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of the stock in a herd are affected (Shupe et al., 1967, J. Am. Vet. Med. Assoc. 151: 191–197). The syndrome is characterized by arthrogryposis, scoliosis, torticollis, and cleft palate. Cleft palate may occur with other abnormalities, but occasionally it is the only detectable deformity (Shupe et al., 1967, op. cit.). The disease is the result of pregnant animals ingesting plants containing teratogenic alkaloids between the 40th and 70th days of pregnancy. One genus noted for its teratogenic alkaloids is *Lupinus* (Keeler et al., 1977, op. cit.; Keeler, 1978, op. cit.). The gestation period in tule elk is approximately 250 days with the rut in September and most calving in May or early June (McCullough, 1969, op. cit.). Thus, the period of insult in tule elk would be in October and November. Dietary records for September 1980, through May 1981 (Gogan, unpubl. data), showed that *Lupinus* spp. (mainly *L. arboreus*) constituted between 2 and 31% of the diet in bimonthly samples between early September and late November 1980, and less than 1% in bimonthly samples between early December 1980, and May 1981. Thus, the diet of tule elk included a plant species with the potential to contain high levels of teratogenic alkaloids during a critical period of fetal development.

The causes of the cleft palate in this calf remain speculative. Circumstantial evidence suggests inbreeding or teratogenic plant alkaloids as possible causes. Closer inspection of fetuses from cervid species with varying levels of inbreeding and use of teratogenic plants may provide insight into the frequency and cause of this deformity.

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Brain Cholinesterase Activity in Starlings and Japanese Quail Dosed with Methiocarb

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Many species of birds damage agricultural crops. Grain (Besser et al., 1968, J. Wildl. Manage. 32: 179–180; Besser, 1973, Internat. Rice Comm. Newsletter (22: 9–14; Shefte et al., 1981, Denver Wildlife Research Center, Bird Damage Res. Rep. No. 124, 7 pp.) and fruit (Guarino et al., 1974, J. Wildl. Manage. 38: 338–342; Dolbeer et al., 1974, Proc. 6th Bird Control Sem., pp. 28–40) can be severely damaged by birds producing significant mon-

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etary loss. In California, damage to wine grapes by European starlings (*Sturnus vulgaris*) and other birds may be extensive (Crase et al., 1976, U.S. Dept. of the Interior, Fish and Wildlife Service, Spec. Sci. Rep.—Wildl. No. 197, 18 pp.; De Haven, 1974, Proc. 6th Vert. Pest Conf., pp. 248–252). Interest is being focused on the use of non-lethal chemical bird repellants that can be applied directly to crops (Schafer and Brunton, 1971, J. Wildl. Manage. 35: 569–572; Crase and De Haven, 1978, Proc. 8th Vert. Pest Conf., pp. 46–50). Among these chemical is methiocarb (Mesurol®: 3,5-dimethyl-4-(methylthio)phenylmethylcarbamate), which has been shown to repell birds from many crops (Crase and De Haven, 1978, op. cit.).

Methiocarb is a carbamate and therefore potentially is a cholinesterase (ChE) inhibitor. As a ChE inhibitor it could kill birds. The objectives of this study were to determine if methiocarb might kill birds by inhibiting brain ChE, the amount of methiocarb necessary to produce ChE inhibition, the magnitude of brain ChE inhibition that suggests methiocarb is the cause of death, and if brain ChE inhibition by methiocarb varied depending upon the time of day the birds were given the compound.

In this study starlings and Japanese quail (*Coturnix japonica*) were used. The starlings were captured with decoy traps at various locations in California. Japanese quail were hatched and raised at the U.S. Fish and Wildlife Service Field Station, Dixon, California. Each of the two species was maintained separately in communal 6.1 × 3.0 × 2.3-m outdoor wire cages and provided food and water ad libitum. Starlings were fed poultry pellets (Albers Egg-Maker 20) and Japanese quail were fed turkey feed (Albers 24–27%). Birds were held at least 6 wk before tests were conducted. Only healthy appearing birds that were above a minimum weight (starlings ≥70 g; Japanese quail ≥100 g) were used in the study. Birds were assigned randomly to treatment groups.

Methiocarb was used in a 99% wetable powder formulation. The chemical was dissolved in propylene glycol. Four evenly spaced, ¼ log doses beginning at 5.6 mg/kg were given by gavage after 2 hr of fasting. The concentration of methiocarb was adjusted so that each bird received 2 µl of propylene glycol per gram of body weight. The birds were exposed to 14 hours light and 10 hours dark. A group of birds was dosed in the morning (1–2 hr after light) (A.M.-birds) and a second group was dosed in the evening (1–2 hr before dark) (P.M.-birds). After dosing the birds were placed in individual cages and observed for 1 hr for clinical signs of toxicity and mortality. After 1 hr surviving birds were killed using chloroform. The birds were frozen and stored at −20 C until brains were analyzed for brain ChE.

Brain ChE (EC 3.1.1.7) activity was determined by the method of Ellman et al. (1961, Biochem. Pharmacol. 7: 88–95) as adapted for brain tissue by Dieter and Ludke (1975, Bull. Environ. Contam. Toxicol. 13: 257–262) and Zinkl et al. (1977, Bull. Environ. Contam. Toxicol. 17: 379–386). All samples were analyzed 1 day after the birds were given methiocarb. ChE activities were calculated in mU (milliunits)/mg of brain. One unit is defined as the conversion of one mole of substrate to product(s) in 1 min. All analyses were carried out at 25 C.

Kruskal-Wallis one-way analysis of variance (Kruskal and Wallis, 1952, J. Am. Stat. Assoc. 47: 583–621) was used to determine if differences in brain ChE activity between A.M. and P.M. birds were present and to determine if a difference between treatment levels existed. If a probability ≤0.05 was found, Scheffe's test was used to determine which (if any) of the treated birds' brain ChE differed from the control birds' brain ChE activities (Kleinbaum and Kupper, 1978, Applied

TABLE 1. Brain cholinesterase (ChE) activity of starlings following oral exposure to methiocarb.

Treatment level in mg/kg	No. of birds				ChE activity					
	A.M.		P.M.		Mean (mU/mg)		SD		% Inhibition	
	Tested	Died	Tested	Died	A.M.[a]	P.M.[a]	A.M.	P.M.	A.M.	P.M.
Control	4	0	4	0	22.8	23.0	2.4	1.8	0.0	0.0
5.6	4	0	4	0	22.5	20.3	4.1	2.5	1.6	11.7
10.0	4	0	4	0	17.0	16.5	2.3	1.1	25.3	28.0
17.8	4	0	4	0	17.5	16.8	2.5	1.0	23.1	26.7
31.6	4	0	4	1	14.9[b]	14.2[b]	0.8	6.4	34.8	38.2

[a] Dose response significant at $P < 0.05$.
[b] Significantly different from the combined control brain ChE activity at $P < 0.05$.

Regression Analysis and Other Multivariable Methods, Duxbury Press, North Scituate, Massachusetts, pp. 271–276). Determination of a dose response was by Jonckheere's (1954, Biometrika 41: 133–145) method.

After dosing both avian species showed signs of intoxication. Within a few minutes both species of birds had wing fluttering, were immobilized, had tachypnea and some regurgitated. Birds that died showed signs of tetany. The birds that survived usually appeared normal at the time they were killed (1 hr after dosing). A single starling given 31.6 mg methiocarb/kg in the afternoon died (Table 1). Among the Japanese quail, deaths occurred in all groups that received methiocarb except the P.M., 5.6 mg/kg group (Table 2).

In neither species did brain ChE activity between A.M. and P.M. birds that received similar doses differ significantly. For starlings only the birds given 31.6 mg methiocarb/kg had significantly depressed brain ChE activities (Table 1). However, for Japanese quail all groups that received methiocarb showed significane brain ChE depression (Table 2). For both species there was a significant dose related brain ChE inhibition (Tables 1 and 2). Greater inhibition of brain ChE activity at similar doses of methiocarb occurred in Japanese quail than in starlings.

The least brain ChE depression found in a bird that died was 46.1% in the A.M., 5.6 mg/kg Japanese quail. The greatest brain ChE depression in quail was 62.6% in a P.M., 17.8 mg/kg bird. In the single starling that died after being given 31.6 mg/kg, 69.4% depression was found.

TABLE 2. Brain cholinesterase (ChE) activity of Japanese quail following oral exposure to methiocarb.

Treatment level in mg/kg	No. of birds				ChE activity					
	A.M.		P.M.		Mean (mU/mg)		SD		% Inhibition	
	Tested	Died	Tested	Died	A.M.[a]	P.M.[b]	A.M.	P.M.	A.M.	P.M.
Control	5	0	4	0	8.0	8.3	0.7	0.9	0.0	0.0
5.6	5	1	5	0	6.1[c]	6.0[c]	1.5	0.7	23.8	27.0
10.0	5	2	5	2	4.4[d]	4.4[d]	0.6	1.1	44.9	46.1
17.8	5	4	5	4	4.3[d]	3.2[d]	0.6	0.7	46.4	60.5
31.6	5	5	5	5	3.7[d]	3.5[d]	0.5	0.4	53.6	57.4

[a] Dose response significant at $P < 0.01$.
[b] Dose response significant at $P < 0.05$.
[c] Significantly different from the combined control brain ChE activity at $P < 0.05$.
[d] Significantly different from the combined control brain ChE activity at $P < 0.001$.

Among the quail 23 of 26 birds that had brain ChE depression of 45% or greater died. Of the starlings only two birds had brain ChE depression of greater than 45%. One of these died with 69.4% depression, while the survivor (P.M., 31.6 bird) had 51.7% depression.

Sufficient data do not exist to determine the potential for methiocarb to act as a lethal agent rather than as a repellant in field situations. Data obtained for four species of African pest birds suggest that the 50% effective repellancy dose (R_{50}) and the LD_{50} are not greatly different. For example, the R_{50} for masked weavers (*Ploceus tainiopterus*) was 1.78 mg/kg (95% confidence interval (CI) of 1.28–2.48) and the LD_{50} was 4.87 (95% CI of 3.14–7.55) (Shefte et al., 1981, op. cit.). In contrast, Schafer et al. (1975, Bull. Environ. Contamin. Toxicol. 14: 641–647) found methiocarb concentrations greater than 100 ppm in food were necessary to kill common grackles (*Quiscalus quiscula*), whereas residues on food in most field studies have been 30 ppm or less, often much less (Bailey and Smith, 1979, Aust. J. Exp. Agric. Anim. Husb. 19: 247–250; Guarino et al., 1974, op. cit.).

Methiocarb has ChE inhibiting activity, and could, as can other carbamate and organophosphate anticholinesterases, kill birds. This study suggests that methiocarb at doses of 5.6 or 10.0 mg/kg sufficiently inhibited brain ChE of Japanese quail to cause death in a few birds. For quail, brain ChE inhibition of about 45% or greater was usually lethal within 1 hr after dosing. Only three of 26 quail whose brain ChE inhibition was greater than 45% survived for 1 hr.

Only one starling died and its brain ChE depression was 69.4%. Only one other starling had brain ChE depression greater than 45%. Thus the data indicate that Japanese quail are more susceptable to methiocarb than are starlings. These conclusions may not be precise, however, because regurgitation of the methiocarb-propylene glycol mixture occurred in more birds and the amount of material regurgitated appeared to be greater in starlings than in Japanese quail. Regurgitation is one of the effects of methiocarb that causes birds to be repelled from crops treated with methiocarb (Zabadal and Hothem, 1979, Wines and Vines 60: 38–41).

In some field studies in which methiocarb was used as a repellant and in which searches for dead birds have been conducted, none have been found (Bailey and Smith, 1979, op. cit.; Zabadal and Hothem, 1979, op. cit.). However, in one study of methiocarb's ability to protect sprouting peas, dead house sparrows (*Passer domesticus*) and greenfinches (*Carduelis chloris*) were found. The dead birds often had copious liquid in and around their bills, and their bodies were stiff and stretched (Porter, 1977, N.Z. J. Exp. Agric. 5: 335–338). These observations suggest salivation and tetany occurred before death, signs seen with anticholinesterase insecticide poisoning.

Therefore, it is possible that certain birds might be very sensitive to methiocarb, and losses could be expected when methiocarb is applied (or misapplied) at a high rate. Our results suggest that if methiocarb is to be implicated in the death of birds after its use in the field, the brain ChE inhibition should be 45 to 50% or more. This contrasts with the more than 70% inhibition that usually is found in birds poisoned by anticholinesterases (Hill and Fleming, 1982, Environ. Toxicol. Chem. 1: 27–38).