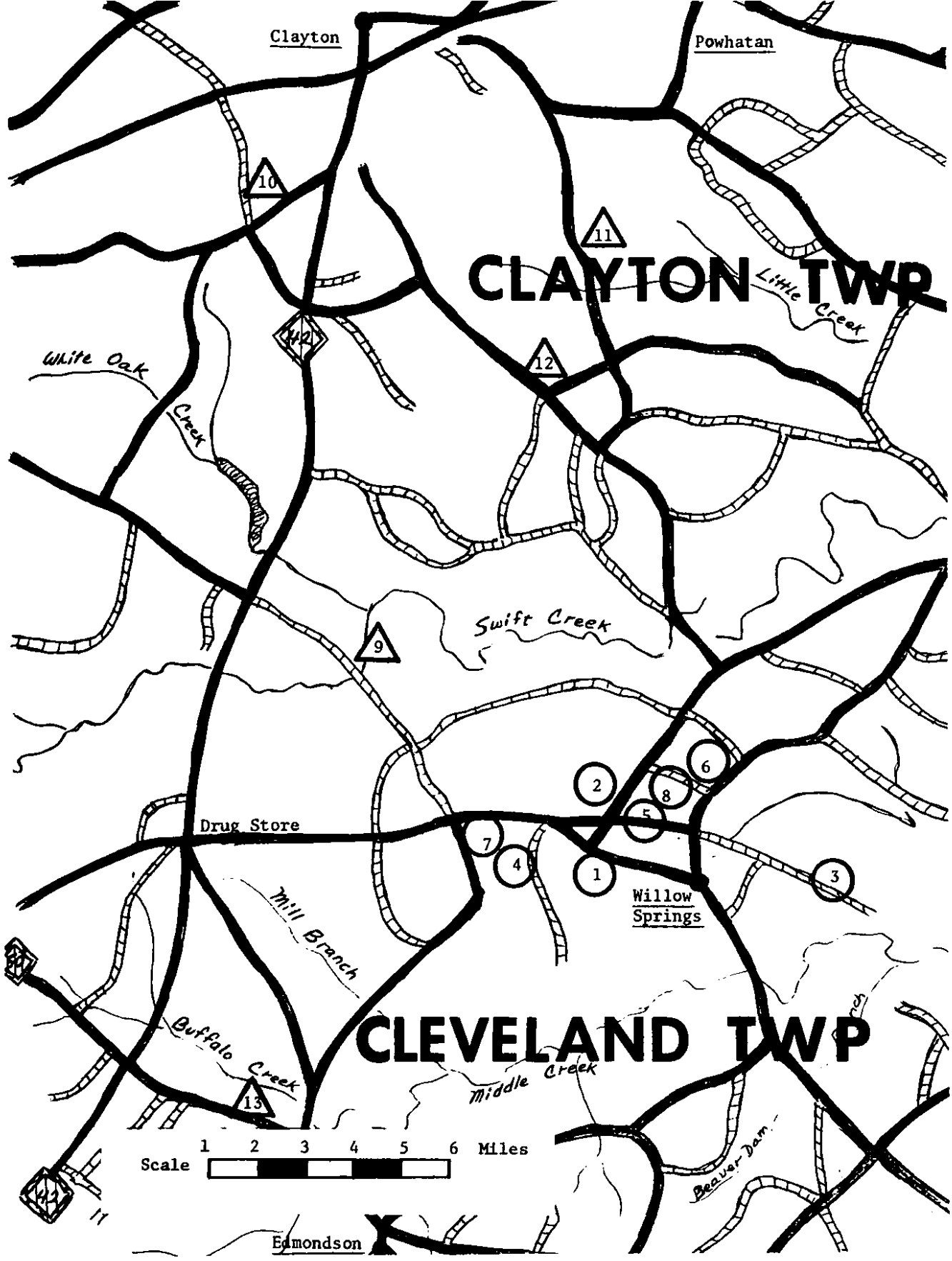

Figure 1. Anticoagulant resistance area 25 miles south of Raleigh, N. Carolina. Circles are farms where control cannot be achieved with anticoagulants. Triangles represent farms not studied intensively: #9, 10, 11 may be experiencing resistance build-ups, #12, 13 are experiencing no control difficulties using anticoagulants.

with no success. On the Johnson farm a half-gallon of pivalyn was used a day, and 200 lbs of bait had been used in one month. On some farms rats literally came out to be fed when the serviceman arrived. Rats also were invading the houses, something that had not occurred previously.

By the summer of 1971 Mr. Flowers suspected factors other than poor techniques or materials were involved. Rats were trapped on several farms and fed pivalyn and/or diphacin for up to a month and a half. Of the 10 rats, only one died within a week; three more died after two weeks of feeding. When rats from other areas were readily killed, he called on Dr. Charles Wright of North Carolina State University. In turn the National Pest Control Association and Bowling Green State University were involved.

With financial assistance available from the NPCA, arrangements were made for live-trapping of rats (which by this time had become very trap shy) by David Patterson from suspected farms and their shipment to Bowling Green laboratories. Feeding tests followed World Health Organization protocol (WHO 1970) to establish resistance levels.

Rats were individually caged, given water ad lib., subjected to no-choice feeding of 0.025% warfarin bait (ground Purina rat lab chow) for six days, followed by 22 days on placebo bait. A total of 25 rats from 4 premises were tested. All survived and were designated "resistant," based on the WHO criteria (table 2). During the test period, days of reduced food consumption or general lethargy were observed for some rats; but these individuals quickly returned to normalcy. Similar responses were observed by English workers in their evaluations (Drummond and Wilson 1968). One rat (#12) died during the post-test observation period, but death was ascribed to a large tumor. Resistant rats consumed up to five times (on a mg/kg basis) the warfarin dose of control animals. That smaller animals consumed relatively larger amounts than larger rats merely reflects differential food consumption.

That all the rats collected from the suspected premises were "resistant" was surprising. In both Britain and Denmark, when rats were collected from the resistance centers, generally less than 50% survived the initial feeding test (Drummond and Wilson 1968). This suggests intensive selection pressure (use of warfarin) has eliminated virtually all susceptible rats on these farms. A similar situation exists in the cores of European resistance sites.

These North Carolina rats have been subjected to standard feeding tests against other anticoagulants (table 3). Prolin (warfarin with an antibiotic) had no effect. Racumin, an European anticoagulant not available commercially in the U. S., killed some of the resistant rats tested, but dosages were elevated and feeding periods extended. All evidence suggests general cross resistance to all anticoagulants. (For discussion of cross resistance in European populations, see Lund 1967; Greaves and Ayres 1969b.)

At this time we do not know the full extent of the North Carolina resistance area. Initial observations and lack-of-control complaints indicate an area about 5 miles in diameter. Currently, acute poisons (largely zinc phosphide) are being used successfully to control rats in this area. Several farms (#12, 13) still report effective control with anticoagulants. Rats recently collected from a residence in Clayton may be resistant (fig. 1).

European data suggest involvement of several chromosome loci. However, the condition best studied is that of an autosomal dominant (Greaves and Ayres 1969a). Until breeding tests are completed, we have no clue as to the genetic nature of this U. S. population.

The physiological mechanism responsible for resistance is hypothesized to be an altered protein involved in the clotting mechanism (development of prothrombin) that has less affinity for the coumarin molecule than for Vitamin K. [In the susceptible rat coumarin blocks the use of Vitamin K, production of prothrombin does not occur, and the animal dies of internal hemorrhage (Greaves and Ayres 1969a; Hermondson et al. 1969).]

Greaves notes that the relative advantage of resistance in a population may be countered by "disadvantages" incurred with the condition. Apparently resistant rats require more Vitamin K than do susceptible rats to keep them in good health. Under both lab and field conditions Vitamin K may not be present in quantities to allow the survival of resistant rats, especially if they are genetically homozygous. Greaves further suggests

Table 2. Summary of no-choice feeding tests showing resistance of North Carolina rats to 0.025% warfarin in ground Purina rat chow bait.

Sex	Weight (g)		Consumption (g) for 6-day test period (0.025% warfarin)									Consumption (g) for 22-day post-test period (placebo)	
			Day of toxicant feeding						daily mean	g bait/ day/ kg rat[1]	total mg warfarin/ kg rat[1]	daily mean	g placebo/ day/ kg rat[2]
	initial	final	1	2	3	4	5	6					
1♀	154	171	14.7	14.7	5.4	14.2	12.4	13.2	12.4	80.5	121.1	13.4	78.4
2♀	142	140	18.3	18.0	10.4	16.5	15.9	17.1	16.0	112.7	169.0	15.0	107.1
3♀	351	336	26.1	27.1	17.6	24.0	23.4	23.0	23.5	67.0	100.6	19.3	57.5
4♀	118	158	14.3	16.1	9.1	14.1	14.4	14.9	13.8	117.0	175.6	14.1	89.2
5♂	304	361	38.3	33.7	19.4	25.6	22.3	21.4	26.8	88.2	132.2	19.7	54.7
6♀	340	274	25.7	25.1	20.7	24.0	23.6	22.3	23.6	69.4	104.0	18.8	68.6
7♂	294	302	34.6	24.3	11.6	33.2	25.5	24.0	25.5	86.7	130.3	20.7	68.5
8♂	170	250	17.2	16.6	17.6	17.4	20.3	16.9	17.7	104.1	155.9	20.2	80.8
9♂	153	250	20.7	20.2	21.7	20.9	22.7	21.2	21.2	138.6	208.2	20.6	82.4
10♀	188	218	23.6	15.7	22.1	23.6	23.8	19.8	21.4	113.8	171.0	16.6	76.1
11♀	245	190	17.2	15.1	16.3	12.6	15.2	17.8	15.7	64.1	96.1	13.0	68.4
12♂	314	297*	30.4	25.3	23.5	12.7	35.2	27.6	25.8	82.2	123.7	22.1*	74.4*
13♀	164	207	19.3	19.5	20.6	18.2	20.0	19.7	19.6	119.5	178.8	16.7	80.7
14♀	190	212	27.3	28.5	26.3	24.4	21.8	22.4	25.1	132.1	198.3	12.9	60.8
15♂	329	344	34.3	14.2	28.6	35.2	35.9	31.9	30.0	91.2	136.8	26.6	77.3
16♀	105	149	21.9	14.1	16.3	13.2	16.3	15.4	16.2	154.3	321.4	13.6	91.3
17♂	262	309	31.8	24.4	27.9	37.2	37.7	32.2	31.9	121.8	182.5	25.5	82.5
18♂	439	353	22.8	26.9	26.9	15.0	15.2	1.4	18.0	41.0	61.6	23.4	66.3
19♂	429	318	24.3	19.9	22.1	24.5	26.5	16.7	22.3	52.0	78.1	15.4	48.4
20♂	194	290	29.9	24.7	27.6	23.6	29.0	24.8	26.6	137.1	205.7	21.9	75.5
21♀	94	190	9.2	8.6	12.2	12.7	13.1	12.5	11.4	121.3	181.6	12.8	67.4
22♀	146	138	30.1	14.8	14.4	13.6	17.0	15.8	17.6	120.5	181.0	18.7	135.5
23♂	369	384	23.2	29.2	27.7	25.8	27.8	23.1	26.1	70.7	106.2	22.0	57.3
24♂	327	285	23.8	20.7	19.8	14.3	10.2	11.0	16.6	50.8	76.3	16.8	58.9
25♂	326	356	16.2	20.0	20.1	21.4	21.0	21.4	20.0	61.3	92.1	19.8	55.6

* died day 16, see text; 1 - based on initial rat weight; 2 - based on final rat weight

Table 3. Summary of no-choice feeding tests using resistant rats and other anticoagulants (Prolin, 0.025%; Racumin, 0.0375%) in ground Purina rat chow bait.

Sex	Weight (g) initial	Weight (g) final	Consumption (g) for 6-day test period (toxicant) Day of toxicant feeding 1	2	3	4	5	6	daily mean	g bait/ day/ kg rat[1]	total mg toxicant/ kg rat[1]	Consumption (g) for 22-day post-test period (placebo) daily mean	g placebo/ day/ kg rat[2]
Prolin (0.025% warfarin and 0.025% Sulfaquinoxaline)													
1♀	171	161	15.2	15.5	13.7	12.8	15.8	13.4	14.4	84.2	126.3	15.5	96.3
2♀	140	254	21.7	26.9	18.9	12.7	18.5	14.3	18.8	134.3	201.8	18.7	73.6
Racumin (0.0375% 3-(u-tetralyl)-4-hydroxycoumarin)													
1♀	212	238	31.6	14.9	22.7	11.9	4.5	3.2	14.8	69.8	157.1	17.6	73.9
2♂	345	?	29.3	26.9	31.8	14.3	3.5	1.7	17.9	51.9	116.8	(A)	----
3♀	150	186	16.8	16.2	14.6	4.6	0.3	7.6	10.0	88.7	150.2	13.3	71.5
4♂	309	?	31.8	31.1	20.0	28.2	9.7	3.8	20.8	67.3	151.2	(B)	----
5♂	335	328	18.9	19.7	16.6	1.0	0.6	0.4*	9.5	28.4	64.0	----	----
6♀	121	134	11.5	0.9	0.9	0.8	7.6	5.9*	4.6	21.7	85.5	----	----
7♂	321	356	22.4	20.9	19.6	10.3	0.4	3.1	12.8	39.9	89.6	19.4	54.5
8♂	264	273	20.7	21.8	17.6	11.2	3.0	9.5	13.7	51.9	119.0	(C)	----

* died day 6; 1 - based on initial rat weight; 2 - based on final rat weight

(A) - Died day 10. Consumption day 7-0.9, 8-0.3, 9-0.5, 10-0.0.

(B) - Died day 8. Consumption day 7-0.3, 8-0.0.

(C) - Died day 8. Consumption day 7-1.2, 8-1.2.

that a cessation of anticoagulant poisoning of resistant populations might cause the resistance to disappear in the population in a few years or less, if the phenomenon is associated with some survival disadvantages (Greaves 1970).

The North Carolina rats had a propensity to bleed. Several rats were toe-clipped upon capture in North Carolina and subsequently died, possibly from blood loss. Resistant animals at Bowling Green were released in a 12' square room for breeding purposes, and two died apparently from blood lost from wounds received in fighting. Such wounds would not normally have proved lethal.

Why did resistance develop on these North Carolina farms? The parallels with their European counterparts are clear: abundant rat harborage, accessible foods, mild climate, probably field and fencerow populations, regular and long term use of warfarin and other anticoagulants. Farmers similarly were content to exist with poor premise sanitation as long as rodenticides could eliminate the most obvious rats. The PCO found it easier to regularly place bait stations than change human behavior.

Under these conditions rats bred readily, and those that carried the genetically-controlled capability to feed indefinitely (or at least frequently) on the warfarin baits and live had the selective advantage. They survived and passed their capability on to at least some of their offspring. Any rats killed were replaced either by the young or individuals emigrating from the surrounding farm lands or adjacent structures.

This pattern, in operation for a decade, more or less, resulted in the emergence of rat populations wholly resistant to warfarin. The distribution of resistance is both a matter of selection pressure and our ability to detect the resistance (Drummond 1970).

In cooperation with the Urban Rat Program in the City of Philadelphia, rat populations in some of the 30 cities having federally supported control programs will be sampled and standard evaluations for resistance made this spring and summer. Hopefully (if their program is not terminated by the State legislature) the New York State Rat Control Laboratory also will be involved, and rats in all of the cities can be studied. In this way some determination of susceptibility levels of rat populations to warfarin in widely separated urban areas will be possible, and identification of actual or potential resistant areas can be made. Rural areas, so far the foci of all resistant populations, will not be examined, however. At this time circumstantial evidence does suggest that several other rural areas in the United States may have developed resistant Norway rat populations.

Summary

The development of anticoagulant resistance by Norway rats in this rural North Carolina area is not surprising. By mutual consent, both farmers and PCO's were depending on the use of anticoagulants to override the poor premise sanitation and building maintenance and keep the rats "under control." This worked reasonably well until the mid-to-late sixties. The intensive use of anticoagulants (mostly warfarin) over a decade or more provided the selective agent to develop resistant populations. Probably this pattern will be repeated elsewhere in the United States.

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