EVALUATION OF CABERGOLINE AS A REPRODUCTIVE INHIBITOR FOR COYOTES (*CANIS LATRANS*)

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ABSTRACT: Cabergoline, a prolactin inhibitor, was evaluated on its potential use as a reproductive inhibitor for coyotes (*Canis latrans*). Groups consisting of six female coyotes were randomly assigned to three treatments and a control group. At 25 to 35 days after fertilization, coyotes were palpated to verify pregnancy status. If an animal was confirmed pregnant, it was dosed with 50 μg, 100 μg, or 250 μg of cabergoline, or a placebo for seven consecutive days on approximately day 40 days of gestation. Five animals dosed with 50 μg of cabergoline, three dosed with 100 μg, and three animals receiving placebo whelped; no animals treated with 250 μg whelped. No drop in serum progesterone or prolactin levels were observed for the 50 μg and 100 μg treated groups. However, progesterone levels declined below 2 ng/ml in animals treated with 250 μg. Prolactin and progesterone levels in the control group followed typical patterns observed in pregnant canines. This study suggests that cabergoline is a potential reproductive inhibitor in coyotes. Future studies should determine if the efficacy of cabergoline in terminating pregnancy in coyotes could be improved with higher doses and at earlier stages of gestation.

KEY WORDS: cabergoline, *Canis latrans*, coyote, fertility control, pregnancy, prolactin, progesterone

(March 6-9, 2000, San Diego, California)

Proc. 19th Vertebr. Pest Conf. (T.P. Salmon & A.C. Crabb, Eds.) Published at Univ. of Calif., Davis. 2000.

INTRODUCTION

Coyotes are one of the most widely distributed predators in North America (Bekoff and Wells 1986), and depredation by them on domestic livestock has been, and continues to be, a serious threat to producers in the western United States. A number of both wild and domestic species such as cattle, deer, and antelope can experience losses due to coyotes, but depredation on sheep is the most economically significant with yearly losses estimated at $17.7 million (U.S. Department of Agriculture 1995). Producers in the western U.S. suggest that problems by predators have risen in recent years and attribute this to an increase in predator numbers, less efficient methods for controlling predators, and ineffective management plans (American Sheep Industry Association 1999).

Losses incurred by coyotes on livestock have traditionally been managed by lethal means such as aerial hunting, trapping, and poisoning (Kirkpatrick and Turner 1985). The effectiveness of these techniques is variable and dependant on environmental conditions, terrain, coyote density, and the magnitude and nature of problems (Knowlton et al 1999). Recently, there has been increased public resistance and criticism of these traditional control methods. Thus, for the resolution of depredation problems to be successful, producers, and resource managers need to incorporate a variety of techniques that integrate social, ethical, and economical concerns, as well as the biology of the species, in the development of management strategies.

The use of fertility control as a means for reducing depredation problems has surfaced in recent years (DeLiberto et al. 1999). The use of this technique as a possible management tool represents an effective and humane alternative because it is non-lethal, predator social structure is preserved, territories are maintained, and non-target species can be protected. Another reason for the interest in reproductive inhibition is the breeding pair hypothesis developed by Till and Knowlton (1983). They indicated that many depredation problems caused by coyotes are from territorial adults provisioning for young. These adult animals switch from feeding principally on small and medium prey, to killing lambs. Till and Knowlton (1983) assumed that territorial breeders are the principal killers of livestock, and that depredations were linked to the presence of pups.

Experiments on fertility control have been conducted, but most compounds studied have had problems associated with an effective delivery system and a requirement for repeated doses (DeLiberto et al. 1999). Cabergoline is an ergot derivative that acts as a dopamine agonist, resulting in a prolonged prolactin-lowering effect. Cabergoline may have potential use in reproductive control because it has little or no side effects, is relatively species specific, is currently in an oral form, and has been shown to terminate pregnancy in the domestic dog (Onclin et al. 1993; Onclin and Verstegen 1997; Post et al 1988) and the red fox (*Vulpes vulpes*) (Marks et al. 2000).

The goal of our research was to evaluate the use of cabergoline as an effective reproductive inhibitor in coyotes. We examined three levels of cabergoline administered during the last trimester of pregnancy.

METHODS

Experiments were conducted at the USDA-Wildlife Services, National Wildlife Research Center' Predation Ecology and Behavior Station in Millville, Utah. This facility provided a unique opportunity to study

depredation processes in a controlled experimental setting, and provided an appropriate infrastructure in which to conduct our research project.

Twenty-four female coyotes were randomly assigned to three treatments and two control groups. Two of the treatment groups (n=6 females each) were housed in 0.1 ha pens and paired continuously with a male coyote. The third treatment group (n=6 females) was housed within individual kennels (4.3 m^2). Males were paired daily with these females continually, except during feeding, for approximately two months. Two control groups consisting of three animals each were used. One control group was housed in 0.1 ha pens with a male coyote and the other control group was housed in the kennels and paired daily with a male coyote.

Observations of mating behavior in the 0.1 ha pens were collected from sunrise to sunset seven days a week until all pairs completed breeding. These data were collected to evaluate mating behavior and document tie dates. At 28 days after the first observed tie, coyotes were palpated to evaluate pregnancy status. If an animal had a confirmed pregnancy, it was randomly placed into one of three treatment groups or a control group. Female coyotes were administered treatments for seven consecutive days beginning 40 days after the middle tie date. Treatments consisted of 50 μg (Group 1), 100 μg (Group 2), 250 μg (Group 3), and a placebo (Group 4). Doses of cabergoline were given in food. Animals housed in the larger pens received doses of an oily based formulation (Galastop: Centralvet, Milano) while those in kennels received a tablet formulation (0.5 mg tablet: Dostinex Tablets: Pharmacia & Upjohn).

Blood samples were obtained by cephalic veinipuncture. Blood was collected weekly until two weeks after the actual or predicted whelping date, and on day 3 and 7 during treatment. The blood samples were centrifuged for 20 min. at 1100 rpm within 2 hours after collection. The supernatant was isolated and stored at -20°C until analyzed.

Serum progesterone concentrations were run to examine treatment efficacy. Progesterone levels were estimated with a direct solid phase enzymeimmunoassay for the quantitative determination of progesterone using an ELISA kit validated for dog serum (American Laboratory Products Company, LTD., Windham, NH).

Serum prolactin concentrations were run only for pregnant females, based on whelping information and progesterone levels, in the 100 μg and 250 μg treatment groups, and for the control group. Prolactin levels were estimated with an enzyme immunometric assay designed for the quantitative measurement of prolactin in canine serum (the Milenia® Canine Prolactin, American Laboratory Products Company, LTD., Diagnostic Products Corporation, Germany).

RESULTS

Five of the treatment animals in Group 1 successfully whelped young. Litter sizes ranged from 1 to 5 pups with an average of 3.6 pups. Three of the females in Group 2 whelped young with litter sizes ranging from 2 to 6 pups with an average of 4.3 pups. None of the females in Group 3 whelped. One female in Group 3 had tary feces on day 4 of treatment, which may have been an indication of abortion. Three of the control animals whelped young, two from the 0.1 ha pens and one in the kennels. The two control animals housed in the larger pens had litter sizes of 4 and 5 pups and the control animal in the kennel whelped 4 pups, an average of 4.3.

For the control, 50 and 100 μg groups there appeared to be a normal decline in progesterone associated with the progression of gestation (Figures 1, 2, 3 and 4). No significant drop in progesterone associated with the treatment of cabergoline was detected. For the 250 μg treatment group, progesterone declined on day 3 and 7 of treatment to below the 2.0 ng/ml. Research suggests that 2.0 ng/ml is the threshold level of progesterone necessary to maintain pregnancy in the domestic dog (Onclin et al 1993; Concannon and Hansel 1977). This is likely the reason these females failed to whelp. Progesterone concentrations did rebound slightly by the post-treatment blood draw.

Prolactin concentrations for the both the 100 μg and 250 μg treatment groups decreased during treatment, but rebounded within a week post treatment. However, the 250 μg treatment group did not rebound as dramatically by the post-treatment blood draw. Prolactin concentrations for the control group were lower on day 3 and 7 post-treatment, but remained stable throughout the remainder of the study.

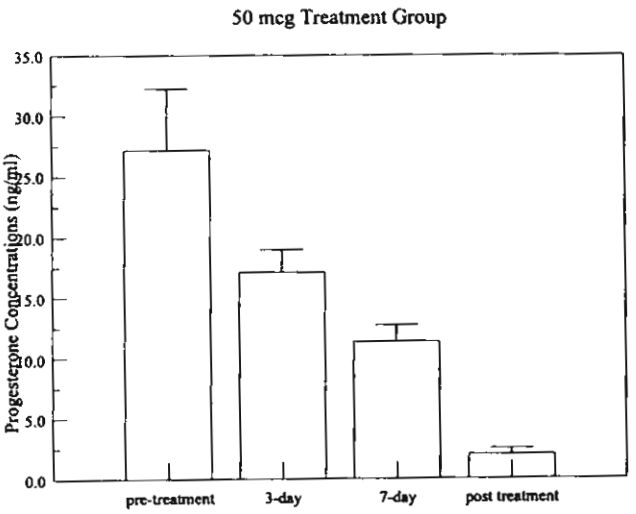

Figure 1. Mean serum progesterone concentrations a week pre-treatment, day 3 and 7 of treatment, and a week post-treatment of pregnant females (n=5) administered a seven consecutive day treatment of 50 mcg cabergoline starting on approximately day 40 of pregnancy.

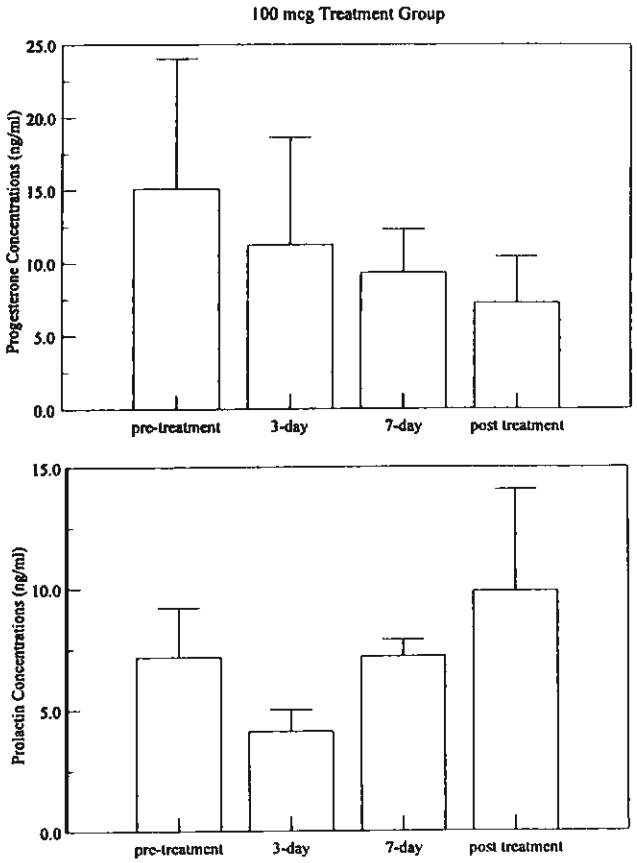

Figure 2. Mean serum progesterone and prolactin concentrations a week pre-treatment, day 3 and 7 of treatment, and a week post-treatment of pregnant females (n=5) administered a seven consecutive day treatment of 50 mcg cabergoline starting on approximately day 40 of pregnancy.

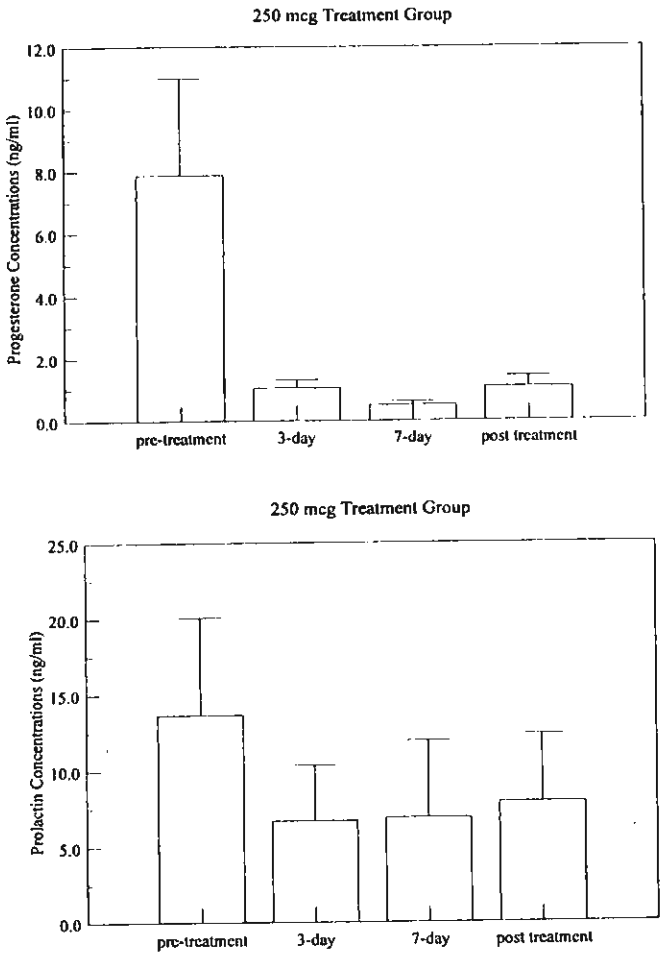

Figure 3. Mean serum progesterone and prolactin concentrations a week pre-treatment, day 3 and 7 of treatment, and a week post-treatment of pregnant females (n=3) administered a seven consecutive day treatment of 50 mcg cabergoline starting on approximately day 40 of pregnancy.

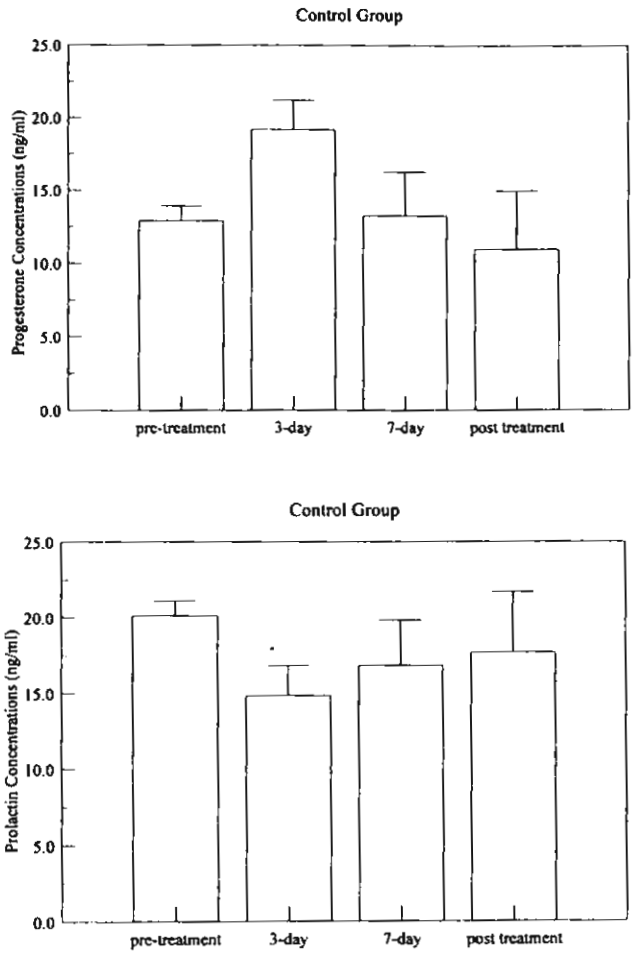

Figure 4. Mean serum progesterone and prolactin concentrations a week pre-treatment, day 3 and 7 of treatment, and a week post-treatment of pregnant females (n=3 for progesterone, n=2 for prolactin) administered a seven consecutive day treatment of 50 mcg cabergoline starting on approximately day 40 of pregnancy.

DISCUSSION

In mammals, progesterone is indispensable for the maintenance of pregnancy (Hodgen and Itskovitz 1988). Any prolonged depression of serum progesterone levels below 2 ng/ml will result in termination of pregnancy (Csapo et al. 1972; Concannon and Hansel 1977; Lombardi 1998). In most mammals, this threshold level of progesterone secretion is accomplished by the corpus luteum (notable exceptions include primates throughout pregnancy and sheep after the first trimester, which utilize the placental progesterone to maintain pregnancy). Therefore, maintenance of the corpus luteum is essential for pregnancy.

Maintenance of the corpus luteum in most mammals is accomplished by luteinizing hormone secreted by the anterior pituitary gland. However, in rodents, felines, and canines luteotrpohic support in mid to late pregnancy is provided by prolactin released from the anterior pituitary (Rothchild 1981; Concannon et al. 1987; Jochle 1997). Consequently, suppression of prolactin during pregnancy in these species will result in regression of the corpus luteum and, ultimately, termination of pregnancy.

Cabergoline suppressed prolactin levels in serum of pregnant coyotes in Groups 2 and 3. However, cabergoline only was successful in terminating pregnancy in Group 3 coyotes. These data suggest that the 50 μg dose considered adequate for treating domestic dogs was not effective in coyotes. Also, although the 100 μg dose resulted in a decrease in mean prolactin levels of coyotes in Group 2, it was apparently insufficient to cause mean serum progesterone levels to fall below the 2 ng/ml threshold for maintenance of pregnancy. Consequently, all animals in Group 2 whelped.

Evidence of a dose dependent response was reported in people (Mattei et al. 1988). They found that hyperprolactinemic patients that were administered a 0.6 mg dose of cabergoline had significant mean maximum decreases of prolactin over patients given a 0.3 mg dose. In addition, patients administered the 0.6 mg dose experienced continued depressed serum prolactin levels from 3 hours to 14 days, while the 0.3 mg dosage group had significant reductions in prolactin levels for only 3 hours to 5 days. Therefore, it is likely that our 50 μg and 100 μg doses were too low to be efficacious.

In Group 3, three animals refused to consume food that contained cabergoline after day 3 of treatment. Cabergoline may have caused the females to become ill and avoid the food and the associated dose. Such an aversion has been noted in domestic dogs (Jochle, pers. comm. 1999). However, these coyotes did not whelp, suggesting that the 250 μg treatment terminated pregnancy in these animals after only one to three doses.

In the red fox (Marks et al. 2000), cabergoline terminated pregnancy during mid gestation, but was unsuccessful later in gestation. Foxes treated twice with cabergoline prior to day 28 of gestation did not whelp. However, pregnancy was not reliably terminated in foxes treated after day 35 of gestation.

Timing of cabergoline administration may have played affected the results in our study. Determination of fertilization and pregnancy is difficult in coyotes. Although our experimental design consisted of initiating treatment on day 40 of gestation, we had no means of accurately determining fertilization. Coyote breeding records for the past 20 years at Predation Ecology and Behavior Center suggest that fertilization generally occurs between the first and middle observed tie dates. Thus, we conservatively estimated fertilization as the observed middle tie date to ensure that all animals had reached the stage of pregnancy whereby prolactin was leuteotrophic. This strategy undoubtably resulted in the initiation of treatment of some females after day 40 of pregnancy. Consequently, if there is a point in late gestation of coyotes when treatment with cabergoline becomes ineffective, it may explain why some of our treated animals whelped. However, this is unlikely because of our complete success in terminating pregnancy with 250 μg and the complete ineffectiveness with 50 and 100 μg of cabergoline.

We are continuing our research on cabergoline and its efficacy in coyotes. This year, female coyotes will be administered dosed with 150 μg for seven days, 250 μg for seven days, or 500 μg for one day. The latter dose of cabergoline was selected to evaluate its potential if only one dose could be delivered in the field. Marks et al. (1996) used fox baits around dens that contained 170 μg of cabergoline. Dens were each treated once in August and once in September resulting in a significantly lower incidence of cubs.

In addition, we will be using a relaxin pregnancy test (Qualitative relaxin assay, Reprocheck, ELISA, Synbiotics Corp., San Diego, CA), as well as palpation to verify pregnancy status and to refine our estimate of fertilization date. The pregnancy test will be run 21 days after the first observed tie and will also be run daily during the treatment regime to determine if pregnancy has been terminated. Additionally, serum progesterone levels will also be used to determine the point during treatment that pregnancy was terminated. Such data will provide valuable information on the number of cabergoline doses required to terminate a pregnancy and the period of gestation during which treatments are effective.

CONCLUSIONS

Producers consider predation a major factor in the decline of the sheep industry (Buys 1975; Gee et al. 1977). Currently, the most effective means of resolving depredation problems is through lethal control methods. Even though these strategies minimize losses to producers, coyotes can still cause significant economic hardship. Additionally, as human population expands into wildlife habitat, lethal control options become limited and controversial. Therefore, there is a need to develop non-lethal control strategies that can be integrated into wildlife damage management programs.

Development of cabergoline as a reproductive inhibitor may further decrease losses due to coyote depredation, making sheep producers throughout the United States more profitable and competitive. Initial studies on cabergoline and other reproductive inhibitors are encouraging. Continued research on alternative control methods such as cabergoline are critical for the survival of the sheep industry.

ACKNOWLEDGMENTS

This project would not have been possible without the dedication of Jessica Tegt, Cody Nielsen, Krista Wenning, Debra Carlson, and Sarah Rose who spent many long days handling, helping with blood draws, and caring for coyotes. We also are indebted to Dr. Wolfgang Jöchle for all his helpful suggestions on study design and use of cabergoline. And finally, many thanks to Terrie Wierenga and the USDA-ARS Poisonous Plant Lab for conducting prolactin and progesterone assays.

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