The use of DiazaConTM to limit fertility in grey squirrels

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Abstract

The grey squirrel is an invasive alien species introduced to Great Britain in the late 19th Century and to Northern Italy during the early 20th Century. As well as displacing the native European red squirrel, grey squirrels cause significant impact to trees and woodlands through bark-stripping activity, and are implicated in the decline of woodland bird populations. In Britain, eradication is no longer an option at a regional scale, but fertility control offers a possible approach to minimise negative impacts. The cholesterol mimic DiazaConTM has been successfully used to inhibit reproduction in some species as it reduces cholesterol available for steroid reproductive hormone synthesis. Results are presented from enclosure studies investigating whether DiazaConTM would reduce cholesterol levels enough and for a sufficient period to reduce fertility in female grey squirrels.

Keywords: cholesterol, contraception, DiazaConTM, fertility control, grey squirrel, 20, 25-diazacholesterol dihydrochloride

Introduction

Since its introduction to Britain in the late 19^{th} Century the grey squirrel (*Sciurus carolinensis*) has been of increasing concern to woodland and conservation managers due both to seasonal bark stripping activity and its impact on woodland biodiversity, particularly the native red squirrel (*S. vulgaris*). Estimates of annual costs to the British timber industry of damage to trees through bark stripping by grey squirrels are £3.40 (€3.84) per ha of vulnerable conifers, and £5 (€5.65) per ha for the three most vulnerable broadleaf timber tree species; sycamore (*Acer pseudoplanatus* L.), beech (*Fagus sylvatica* L.) and oak (*Quercus sp.*) (Williams et al., 2010). As tree damage is believed to be triggered in part by an increase in juveniles in the population during late spring, following breeding in January/February, fertility control offers a non-lethal approach to minimising impacts.

DiazaConTM is a cholesterol mimic that inhibits cholesterol production and blocks steroid hormone production and thus can reduce fertility in both males and females. A reduction in cholesterol of approximately 40% will reduce reproduction in many bird species and rats (Yoder et al., 2005). An initial study (Yoder et al., in press) showed that DiazaConTM treatment over 8 days reduced cholesterol ≥40% for 2 months in grey squirrels. As grey squirrels will not breed in cages in captivity, enclosure studies were used to investigate whether DiazaConTM would reduce cholesterol levels enough and for a sufficient period to reduce fertility in female grey squirrels.

Materials and methods

DiazaConTM was provided by the Avitrol Corporation (Tulsa, OK) via National Wildlife Research Center (NWRC). Enclosure studies were carried out in 2007 and 2009. In each year 32 female grey squirrels were caught from the wild and held in individual cages to acclimatize before the treatment feeding period. Sixteen females were allocated each to treatment or control. For both studies DiazaConTM was presented on whole grain wheat coated with corn oil and sugar to achieve a dose of 55mg/kg bodyweight (assuming 20g bait was eaten each day over a 10 day feeding period). Control animals received untreated bait, and daily bait intake was monitored for both control and treated groups.

After treatment the females were released into enclosures with males and monitored monthly thereafter for reproductive success. Treatment females were allocated six each to 2 large and two each to 2 small enclosures, based upon mean daily DiazaConTM dose intake. The same numbers of control females were allocated to the enclosures based upon pre-feeding period bodyweight, with animals of similar weight allocated to each enclosure where possible. Before the feeding period all females were weighed, a

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vaginal smear taken to check the stage of the oestrous cycle and a blood sample was taken for cholesterol and desmosterol testing at NWRC and for progesterone assay. At each recapture females were visually assessed for lactation (raised, hairless nipples), weighed, and blood samples and vaginal smears taken. DNA testing was used to confirm relatedness of juveniles/litters found to females.

Results

In 2007 the DiazaCon[™] dose levels achieved ranged from 8.9 to 25.7 mg/kg (mean 16.6 mg/kg) (n=16). Plasma cholesterol reduced in control and treated groups post treatment, then returned to above pretreatment levels for the control group and remained at this level until the end of the study. For treatment females plasma cholesterol levels remained below 50% of that of the control group for 68 days. By the end of the 4 month study period there was no significant difference between treatment and control groups. Desmosterol levels were correlated with dose of DiazaCon[™]. As only 2 control females had litters, both with 2 kittens, results in terms of effect of DiazaCon[™] on reproduction were inconclusive.

In 2009 a greater range of dose intake was achieved with a mean intake of 28.7 mg/kg and range 9.0-66.2 mg/kg (n=13). The target dose of 55 mg/kg was achieved for 2 females. Cholesterol response showed a significant increase in cholesterol for control animals (n=16) on release into the enclosures, as observed in 2007. The decline in cholesterol post feeding was less pronounced than observed in 2007. A significant reduction in cholesterol was observed for the treatment group up to 4 months post treatment with treatment cholesterol at only 66% of the control group at this time. Mean desmosterol response reflected the observed decrease in cholesterol. Although more pregnancies were observed during the 6 months that females were held in the enclosures only one control female bred soon after release into the enclosures and the rest bred between May and late June. Nine control females bred (7 litters found) and 5 treatment females bred (3 litters found). One treatment female bred after the end of the study.

Discussion

These studies demonstrate that DiazaConTM fed over a 10 day period is effective at reducing cholesterol levels by \geq 40% in female grey squirrels for up to 4 months. A lack of information on seasonal changes in plasma cholesterol for grey squirrels limits interpretation of the results. As most females bred after cholesterol levels in treated females had returned to pre-treatment levels the results are inconclusive in terms of effect of DiazaConTM on fertility in grey squirrels.

References

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