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Synchrony of onset and cessation of breeding activity in brushtail possums *(Trichosurus vulpecula)* in coastal Otago, New Zealand

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ABSTRACT

The onset of breeding was monitored in 117 female brushtail possums trapped at a single location between December and May, with reproductive status being determined at the time of capture. The cessation of breeding activity was monitored in captive animals in which reproductive status was determined in October (N=38), November (N=39) and January (N=30), by laparoscopic observation of their reproductive tracts. Comparison was made between animals that had pouch young removed (RPY) 7-9 days previously, and possums that had not had pouch young present for at least 40 days.

Only 1/57 possums trapped over the period from 1 December to 31 March had recently bred. In contrast, 49/60 (82%) possums captured between 1 April and 6 May, were either pregnant or had new-born pouch young. In spring (mid-October) 45% of possums continued to cycle, only 13% were cycling in late November and all were anoestrous in January. There were no relationships between liveweight or RPY and reproductive status.

Keywords: brushtail possum; seasonal breeding; anoestrus; laparoscopy.

INTRODUCTION

The brushtail possum *(Trichosurus vulpecula)* is an arboreal marsupial that is seasonally polyoestrous (Tyndale-Biscoe, 1955; Pilton and Sharman, 1962). The female is monovular, has an oestrous cycle of 24-26 days and gestation length of 17-18 days (Hartman, 1917; Tyndale-Biscoe, 1955; Curlewis and Stone, 1986). After birth, the single-born young enters the pouch, attaches permanently to the teat for the first 70-90 days of life and emerges from the pouch for intervals of increasing duration from Day 120-140. It may leave the pouch permanently from Day 150 and is completely weaned at about 240 days of age (Lyne and Verhagen, 1957; Crawley, 1973).

Although births have been recorded in every month of the year, within a given population breeding is highly seasonal (Kerle, 1984; Gemmell, 1987). The main breeding season is in autumn (Smith *et al.,* 1969: Crawley, 1973) when 80 -100% of adult females produce young (Cowan, 1990). The onset of breeding in autumn is reported to vary from early April to early June, and to be earlier in times of good nutrition when female bodyweights are high (Cowan, 1990). The relationship between body condition and ecological performance suggests that reproduction is regulated by food supplies (Humphreys *et al.,* 1984).

A second, more variable breeding season involving fewer females occurs in spring. The percentage of females that breed in spring varies widely between locations. Possums in exotic forest or pasture/scrub locations, breed in spring more often than those in established native forest, with up to 33% of females in such locations breeding at this time of the year (Cowan, 1990). In some locations, less than 1.0% of females produce young in spring (Green, 1984; Cowan, 1990). It is suggested that level of nutrition is the major influence on the occurrence of spring breeding and that double-breeding (producing young in both autumn and spring) occurs only if autumn breeding was early, and then only in females of above average body condition (Cowan, 1990).

The mechanisms controlling the onset and cessation of breeding activity in possums are poorly understood, but it has been demonstrated that photoperiod plays a role in the timing of the breeding season (Gemmell, 1990; Gemmell *et al.,* 1993; Gemmell and Sernia, 1995). Exposure to an artificial short-day photoperiod, or treatment with melatonin implants, will induce ovulation and result in pregnancies in summer when possums are normally anoestrous (Gemmell, 1987; Gemmell, 1990). Cowan (1990) has demonstrated that the incidence of breeding in the spring is not related to latitude, at least within New Zealand.

The possum is a pest of considerable economic importance in New Zealand. It causes extensive damage to native and exotic forests, and is an important vector of several diseases. Current methods of controlling possums have little effect on reducing the national population. Research efforts are now aimed at suppressing reproduction in this species, and attempts are being made to identify reproductive processes that may be susceptible to disruption. There is a need to define more accurately the onset and duration of the breeding season within a population of possums in a given location. A further requirement is an understanding of the influence that body condition and the presence and subsequent removal of pouch young has on breeding activity. In this study, we have determined the synchrony of onset of breeding activity within a single wild population of brushtail possums. We have also used a population of captive animals to characterise the cessation of breeding activity in possums maintained under conditions of good nutrition, to investigate the relationship between liveweight and breeding activity.

MATERIALS AND METHODS

Capture

Possums were trapped at sites in coastal Otago, all within 15 kilometres of Invermay Agricultural Centre (latitude 45° 14' S). All locations were at the interface between bush and/or *pinus radiata* forestry blocks and pasture. Each location included steep gullies containing regenerating bush and scrub (major species present were manuka, kanuka and gorse), that were adjacent to clover/ryegrass pastures grazed by sheep, deer and/or cattle. Possums were captured using wire-sided box-traps, baited with apple. Traps were checked each morning and animals were transferred to hessian sacks for transportation to Invermay.

Onset of Breeding Activity.

Possums were collected from one site over a six-month interval (early-December to mid-May 1996), which included the putative transition period from the non-breeding to the breeding season. A total of 117 adult females (mean liveweight at capture 2.65 ± 0.05 kg) were trapped within a 0.5 km radius. All captured animals (male, female or juvenile) were removed from the site of capture.

Reproductive status of females at the time of capture was classified as being either; (a) empty - non-pregnant, mammary glands regressed and no young present; (b) pregnant - pouch young born in captivity within 18 days of capture; (c) with new pouch young - pouch young estimated to be less than 40 days of age; (d) with old pouch young - pouch young estimated to be over 100 days of age, or (e) with back-rider juvenile present. Pouch young in categories c, d and e, would have been born in the current autumn, the previous spring or the previous autumn, respectively.

Cessation of Breeding Activity.

Adult female possums captured over the period from April to November, were held in captivity at the possum facility at AgResearch, Invermay. All animals were group-housed (6-10 possums/pen) under conditions of natural photoperiod, and were fed a mixed diet of fresh fruit, bread and cereal-based pellets. Fresh branches of *pinus radiata* were included in all pens as a source of browse, and fresh water was always available. Under these housing conditions, animals do not lose weight or exhibit any clinical signs of post-capture stress (McLeod *et al.,* 1997). All groups of female possums were housed in the absence of males.

Reproductive status was assessed three times during spring and summer (mid-October, late-November and mid-January), by laparoscopic observation of the reproductive tract (see below). At each of these observations, comparison of reproductive status was made between animals that had had their pouch young removed 7-9 days previously, with that of possums that had not had pouch young present for at least 40 days (range 41-90 days).

Laparoscopy.

Reproductive status of female possums was based on the condition of the reproductive tract according to the criteria of Crawford *et al.,* (1997a). In brief, animals that have pouch young present or that are seasonally anoestrus, have a small and undeveloped vaginal cul-de-sac and uteri. Reproductive tract morphology does not change following RPY in anoestrous animals, and only small antral follicles are present on the ovaries. In contrast, in cyclic animals the cul-de-sac and uteri increase in size following RPY, to reach a maximal size after 8-12 and 15-20 days, respectively. In addition, there is an increase in the size and number of large antral follicles present on the ovaries.

The laparoscopic method has been described in detail by Crawford *et al*., (1997a). Briefly, anaesthesia was induced and maintained by halothane inhalation (Fluothane; ICI New Zealand Ltd, Lower Hutt, New Zealand). Local anaesthetic (lignocaine hydrochloride BP 2% w/v; Techvet Laboratories Ltd, Auckland, New Zealand) was administered subcutaneously at the sites of endoscope (4 mm diameter: Karl Storz GmbH & Co.Tuttlingen, Germany) and manipulating probe insertion. The diameter of the open jaws of the manipulating probe (6 mm) was used to estimate sizes of reproductive tract organs and diameter of antral follicles. Morphology of the vaginal cul-de-sac and uteri was assessed for changes in size, degree of vascularisation, colour and swelling. The diameter and number of antral follicles present on the surface of the ovaries was also estimated.

Age of pouch young was estimated at the time of capture of adult females (onset of breeding activity) or at the time of RPY (cessation of breeding activity). Estimated age was taken as the mean age from crown-rump and head length measurements (Tyndale-Biscoe, 1955).

Capture and housing of possums, and all experimental procedures performed, had been approved by the AgResearch Animals Ethics Committee according to the Animals Protection (Codes of Ethical Conduct) Regulations, 1987.

Analysis of data

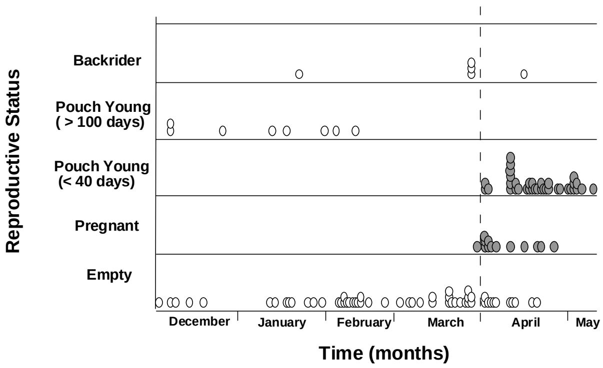
Differences between times of year in the number of animals that were cyclic (oestrous) or non-cyclic (seasonally anoestrous) were compared by Chi-squared analysis. Differences between groups in liveweight were compared by one-way ANOVA.

RESULTS

Onset of breeding activity

Over the 5-month capture period between December and May, a total of 117 adult female possums were collected. The reproductive status of individual females at the time of capture is shown in Figure 1. Over the 17-week period from 1 December to 31 March, a total of 57 adult female possums were caught. Only one of these animals (pregnant at the time of capture) had recently bred. Another eight females (14%) had pouch young aged over 100 days, 4 (7%) were carrying backriders and 44 (77%) were empty. Of the 60 females trapped over the 5 week period from 1 April to 6 May, 80% were either pregnant (n=12) or had new-born pouch young (n=36) and only 18% (n=11) were empty. One further animal was carrying a backrider.

FIGURE 1: Reproductive status at the time of capture of individual female brushtail possums trapped between December and May at a single location in coastal Otago. Those animals that had successfully recently bred, are shown by the shaded symbols.



Mean liveweight did not differ between animals that were empty (2.59 ± 0.06 kg, N=49), pregnant (2.63 ± 0.13 kg, N=14), with new-born pouch young (2.68 ± 0.06 kg, N=35) or carrying backriders (2.54 ± 0.15 kg, N=5). The mean bodyweight of those animals with older pouch young was significantly heavier than that of all other groups (3.24 ± 0.09 kg, N=8, $P < 0.05$).

Onset of seasonal anoestrum

The numbers of possums that were classified as anoestrous or as showing cyclic activity at each of the three laparoscopic observations are shown in Table 1. There were no significant differences in mean bodyweight between these groups of animals, either at the time of capture or at the time of laparoscopy. The percentage of possums that were anoestrus at the time of laparoscopy changed significantly with time of year, being 55.3, 87.2 and 100.0% in mid-October, late November and mid-January, respectively ($P < 0.01$).

TABLE 1: Proportion of female brushtail possums classified as cyclic or seasonally anoestrous in October, November and January, their pouch young status and mean liveweight at times of capture and of laparoscopy.

	Percent (n)	RPY	Liveweight (kg) [1]At capture	At laparoscopy
mid-October				
Anoestrous	55.3 (21)	53.3 (8)	2.90 ± 0.11	2.95 ± 0.09
Cyclic	44.7 (17)	46.7 (7)	3.13 ± 0.13	3.16 ± 0.14
Total	(38)	15	3.00 ± 0.09	3.04 ± 0.06
late-November				
Anoestrous	87.2 (34)	100 (10)	2.89 ± 0.09	2.87 ± 0.06
Cyclic	12.8 (5)	0 (0)	3.02 ± 0.22	3.09 ± 0.09
Total	(39)	10	2.91 ± 0.09	2.90 ± 0.06
December				
Anoestrous	100 (30)	100 (5)	2.96 ± 0.09	3.02 ± 0.10
Cyclic	0 (0)	0 (0)	-	-
Total	(30)	5	2.96 ± 0.09	3.02 ± 0.10

[1] Number of animal from which pouch young were removed 7-9 days prior to laparoscopy

Mean liveweight of cyclic females was not significantly different from that of anoestrous animals, either at the time of capture or at laparoscopy. The mean estimated age of pouch young removed prior to each laparoscopy did not differ significantly between times of the year, or between females subsequently classified as cyclic or anoestrous. Removal of pouch young did not influence the incidence of cyclic activity at any of the laparoscopy dates.

DISCUSSION

In this study, the onset of breeding activity in female possums was highly synchronous between individual wild-caught animals trapped at a single location. None of the possums captured between early December to late March had recently bred, whereas 80% of adult females captured over the succeeding 5 weeks had. This confirms the findings of Jolly *et al.,* (1995), that within a given population, the onset of seasonal breeding is highly synchronised. The degree of variation in the timing and synchrony of breeding activity in the same population between years, or between different locations in the same region and in the same year, remains to be determined.

The present investigation offered a novel approach for determining the cessation of the breeding season. This was monitored in a captive population of possums, where all animals had access to a high level of nutrition and changes in liveweight in individual animals could be determined. In addition, ovarian activity was the parameter monitored, rather than breeding success (presence of pouch young). Therefore, reproductive status was not confounded by possible seasonal effects of male reproduction or by fertilisation/conception rates. Furthermore, laparoscopy which has seldom been used to monitor reproduction in possums, provides a very accurate method of assessing reproductive status in possums (Crawford *et al.*, 1997a).

The proportion of animals that remained reproductively active (exhibiting oestrous cycles) in spring (45%, mid-October) was much higher than that previously reported for animals rearing pouch young at that time of year. The highest estimates recorded in similar studies of possum populations in both North and South Islands (based on the presence of pouch young) seldom exceeded 30% (Cowan, 1990). This suggests that either there is wide variation in the proportion of females breeding in spring between different regions or between successive years, or that a high proportion (up to 40%) of cyclic females may fail to breed successfully at this time of year. This could be due to a failure to mate, the failure of fertilisation/conception or to high embryonic or new-born mortality rates. In some regions of New Zealand, < 1% of females breed in spring (Cowan 1990). It would be of interest to determine the proportion of females that still undergo oestrous cycles in spring in these populations.

Previous reports, particularly those relating to possums breeding in spring, suggest a strong correlation between body condition and breeding success. The present study has failed to identify any relationship between liveweight and reproductive status, either at the onset or cessation of seasonal breeding in adult female possums. Furthermore, the presence or absence of oestrous cyclicity in spring, could not be related to dynamic changes in liveweight (recorded over the period of captivity). Therefore, reproductive status is not likely to be associated with weight gain or weight loss. It should be noted however, that possums held in this colony invariably do not lose weight (McLeod *et al.,* 1997).

It is well documented that suckling by pouch young suppresses ovarian cyclicity in early and mid breeding season (Pilton and Sharman, 1962). However, in the present study the incidence of oestrous cyclicity in spring was not affected by RPY (see Table 1). This indicates that the suppressive effects of suckling on reproduction, and the abrupt removal of these suppressive effects, had little influence on reproductive status at this time of year.

In the present study, all females were housed in the absence of males. We have recently shown that presence of males can increase the incidence of ovulation following RPY by up to 80%, at least during the main (autumn) breeding season (Crawford *et al.*, 1997b). Presence of males can also increase the incidence of ovulation following RPY in spring, but with a lower success rate than achieved in autumn (JL Crawford and BJ McLeod - unpublished). Therefore, it is possible that an even higher proportion of females would have continued to exhibit oestrous cycles in the present study, if they had been housed with males.

The rapid decline in the proportion of animals showing oestrous cyclicity in late spring November) and the absence of cyclic animals in summer, suggest the presence of an overriding mechanism such as photoperiod, suppressing ovarian cyclicity. However, although Gemmell and coworkers have demonstrated a strong correlation between photoperiod and the onset of breeding (Gemmell, 1990; Gemmell and Sernia 1992) they have been unable to demonstrate a photoperiodic influence on the cessation of seasonal breeding (Gemmell and Sernia 1995). Furthermore, the present study has shown that within a captive group of possums maintained under the same photoperiodic and nutritional conditions, some may continue to cycle when others have become anoestrous. The mechanisms controlling this phenomenon remain to be determined.

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REFERENCES

Cowan, P. E. 1990. Brushtail possum. pp 67-98 *in*: C M King (ed) The Handbook of New Zealand Mammals. Oxford University Press, Oxford.

Curlewis, J.D.; Stone, G. M. 1986. Reproduction in captive female brushtailed possums *Trichosurus vulpecula. Australian Journal of Zoology* **34:** 47-52.

Crawford, J.L.; Shackell, G. H.; Thompson, E. G.; McLeod, B.J.; Hurst, P. R. 1997a. Preovulatory follicle development and ovulation in the female brushtail possum *(Trichosurus vulpecula)* monitored by repeated laparoscopy. *Journal of Reproduction and Fertility* **110:** 361-370.

Crawford, J.L.; McLeod, B.J.; Thompson, E. G.; Hurst, P. R. 1997b. Presence of males effects the incidence of ovulation following pouch young removal in brushtail possums*(Trichosurus vulpecula)* . *Animal Reproduction Science* **51:** 45-55.

Crawley, M.C. 1973. A live-trapping study of Australian brush-tailed possums, *Trichosurus vulpecula* (Kerr), in the Orongaronga Valley, Wellington, New Zealand. *Australian Journal of Zoology* **21:** 75-90.

Gemmell, R.T. 1987. Effect of melatonin and removal of pouch young on the seasonality of births in the marsupial*Trichosurus vulpecula. Journal of Reproduction and Fertility* **80:** 301-307.

Gemmell, R.T. 1990. Influence of daylength on the initiation of the breeding season of the marsupial possum *Trichosurus vulpecula. Journal of Reproduction and Fertility* **88:** 605-609.

Gemmell, R.T.; Sernia, C. 1992. The role of daylength on the initiation of the breeding season of the brushtail possum *(Trichosurus vulpecula).Journal of Reproduction and Fertility* **95:** 701-708.

Gemmell, R.T.; Gepon, G., Sernia, C. 1993. Effect of photoperiod on the breeding season of the marsupial possum *Trichosurus vulpecula). Journal of Reproduction and Fertility* **98:** 515-520.

Gemmell, R.T.; Sernia, C. 1995. Effect of changing from a short-day to long-day photoperiod on the breeding season of the brushtail possum *(Trichosurus vulpecula). Journal of Experimental Zoology* **273:** 242-246.

Green, W. Q. 1984. A review of the ecological studies relevant to management of the common brushtail possum. pp 483-499. *in:* Smith, A. and Hume, I. *ed* Possums and Gliders, Surrey Beatty and Sons, Chipping Norton, New South Wales, Australia.

Hartman, C. 1917. The oestrous cycle in the opossum. *Biological Bulletin* **37:** 353-321.

Humphreys, W.F.; Bradley, A.J.; How, R.A.; Barnett, J.L. 1984. Indices of conditions of phalanger populations: a review. pp 59-77. *in:* Smith, A., Hume, I. *ed.* Possums and Gliders, Surrey Beatty and Sons, Chipping Norton, New South Wales, Australia.

Jolly, S.E.; Scobie, S.; Coleman, M.C. 1995. Breeding capacity of female brushtail possums *(Trichosurus vulpecula)* in captivity. *New Zealand Journal of Zoology* **22:** 325-330.

Kerle, J. A. 1984. Variation in the ecology of *Trichosurus* : Its adaptive significance. pp 483-499. *in:* Smith, A. P., Hume, I.D. *ed. Possums and Gliders.* Sydney, Australian Mammal Society.

Lyne, A.G; Verhagen, A.M.W. 1957. Growth of the marsupial *Trichosurus vulpecula* and a comparison with some higher mammals. *Growth* **21:** 167-195.

McLeod, B.J.; Thompson, E. G.; Crawford, J. L.; Shackell, G. H. 1997. Successful group-housing of wild-caught brushtail possums *(Trichosurus vulpecula). Animal Welfare* **6:** 67-76.

Pilton, P.E.; Sharman, G.B. 1962. Reproduction in the marsupial *Trichosurus vulpecula*. *Journal of Endocrinology* **25:** 119-136.

Smith, M.J.; Brown, B.K.; Frith, H.J. 1969. Breeding of the brushtailed possum, *Trichosurus vulpecula* Kerr in New South Wales. *CSIRO Wildlife Research* **14:** 181-193.

Tyndale-Biscoe, C. H. 1955. Observations on the reproduction and ecology of the brush-tailed possum *Trichosurus vulpecula* Kerr (Marsupialia) in New Zealand. *Australian Journal of Zoology* **3:** 162-184.