are in a sample of hunters holding licenses. We found 7.2 dove hunters per 100 telephone contacts, in contrast to state mail surveys of the Southeast where dove hunters seem to constitute about 21 to 37 percent of the licensed hunters for several surveys in our files.

Any comparison of these two methods of survey must also consider the costs. We do not have exactly comparable cost data for a mail survey, but one state biologist kindly furnished us with records for a 1973-74 mailing of 10,800 questionnaires from which we have derived an estimate allowing for 6 percent inflation of 88 cents per questionnaire mailed, comparable to our latest cost estimate of $1.92 per call attempted.

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ORAL ACCEPTANCE AND ANTIFERTILITY EFFECTS OF MICROENCAPSULATED DIETHYLSTILBESTROL ON WHITE-TAILED DOES[1]

by

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ABSTRACT

The acceptance and antifertility action of microencapsulated diethylstilbestrol (DES) administered in feed was investigated with penned female white-tailed deer (*Odocoileus virginianus*). A switchback designed oral acceptance test at 0, 250, 500, 750, and 1,000mg was conducted just before the breeding season. The 1,000mg level was as well accepted as the other three concentrations, but none were as well accepted as the control. Six does were presented 1,000mg of DES, homogenized in 1.362kg of feed, every 17 days throughout the breeding season. Five of the six does demonstrated aversion to the compound. Consumption of 131mg or less did not prevent normal pregnancy. The sixth doe, which consumed 182 and 428mg at the first two feedings, bred again after each feeding indicating that these levels might have interrupted pregnancy. Possible reasons for the poor acceptance of DES during the breeding season are discussed. If the rejection is due to metabolic aversion, microencapsulated DES may never work as a multiple-dose antifertility agent; if it is due to taste or smell, a different microencapsulation formulation might overcome the aversion problem.

INTRODUCTION

Diethylstilbestrol (DES), a biologically active synthetic estrogen, is an effective postcoitum contraceptive (Diczfalusy 1968). However, like the natural estrogens, this compound at high concentrations results in significant reduction in feed intake (Bull et al. 1974). Harder and Peterle (1974) found poor acceptance when they fed DES in a bait carrier to free-ranging white-tailed deer in Ohio. By microencapsulating DES, I hypothesized that its taste and smell would be masked and that its acceptance by white-tailed deer might thus be increased. Microencapsulation is a technique which gives each individual drug particle a protective coat from which the drug can be released at a rate depending upon moisture, pH, physical force, or combinations of these (Luzzi 1970). In this study, DES was microencapsulated with a type of food shellac designed to dissolve and release the compound in the rumen. The manufacturer, Abbott Laboratories, reported that this type of coating was able to increase the acceptance of antibiotics by swine from 3 grams per ton for uncoated to 100 grams per ton for coated (Macy, personal communication, 1975).

[1] A contribution from a cooperative project between the National Park Service and the U. S. Fish and Wildlife Service, U. S. Department of the Interior.

This study was conducted at Mammoth Cave National Park, Kentucky. Its objectives were: (1) to determine if a microencapsulated DES presented in feed at 0, 250, 500, 750, or 1,000mg would be accepted by does; and (2) to determine the antifertility effects of DES when fed on a regular schedule during the breeding season.

I thank Lowell Macy of Abbott Laboratories for formulating and supplying the microencapsulated DES.

PROCEDURE

Test one: *Oral acceptance switchback design.* The five DES concentrations (0, 250, 500, 750, and 1,000mg) were fed to does in October, 1973. The experimental design followed standard switchback procedures for two or more treatments with the first and third feedings at the same concentration and the second feeding at a different concentration, either higher or lower. Treated feed was offered during 24-hour treatment periods, and there was a 6-day adjustment period between treatments. Ten does, yearling or older, were placed in individual stalls, 3.0 × 4.6 meters, and two were randomly allotted to each DES concentration. Pretesting showed the does would readily consume 1.362kg (3 lb.) daily. During the three 6-day adjustment periods, each doe was offered only 1.362kg per day of a dairy ration containing 16 percent protein; on the 3 treatment days the microencapsulated DES was mixed with the dairy ration. Feed consumption was measured daily, and the data was analyzed by the switchback design procedures for five treatments as outlined by Lucus (1956).

Test two: *Postcoitum antifertility effects.* The breeding season began on November 11, when 9 of the 10 does from test one, plus one additional untested doe were randomly placed (6 treated, 4 control) on a DES feeding regime. Every 17 days throughout the breeding season, beginning on November 11, 1973, 1,000mg DES was added to the treated does' ration. The oral dose sufficient to cause resorption or abortion in white-tailed deer is unknown, and the 1,000mg level was selected on the basis of the research by Hill and Pierson (1958) who aborted cattle in early gestation with a 100mg intramuscular dose of DES. The rule of thumb applied was that an oral dose must be 10 times greater than a given intramuscular dose to have the same effectiveness. Two does and one adult male were placed in two connecting stalls. The 16 percent dairy ration was available *ad libitum* for 16 days; on the evening of the 16th day, the deer were fasted (except for the February 15, 1974, feeding) and on the morning of the 17th day, the male was removed and the females were separated into individual stalls and given 1.362kg of dairy ration containing 1,000mg DES for the treated does and 0mg for controls. The following morning feed consumption was measured, the males were returned and allowed to mix with the two females, and all were placed on the regular ration. In February, at the end of the breeding season, the bucks were removed and the does were held in individual stalls and allowed to fawn. Data for feed consumption, fawning dates, and fawns per treatment were analyzed by one-way analysis of variance (ANOV), and mean separation was by Scheffe's test (Snedecor and Cochran 1967); $P \leq .05$ was the criterion of significance.

RESULTS AND DISCUSSION

Test one. All 10 does consumed some microencapsulated DES during all three treatment periods (Table 1). Except for doe 1, does receiving a lower concentration at the second feeding ate more of the bait, and those receiving a higher concentration ate less than during the first feeding. All except doe 3 ate less during the third feeding than during the first, even though the DES concentration level was the same. Only twice did animals eat the entire 1.362kg ration — the first feeding for doe 8 (250mg DES) and the first feeding for doe 10 (0mg).

Mean feed consumption decreased as the DES concentration increased except that the 1,000mg mean was slightly higher than the 750mg mean. An ANOV showed a significant difference among treatments and mean separation showed the following (means not underscored by the same line are significantly different):

Treatment:	0mg	250mg	500mg	1,000mg	750mg
Mean Feed Consumption (g):	1053.4	848.9	585.5	327.0	315.2

Test two. At the first feeding on November 28, only half the animals in either the treated or control groups ate any of their ration (Table 2), probably because of thunderstorms with 5 inches of rain that night. One treated animal, doe 6, consumed 182mg of DES and the other two (does 7 and 8),

consumed less than 40mg. During the second feeding, all does consumed less than 70mg of DES except doe 6, which consumed 428mg. Doe 1 died of causes unrelated to experimental treatment after the second feeding. During the following feedings, four of the remaining five animals refused most or all of their feed and never consumed as much as 50mg of DES. Doe 6 continued to eat more than the others (accounting for 72 percent of the 1,391mg of DES consumed by all does) but also showed a progressive decline in consumption. An ANOV showed a significant difference between the treated and control groups in feed consumption during the first four feedings; consumption averaged 86.7g for the treated does versus 522.6 for the controls.

Table 1. Consumption of microencapsulated DES by penned, nonbreeding white-tailed does on three dates when five concentrations were offered in single daily rations (1.362kg of feed) according to a switchback design.

	Oct. 13, 1973		*Oct. 20, 1973*		*Oct. 27, 1973*	
Doe No.	*DES in feed (mg)*	*DES consumed (mg)*	*DES in feed (mg)*	*DES consumed (mg)*	*DES in feed (mg)*	*DES consumed (mg)*
1	1000	351	750	151	1000	132
2	1000	159	250	127	1000	113
3	750	129	500	210	750	177
4	750	331	0	0	750	156
5	500	102	1000	59	500	0
6	500	491	0	0	500	468
7	250	228	750	263	250	121
8	250	250	500	163	250	90
9	0	0	1000	263	0	0
10	0	0	250	46	0	0

I thought that the stress of moving the males out and separating the does at each feeding may have affected consumption, so in February the males were permanently removed and the females separated into individual stalls. Feed consumption was recorded daily until a pattern was established and then the five treated does were again offered the DES ration. They refused almost all of it and together the five does consumed only 57.7mg of DES (Table 2).

Two animals died before fawning. Doe 1, a treated animal which died on December 15, had ovulated, but no conceptus or membranes were visible in the uterus. Doe 9, a control, was pregnant when she died on February 12. The remaining eight does fawned between May 26 and July 7. The three control and five treated does gave birth to six and seven fawns, respectively. An ANOV showed no significant difference between treatments in fawning dates or number of fawns per doe.

DES consumption of 70mg or less was insufficient to interrupt pregnancy in five of the pregnant does. However, consumption of 182mg or more apparently interfered with early pregnancy in doe 6. This doe bred on November 12, and on November 28 she consumed 182mg of DES. On December 5, she again came into heat and was bred, and on December 15 she consumed 428mg. She was not pregnant on January 1, as she bred a third time on January 5. If she conceived she would have been pregnant about 16 and 10 days, respectively, at the time of DES ingestion and probably at the preimplantation state of gestation when 182 and 428mg interrupted her first and second pregnancies. The 260mg dose on January 1 did not interfere with ovulation, as she ovulated and conceived 4 days later. Consumption of 131mg on January 18 failed to interrupt her third pregnancy. There was no direct evidence that this doe was actually pregnant in November and December, but she had shown no previous history of abnormal breeding behavior. Since she was the only animal consuming any quantity of DES, I believe that her breeding pattern was probably a treatment effect.

Unlike some steroids such as 6-chloro Δ^6-17 acetoxyprogesterone (CAP) (Wagner 1964) and quinestrol (Falconi et al. 1972), DES did not appear to be stored during test one and then later metabolized during the breeding season in sufficient quantity to interfere with the estrus cycle. All nine surviving does from the switchback experiment came into estrus and conceived before the first feeding on November 28, as evidenced by back dating the fawning dates to obtain the conception

Table 2. Consumption of bait by penned white-tailed does on five dates during the breeding season when 0 or 1000mg of microencapsulated DES was offered in single daily rations (1.362kg of feed); after January 18, males were permanently removed and new feeding patterns established.

	Treatment days								*Pretreatment*	*Treatment day*	
	Nov. 28, 1973		*Dec. 15, 1973*		*Jan. 1, 1974*		*Jan. 18, 1974*		*Feb. 14, 1974*	*Feb. 15, 1974*	
Doe No.[a]	*Bait consumed (g)*	*DES consumed (mg)*	*Bait consumed (g)*	*DES consumed (mg)*	*Bait consumed (g)*	*DES consumed (mg)*	*Bait consumed (g)*	*DES consumed (mg)*	*Untreated feed consumed (g)*	*Bait consumed (g)*	*DES consumed (mg)*
Treated (1000mg DES in bait)											
1	0	0	5	4		(Died on test between the second and third feedings)					
3	0	0	90	66	0	0	26	19	815	34	25
6	249	182	587	428	357	260	180	131	1259	44	32
7	50	36	61	44	9	7	0	0	672	1	<1
8	15	11	69	50	20	15	67	49	928	0	0
10	0	0	92	67	30	22	0	0	276	0	0
Control (0mg DES in bait)											
2	420	——	1009	——	522	——	77	——	676	146	——
5	0	——	641	——	606	——	1327	——	1232	1218	——
9	80	——	993	——	1139	——	273	——	(Died February 12, 1974)		
11	0	——	62	——	1154	——	68	——	798	953	——

[a] Does 1-10 have the same numbers as in Table 1; doe 11 was new.

dates. These results are in agreement with metabolic studies using labeled DES. When 20mg of ^{14}C-labeled DES was fed to steers, a minimum of 85 percent was excreted as free DES in the feces (Aschbacher and Thacker 1974); and when 10mg of tritium-labeled DES was fed to steers, only 0.35 ppb was retained in the internal fat (Mitchell et al. 1959). However, it should be noted that there might be differences in DES storage between castrated and intact animals.

In the present study, the microencapsulated DES produced a physiological response causing decreased DES consumption below the threshold necessary to interfere with reproduction. Taste, smell, metabolic aversion, or a combination of these may have influenced DES intake. If the response was strictly an initial smell or taste aversion, rather than a learned metabolic aversion, one would expect uniform rejection over a period of time, but rejection was irregular and generally increased. However, zero DES consumption by some does on the third and fourth feeding during the breeding season indicates that smell may have served as a cue for rejection.

DES concentration appeared to affect the degree of taste or smell aversion as demonstrated by the switchback experiment. Consumption on the second feeding increased for those does placed on the lower concentrations and decreased for those placed on a higher concentration. Overall, DES consumption decreased as the concentration increased, but plateaued at the 750 and 1,000mg levels, with consumption decreasing over a period of time.

Metabolic aversion caused by nausea is one possibility. Humans ingesting DES may experience nausea sometimes severe enough to terminate treatment (Castrodale et al. 1942). However, no signs of nausea were observed in any deer following treatment. Another metabolic cause of rejection may be induced or natural titers that affect satiety. Bull et al. (1974) postulated that there is a feeding center in the lateral hypothalamus whose output of impulses is quantitatively attenuated by impulses of varying intensity from a ventromedial hypothalamic satiety center, and that estrogens inhibit one or more of these impulses influencing feed intake. This depressed intake is a rapid response that is reversible in a short time. Our pen studies have shown that pregnant does consume less feed during the winter months, suggesting a similar relationship between estrogen levels and consumption, since estrogen titers rise during pregnancy in ruminants (Catchpole 1964). Infusing estradiol into goats decreased their feed intake (Forbes 1971), and the greatest decrease occurred during estrus (Forbes and Rook 1970), suggesting that natural and induced estrogen levels have an additive effect on consumption. In the present study, as pregnancy progressed, the four control does showed no regular pattern in feed intake, but the five treated does ate progressively less after the second feeding, until their consumption totaled only 79 grams of feed (58mg of DES) during the fifth and final feeding in February. Harder and Peterle(1974) reported increased consumption during gestation when they offered DES to wild white-tailed deer in a shelled-corn bait; however, nontarget species were frequent visitors to the feeders, which may have accounted for the increased consumption. They reported a progressive aversion pattern similar to that in the present study when they offered wild deer tableted DES embedded in apple quarters.

If the rejection of DES by white-tailed deer is due to metabolic aversion, then perhaps even microencapsulated DES will not work as an antifertility agent where two or more feedings are required. If it is due to taste or smell, a different microencapsulation formulation might overcome the aversion problem.

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NASAL BOTS OF WHITE-TAILED DEER IN THE SOUTHEASTERN UNITED STATES[1]

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ABSTRACT

Nasal bots (*Cephenemyia* sp.) were found in 107 (4.4 percent) of 2,423 white-tailed deer (*Odocoileus virginianus*) examined from the following states: Alabama, Arkansas, Florida, Georgia, Kentucky, Louisiana, Maryland, Mississippi, North Carolina, South Carolina, Tennessee, Virginia, and West Virginia. Infected deer were not found in Alabama, Kentucky, and Tennessee. The parasite was most prevalent in the winter and summer. There were no significant differences in infestations between sexes or age groupings. The average infestation was 9 larvae per infested deer and only 5 deer harbored more than 30 larvae. *Cephenemyia* sp. did not appear to be a significant disease factor for white-tailed deer of the southeastern United States.

INTRODUCTION

The parasitic larvae of *Cephenemyia* sp. are frequently discovered by hunters when field dressing deer. The parasites, variously termed nasal, pharyngeal, head, or throat bots, are relatively large larvae of dipterous insects that require deer as hosts. Their presence frequently causes alarm that moves the sportsman to seek more information. These inquiries usually relate to the public health significance of *Cephenemyia*, life history, and pathogenicity for deer and domestic livestock. Data herein presented hopefully will serve as an aid to biologists in answering such questions.

MATERIALS AND METHODS

From February 1960 to December 1973, 2,423 white-tailed deer from 164 counties of 13 southeastern states were examined for *Cephenemyia* larvae. Of these, 1,417 were heads from hunter-killed deer; 905 were deer collected for herd health evaluations; and 101 were sick or dead deer submitted for diagnostic examinations.

Retropharyngeal pouches and oral cavities were examined by removal of the lower jaw. Nasal passages were exposed with the aid of a saw, bone forceps, and screwdriver. The esophagus,

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