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Background Prevalence of Tetracycline-like Fluorescence in Teeth of Free Ranging Red Foxes (*Vulpes vulpes*), Striped Skunks (*Mephitis mephitis*) and Raccoons (*Procyon lotor*) in Ontario, Canada

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ABSTRACT: We collected and examined teeth from 3406 red foxes (*Vulpes vulpes*) collected in Ontario, Canada, from 1978 to 1986, prior to large scale rabies vaccine baiting. We found tetracycline-like fluorescence in five (0.2%) of the samples. Also, we observed similar fluorescences in five (0.4%) of 1103 striped skunks (*Mephitis mephitis*) and in six (0.8%) of 744 raccoons (*Procyon lotor*). The low prevalence of such marks would not appear to invalidate the use of tetracycline as a marking agent in vaccine baiting trials.

Key words: Fluorescence, tetracycline, teeth, Ontario, *Vulpes vulpes*, *Mephitis mephitis*, *Procyon lotor*.

A standard method of estimating the acceptance of vaccine-baits during oral vaccination trials for rabies control is to incorporate tetracycline within the bait matrix (Wandeler et al., 1988). Potential rabies vectors eating baits readily incorporate the tetracycline in the dentine and cementum of their teeth. Presence of tetracycline in the teeth is identified by a characteristic yellow fluorescence emission under ultra violet light microscopy (Johnston et al., 1987). However, teeth of sampled animals also may contain tetracycline from environmental sources or other substances with fluorescence similar to that caused by tetracycline from a bait (World Health Organization, 1992). Our objective was to estimate the possible error in evaluating bait acceptance due to background or non-specific fluorescence, by examining teeth of rabies vectors sampled previous to large scale rabies baiting experiments in Ontario, Canada.

Red foxes (*Vulpes vulpes*), striped skunks (*Mephitis mephitis*) and raccoons (*Procyon lotor*) were captured by hunters and

trappers as part of their standard activities (Novak, 1987) and subsequently collected by rabies unit staff. Specimens examined in this study were captured either previous to the initial distribution of baits containing tetracycline in 1978 to 1983, or in 1984 to 1986 when oral baiting trials were taking place. In baiting years, samples were collected >5 km outside of baited locations for raccoons and skunks, and <50 km for foxes; this was the estimated size of home ranges and movements of each species (Rosatte, 1987; Sanderson, 1987; Voigt, 1987). To simplify comparison with the results of future baiting programs, samples were grouped into three general geographic categories. Fox specimens were obtained throughout the province from the following counties in southwestern Ontario: Bruce (44°48'N, 81°25'W), Grey (44°20'N, 80°45'W), Huron (43°40'N, 81°30'W), Lambton (42°45'N, 82°05'W), Perth (43°30'N, 81°05'W), and Simcoe (44°25'N, 79°50'W); in southeastern Ontario: Leeds-Grenville (44°50'N, 75°40'W), Ottawa-Carleton (44°20'N, 75°35'W), Prescott (45°30'N, 74°45'W), and Stormont-Dundas-Glengarry (45°15'N, 74°40'W); and in northern Ontario: Cochrane (50°00'N, 83°00'W), Manitoulin (45°45'N, 82°30'W), Sudbury (47°10'N, 82°00'W), Thunder Bay (49°30'N, 88°30'W), and Timiskaming (47°45'N, 80°20'W). Raccoons were collected only in southwestern Ontario from Huron and Perth Counties. Skunks also were collected only in southwestern Ontario from Huron, Lambton, Perth and Simcoe Counties.

Canine teeth were extracted, prepared, sectioned and examined for the presence

TABLE 1. Incidence of fluorescence in canine teeth of Ontario red foxes (*Vulpes vulpes*), 1978 to 1986.

Area	Year collected	Number sampled	Number with fluorescence (%)
Southwestern Ontario	1978	677	3 (0.4)
	1979	931	0
	1980	293	2 (0.7)
	1981	329	0
	1984*	165	0
	1985*	139	0
	1986*	157	0
Southeastern Ontario	1979	26	0
	1980	115	0
	1981	153	0
Northern Ontario	1977	39	2 (2.6)
	1979	231	0
	1981	151	0

* Specimens collected during a baiting year in locations >50 km from any baited area.

of fluorescence as described by Johnston et al. (1987). Tetracycline-like fluorescence in sampled teeth was considered positive if it was of characteristic yellow color and intensity similar to that seen in experimental trials of tetracycline deposition in captive fox teeth (Johnston et al., 1987).

With some exceptions, the prevalence of tetracycline-like fluorescences in the teeth of all species examined was extremely low (Tables 1, 2). Incidences of positive fluorescence were the result of point occurrences: a number of positive specimens collected within a single county within the year. The highest prevalences found were similar to the 3.2% prevalence of fluorescence found in the teeth of juvenile foxes sampled in Switzerland (Kappeler, 1991). In a similar study, no fluorescence was found in 18 foxes from non-baited areas in metropolitan Toronto (Rosatte et al., 1992).

Other fluorescences also were seen in the teeth of 64 (1.9%) of 3406 sampled foxes; but due to differences in color intensity and location within the teeth, these were not attributed to tetracycline. Kappeler (1991) observed similar nonspecific fluo-

TABLE 2. Incidence of fluorescence in canine teeth of Ontario striped skunks (*Mephitis mephitis*) and raccoons (*Procyon lotor*), collected 1978 to 1986, southwestern Ontario.

Species	Year collected	Number sampled	Number with fluorescence (%)
Striped skunks	1978	176	3 (1.7)
	1979	657	0
	1981	80	0
	1984*	63	0
	1985*	75	0
	1986*	52	2 (3.8)
Raccoons	1978	156	5 (3.2)
	1979	209	0
	1980	55	1 (1.8)
	1981	200	0
	1984*	26	0
	1985*	61	0
	1986*	37	0

* Specimens collected during a baiting year in locations >5 km from any baited area.

rescences and suggested that they were the result of other substances. The complexity of possible fluorescence in sampled teeth underlines the importance of having trained observers evaluating baiting trails.

Tetracycline deposition in the teeth of potential rabies vectors was most likely the result of agricultural and animal husbandry practices. Tetracycline is a common antibiotic used in treatment of livestock (Canadian Animal Health Institute, 1993). Topical, intramuscular and oral administration of tetracycline is not considered to produce sufficient concentrations of the drug in flesh of livestock to mark animals eating offal or carcasses (M. Hazlett, pers. comm.). Kappeler (1991) suggested that the intra-uterine administration of 2 to 3 g of tetracycline to treat metritis (placental retention, inflammation and possible infection), followed by expulsion of the afterbirth either naturally, or via the administration of oxytocin would provide a ready source of the drug to mark wild animals. Kappeler (1991) calculated that this procedure results in 30,000 to 60,000 tetracycline contaminated afterbirths

yearly in Switzerland (5 to 10% of all births), which may be eaten by foxes. Treatment of metritis also is a common veterinary activity in Ontario (M. Hazlett, pers. comm.). Dohoo et al. (1982) determined that 294 (9.0%) of 2711 lactating Holstein-Friesian dairy cattle suffered from reproductive tract infections, including acute metritis, as diagnosed in the first 21 days of lactation. As in Switzerland, treatment of reproductive tract infections may account for some of the tetracycline in the teeth of Ontario foxes, skunks and raccoons. Although the amount of tetracycline contained in a contaminated after-birth is unknown, <50 mg tetracycline will mark 100% of juvenile foxes; ≥75 mg are necessary to consistently mark adult animals (Lawson et al., 1992).

In summary, we observed tetracycline or tetracycline-like marks in teeth of carnivore species targeted by present and future baiting programs in Ontario. Although the prevalence of such marks can reach 3% in localized situations, they do not appear to invalidate the use of tetracycline as a marker to evaluate the results of large scale bait distribution trials.

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