

## Reproduction of the Red-Bellied Pademelon, *Thylogale billardierii* (Marsupialia)

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### Abstract

*Thylogale billardierii*, which is abundant in Tasmania, is a seasonal breeder with most births in the months April, May and June. Parturition is followed by mating, and the zygote so produced remains dormant until either suckling becomes intermittent near the end of pouch life or the young is lost. The mean length of the oestrous cycle was determined at 30.3 days, not significantly longer than the duration of gestation (30.2 days). Removal of pouch young results in the birth of a new young 28.7 days later. Removal of the corpus luteum results in oestrus 11 days later. Pouch life is 202 days, and vacation of the pouch by the young coincides precisely with parturition and post-partum mating. The young mature at about 14-15 months.

### Introduction

The red-bellied pademelon, *Thylogale billardierii* (Desmarest), was previously distributed over southern south-east Australia (Calaby 1971) but is now restricted to Tasmania and some of the larger Bass Strait islands. Little is known about the physiology of this medium-sized macropodid marsupial, even though it is extremely abundant in the wetter areas of Tasmania. Sharman (1961) examined the chromosomes of this species and Kirsch (1977) carried out serological studies. Sharman and Berger (1969) suggested that this species exhibits embryonic diapause, and Morton and Burton (1973) described some reproductive behaviour and made an estimate of the duration of gestation.

The present study of reproduction of the pademelon provides data on the breeding season, oestrous cycle, gestation and attainment of maturity. Data on pouch young growth are presented separately (Rose and McCartney 1982).

### Materials and Method

#### *Animals and their Maintenance*

Most of the data was collected from 1977 to 1980. Five pademelons were obtained from the Jock Marshall Reserve at Monash University, through the courtesy of Dr John Nelson. Sixteen live animals were snared or netted in various localities in eastern Tasmania. The captive population comprised 13 females and eight males but two males died soon after capture. Six of the females were mature and another three reached adult weight during the study. A total of 140 pouch young were obtained from eastern and south-eastern Tasmania. To these observations were added data from 102 females with pouch young, from south-west Tasmania, provided by Mollison (personal communication); these data were in the form of head length, weight of the pouch young, and date.

The animals were housed in an enclosed area of about 0.5 ha of quasi-natural bushland at the University of Tasmania in Hobart. They fed on pasture and shrubs supplemented with stock pellets and oats, as well as bread, apples and lucerne hay. A long hand-held net was used to capture the animals within the enclosure. Pademelons were identified by the use of ear-tattoos.

#### Techniques

Pademelons were judged to have attained maturity if a female produced a young or a male had sperm present in his urine.

Vaginal smears were obtained daily from the posterior vaginal sinus of five females, stained with Shorr's (1941) stain, and evaluated by means of the criteria adopted by Peters and Rose (1979). The smears were observed throughout a total of 10 oestrous cycles. Mating was observed during daylight on seven occasions, and the period between observed matings were used to calculate the length of the oestrous cycles. The length of gestation was obtained from data from each of five females, by noting the length of time between mating and birth. The time to birth after the removal of pouch young was obtained in six cases. After parturition animals were examined for the presence of a copulatory plug.

Seasonality of breeding was examined by calculating the dates of parturition for pouch young obtained from the wild (this study) and from Mollison's unpublished data. Dates of parturition were calculated by means of the growth curves which we have constructed (Rose and McCartney 1982).

The corpus luteum was removed from two females (with pouch young) under Halothane anaesthetic. Their young were then removed and the animals maintained in a warm room. Vaginal smears were taken daily until oestrus.

### Observations

#### Males

*Age and weight at maturity.* In captivity four males attained maturity at weights between 3.9 and 4.75 kg. The earliest age at which a male matured was 14 months. Data from Mollison show that there is a marked increase in the ratio of testes to body weight at 4 kg. Males are fertile soon after spermatorrhoea first occurs.

Table 1. Monthly summary of reproduction in captivity

Information presented is the monthly distribution of the total number of observations from nine females over the period 1977-80

	J	F	M	A	M	J	J	A	S	O	N	D
Birth	2	2	—	1	2	2	1	3	2	—	—	1
Mating	2	2	—	2	1	2	2	3	1	2	4	2
Oestrus	2	2	1	2	1	3	2	3	3	3	5	2

*Behaviour.* As a prelude to mating, the male trailed the female for some time emitting a clucking sound; this was followed by a period during which he sniffed the pouch and genital region. During mounting the male grasped the female around the flanks. Powerful thrusts, which almost lifted the smaller female off the ground, accompanied intromission. Morton and Burton (1973) give a similar account of mating. In captivity, males mated in most months of the year (Table 1) but the situation in the wild is unknown.

#### Females

Data on female reproduction are summarized in Table 2.

*Sexual maturity.* Three females reared in captivity reached a weight of 4.5 kg or more. Only one of these gave birth (at 14 months), her weight at this age being

3.5 kg. Of the other two, one was released at the age of 19 months and weight of 4.5 kg, without having produced young, and the other is still in captivity and nulliparous (age over 30 months, weight 4.7 kg). Our field data and those from Mollison indicate that some females are mature at a weight of 3.5 kg.

*Oestrous cycles.* The mean oestrous cycle length was 30.3 days (Table 2). Three stages of the oestrous cycle were apparent from the smears: pro-oestrus, oestrus and post-oestrus. During pro-oestrus, which lasted 5–8 days, large numbers of partly cornified cells were present. There was a gradual increase in the thickness of the smear and the proportion of leucocytes steadily decreased. Oestrus lasted less than 2 days, and was characterized by the absence of leucocytes and the presence of a majority of epithelial cells with pycnotic nuclei. If mating had occurred, the smear contained many sperm and structures usually referred to as prostatic bodies. For 3–5 days after oestrus the smear became thicker, which indicated that large numbers of angular enucleate epithelial cells had sloughed off. Shortly thereafter, leucocytes reappeared and the number of pycnotic epithelial cells decreased. During the remainder of the cycle, the proportions of the cellular constituents varied erratically, and the smear frequently contained strands of mucus and parabasal bodies.

Table 2. Duration in days of various parameters of reproduction

	<i>N</i>	Mean	Range
Oestrous cycle	12	30.3	30–32
Gestation length	5	30.2	30–31
Removal of pouch young to birth	6	28.7	27–31
Removal of pouch young to oestrus	6	29.2	27–31
Corpus luteotomy to oestrus	2	11	11
Pouch life	6	202	196–212

*Mating.* Mating, as determined by visual observations, or as inferred from either the presence of a white gelatinous mass around the opening of the urogenital sinus or the presence of sperm in the vaginal smear, was usually limited to a period of less than 24 h, but occurred over a longer period in two cases.

*Gestation.* The length of gestation was obtained for five females: 30, 30, 30, 30 and 31 days (Table 2). Birth was always followed within 24 h by a post-partum oestrus, so that parturition and mating usually occurred during the same night. Removal of the pouch young initiated oestrous cycles and development of the diapausing blastocysts, and in six cases was followed by parturition 27–30 days later (Table 2). On the five occasions for which records are available, oestrus followed within 1 day of the natural permanent vacation of the pouch. In three of these cases parturition and oestrus (mating) occurred during the same night. Final emergence of the young was associated with a marked contraction of the pouch.

*Breeding season.* Table 1 summarizes data on breeding in captivity. It can be seen that oestrus, as detected by mating or vaginal smears, occurred in all months of the year. Although matings took place in all months apart from March, births followed in only nine months of the year. The marked seasonality of parturition is apparent from Fig. 1, which is derived from the data on shot wild animals. The

distribution of birth dates is similar for the three different regions and periods, despite possible differences in habitat, available food supply and seasonal conditions. The three regions are similar in that they are close to tall open forest. Most young were born in the autumn and winter but some were born in summer, when the winter-born young would be leaving the pouch. It is not known whether the summer births are due to the reactivation and resumption of development of diapausing blastocysts or are the result of ovulation and an uninterrupted pregnancy. However, the latter alternative may be supported by the finding that four wild-caught females whose young were removed in February failed to produce young while kept apart from a male for 30 days. No blastocysts were found in these four females after dissection.

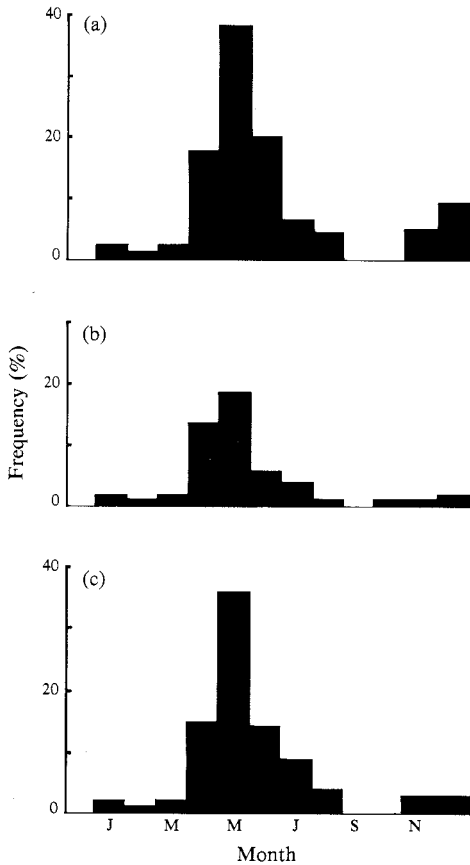


Fig. 1. Frequency distribution of the number of births in each month, in pademelons from three parts of Tasmania.

(a) Florentine Valley, south-west Tasmania, 1959-61;  $n = 102$ .

(b) Avoca, eastern Tasmania, 1978;  $n = 51$ .

(c) Tasman Peninsula, south-eastern Tasmania, 1980;  $n = 89$ .

*Effect of removal of the corpus luteum.* After the surgical removal of the corpus luteum from two females carrying pouch young, oestrus followed 11 days later. On the terminology and assumptions of Tyndale-Biscoe *et al.* (1974), this 11-day period corresponds approximately to the length of time for a new follicle to develop and ovulate, and is designated the follicular phase. The luteal phase occupies the remainder of the oestrous cycle in many mammals; in the pademelon this will approximate to the length of the oestrous cycle (30 days) minus the length of the follicular phase (11 days), i.e. 19 days. Histological examination of corpora lutea (unpublished data) also indicate that the luteal phase lasts 18-19 days.

## Discussion

The reproduction of the Tasmanian pademelon appears to conform to that of most macropodid marsupials (Tyndale-Biscoe *et al.* 1974). Parturition is closely followed by post-partum mating; the zygote produced at this mating remains dormant unless the pouch young is removed. It is as yet unclear whether in the wild most dormant blastocysts are reactivated or lost near the end of the pouch life of the current young. It may be that the viability of the blastocyst is decreased outside of the main breeding season, as in some populations of the quokka *Setonix brachyurus* (Shield and Woolley 1963). The absence of a blastocyst in the uteri of four wild females caught in late summer may indicate that loss of blastocysts is an alternative explanation to failure of blastocysts to reactivate. Sharman (1955) has demonstrated that on Rottneest I. blastocysts were not present in the quokka during seasonal anoestrus.

In captivity, females came into oestrus throughout the year. This may indicate that nutritional factors are involved with female fertility in the wild. Seasonal breeding is characteristic of all Tasmanian kangaroos. *Macropus giganteus* (Pearse, personal communication), *M. rufogriseus rufogriseus* (J. C. Merchant, cited by Tyndale-Biscoe *et al.* 1974), and the pademelon *T. billardieri* breed in such a way that young leave the pouch in the spring and summer. Although there are variations of diet between the three species, it is obvious that the young vacate the pouch at the period of greatest vegetation growth. By contrast, the two Tasmanian rat-kangaroos *Potorous tridactylus* (Buchmann, personal communication) and *Bettongia gaimardi* (Rose, unpublished data) breed throughout the year in the wild, but these animals are less specialized feeders than the larger macropods.

The close temporal relationship of vacation of the pouch with parturition and post-partum oestrus is interesting, although it is not unique to the pademelon. It also occurs, for example, in *M. rufus* (Sharman and Calaby 1964), *M. agilis* (Merchant 1976) and *B. gaimardi* (Rose, unpublished data). It would not be surprising if there was an underlying mechanism synchronizing these events, as it is most important that a young leaves the pouch before the birth of a new young. Further work on this topic is under way.

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## References

- Calaby, J. H. (1971). Current status of Australian Macropodidae. *Aust. Zool.* **16**, 17-31.  
Kirsch, J. A. W. (1977). Comparative serology of marsupials. *Aust. J. Zool. Suppl. Ser.* No. 52.  
Merchant, J. C. (1976). Breeding biology of the agile wallaby, *Macropus agilis* (Gould) (Marsupialia : Macropodidae) in captivity. *Aust. Wildl. Res.* **3**, 93-103.

- Morton, S. R., and Burton, T. C.** (1973). Observations on the behaviour of the macropodid marsupial *Thylogale billardieri* (Demarest) in captivity. *Aust. Zool.* **18**, 1-14.
- Peters, D. G., and Rose, R. W.** (1979). The oestrous cycle and basal body temperature in the common wombat (*Vombatus ursinus*). *J. Reprod. Fertil.* **57**, 453-60.
- Rose, R. W., and McCartney, D. J.** (1982). Growth of the red-bellied pademelon, *Thylogale billardieri*, and age estimation of pouch young. *Aust. Wildl. Res.* **9**, 33-38.
- Sharman, G. B.** (1955). Studies of marsupial reproduction. III. Normal and delayed pregnancy in *Setonix brachyurus*. *Aust. J. Zool.* **3**, 56-70.
- Sharman, G. B.** (1961). The mitotic chromosomes of marsupials and their bearing on taxonomy and phylogeny. *Aust. J. Zool.* **9**, 38-60.
- Sharman, G. B., and Berger, P. J.** (1969). Embryonic diapause in marsupials. *Adv. Reprod. Physiol.* **4**, 211-40.
- Sharman, G. B., and Calaby, J. H.** (1964). Reproductive behaviour in the red kangaroo, *Megaleia rufa*, in captivity. *CSIRO Wildl. Res.* **9**, 58-85.
- Shield, J. W., and Woolley, P.** (1963). Population aspects of delayed birth in the quokka (*Setonix brachyurus*). *Proc. Zool. Soc. Lond.* **141**, 783-90.
- Shorr, E.** (1941). A new technique for staining vaginal smears. III. A single differential stain. *Science (Wash. D.C.)* **94**, 545-6.
- Tyndale-Biscoe, C. H., Hearn, J. P., and Renfree, M.** (1974). Control of reproduction in macropodid marsupials. *J. Endocrinol.* **63**, 589-614.