Influence of melatonin on the initiation of the breeding season of the marsupial bandicoot, *Isoodon macrourus*

R. T. Gemmell

*Department of Anatomy, University of Queensland, St Lucia, Brisbane, Queensland 4067, Australia*

**Summary.** Melatonin implants were administered to 6 female bandicoots during the months of May and July. These animals, together with 6 control bandicoots were housed in large outside enclosures with mature males. Births were observed in the 6 control animals from 26 July to 2 September, but no births were observed in the 6 bandicoots with melatonin implants. These results would suggest that photoperiod, which is known to influence melatonin concentrations, may be a factor in the initiation of births in the bandicoot. However, the gradual build-up of births would suggest that other factors such as temperature and rainfall may also have some influence.

**Introduction**

The bandicoots which reside along the Eastern Australian coast are all seasonally breeding marsupials, most of the births occurring in the spring and summer months (Heinsohn, 1966; Gordon, 1971; Stoddart & Braithwaite, 1979; Gemmell, 1982; Barnes & Gemmell, 1984). The cycle of breeding activity is more pronounced as the latitude increases. In Tasmania, Victoria and New South Wales definite periods of non-breeding or anoestrus were observed, but in Queensland lactating northern brown bandicoots (*Isoodon macrourus*) were observed throughout the year (Hall, 1983), although there was a decrease in breeding during the months April to June (Gemmell, 1982, 1986b). This reduction in the degree of seasonality of reproduction in the bandicoot as the latitude decreases would indicate that the bandicoot is exhibiting a 'high degree of flexibility and opportunism associated with the breeding of most small mammals' (Bronson, 1985). A total of 197 births were monitored from January 1980 to December 1984 in a semi-captive population of northern brown bandicoots housed in Queensland. The complete histogram of the monthly frequency of births indicated that the breeding rate was depressed from March to June, with a gradual increase in births during July and August. Examination of the annual record revealed a variation in the start of the breeding season with the first births occurring after at least a 1 month period of no births, on 1 August 1980, 7 July 1981, 27 May 1983 and 28 July 1984. Births occurred throughout the year of 1982 (Gemmell, 1986b).

Correlations between breeding activity and environmental variations such as daylength, temperature and rainfall have shown that there is a strong association between the number of births and the rate of change of minimum temperature (Barnes & Gemmell, 1984). A subsequent study of plasma testosterone concentrations in the male bandicoot suggested that the annual plasma testosterone profile correlated well with the rate of change of daylength, an increasing daylength being associated with an increase in plasma testosterone concentration (Gemmell et al., 1985). From the above observations, a possible mechanism has been proposed for triggering the start of the breeding season in the bandicoot in Queensland in which the rate of change of daylength may play some part in initiating the breeding season, although the more variable factors such as temperature and rainfall may also be involved (Gemmell, 1986b).

The rate of change in daylength influences the body metabolism of various mammals and is thought to be mediated via the secretion of melatonin by the pineal gland (Reiter, 1980; Kennaway,
1984). The bandicoot is a long-day breeder and if daylength influences reproduction births would be initiated by an increase in daylength and a subsequent decrease in melatonin concentrations. In this study the effect of melatonin implants on the initiation of the breeding season of the northern brown bandicoot was examined.

**Materials and Methods**

Twelve adult female and 4 adult male bandicoots, *Macropus eugenii* housed in 2 enclosures were used in this study. Three control bandicoots without implants and 3 bandicoots with implants were housed with 2 adult male bandicoots in each enclosure. Details of capture and maintenance of bandicoots have been described previously (Gemmell, 1982). The pouches of the female bandicoots were examined weekly. The head lengths of newly born pouch young were used to estimate their day of birth.

**Melatonin implants**

Bandicoots 1-6 were adult females which did not receive an implant. Bandicoots 7-12 received (under halothane anaesthesia) a subcutaneous implant of melatonin, on the backs between the shoulders. These implants consisted of a sealed envelope of Silastic sheeting, surface area 25 cm² (500-1 sheeting, Dow Corning, Midland MI, U.S.A.) containing 0.02 g melatonin (Sigma, Poole, Dorset, U.K.) (Lincoln *et al.*, 1984). All 6 bandicoots received an implant on 8 May and a subsequent implant on 4 July. Blood samples were obtained weekly from all 12 female bandicoots until a young was first observed in the pouches of the 6 control animals or until the end of September in the case of the animals with melatonin implants.

**Progesterone assay**

Plasma progesterone concentrations were determined by radioimmunoassay using the method described previously for the bandicoot (Gemmell, 1979), using sheep anti-progesterone-11-hemisuccinate—bovine serum albumin (antiserum No. 334) kindly donated by Dr R. J. Cox, CSIRO, Prospect, New South Wales, Australia. The antiserum is highly specific for progesterone, the only significant cross-reaction being that with 11β-hydroxyprogesterone (11.9%). The limit of sensitivity of the assay was 25 pg/tube and the intra- and inter-assay coefficients of variation were 13.5% (*n* = 5) and 17.0% (*n* = 10) respectively. The assay buffer blank was less than 0.1 ng/ml and the efficiency of extraction was 83.0% (*n* = 10).

**Results**

Bandicoots 1 and 2 (controls) gave birth on 26 July. Bandicoots 3 and 4 on 24 August, Bandicoot 5 on 1 September and Bandicoot 6 on 2 September. All 6 bandicoots had basal concentrations of plasma progesterone before their first pregnancy of the breeding season and the cleanliness of the pouch and the size of the teats did not change until they became pregnant. The weekly plasma concentrations of progesterone are presented for 3 control bandicoots (Fig. 1).

The plasma progesterone profile of the 6 bandicoots that had received a melatonin implant (Nos 7-12) remained at basal levels, except in Bandicoot 12 in which concentrations of progesterone indicative of an oestrous cycle were observed in September. The pouch morphology of Bandicoots 7-12 was similar to that of Bandicoots 1-6 in that pouches were clean throughout August and September. However, the teat size did not increase, with the exception of Bandicoot 12, during September. The weekly plasma concentrations of progesterone are presented for 3 bandicoots with melatonin implants (Fig. 2).

**Discussion**

Melatonin administered at the start of the breeding season inhibits ovulation and subsequently pregnancy in the bandicoot. Similar treatment of long-day breeding eutherian mammals has been
shown to inhibit reproductive activity. When administered to the Djungarian hamster melatonin prevented the reproductive stimulatory effects of long days (Hoffman, 1973). Similarly, melatonin administered to male rabbits caused the testicular regression normally initiated by exposure to short days (Boyd, 1985). However, in Syrian hamsters stimulation and inhibition of reproductive activity have been reported with constant-release melatonin implants (Goldman, 1983). Continuous exposure of animals to melatonin is not mimicking physiological parameters, since melatonin is normally released as a nocturnal pulse. Melatonin has been administered to two other marsupial species, the tammar wallaby (Macropus eugenii) and Bennett’s wallaby (Macropus rufogriseus): seasonal diapause in both wallabies was rapidly terminated (McConnell & Tyndale-Biscoe, 1985; Loudon et al., 1985). Unlike the wallabies and kangaroos, the bandicoot does not exhibit embryonic diapause (Gemmell et al., 1986). The results with the bandicoot would indicate that the administration of melatonin to long-day breeding mammals, whether marsupial or eutherian, has the same effect, i.e. the inhibition of ovulation and pregnancy.

Melatonin has also been shown to inhibit sexual development in the male white-footed mouse, Peromyscus leucopus (Petterborg & Reiter, 1980). The inhibition of development of the testis obtained with melatonin would also be obtained with exposure to a short photoperiod (Petterborg & Reiter, 1980; Forger & Zucker, 1985). Sexual maturity in the female bandicoot can also be delayed. Females born in the months July–September can produce their first litter in the same season and, give birth at 193·8 ± 7·3 days (N = 5) of age. Bandicoots born in the same period of the year, but for which the first litter is delayed until the next breeding season, give birth at 354·3 ± 7·7 days (N = 6) of age (Gemmell, 1986a). If photoperiod and melatonin have a role in controlling seasonality of breeding in the mature bandicoot, it is probable that the same environmental cues which inhibit breeding in adult females also affect the maturation of juvenile females.
A correlation between an increase in plasma progesterone concentration and an increase in teat size has been reported previously for the bandicoot. Hall (1983) observed that nulliparous females had small, dry pouches with small teats, although just before birth the teats enlarged and the pouch became moist. In a later study, Gemmell (1986b) observed that the increase in plasma progesterone concentration, whether during pregnancy or an oestrous cycle, was always accompanied by an increase in teat size. In the 6 bandicoots receiving melatonin in the present study, an enlargement of teats was only seen with Bandicoot 12 during the same weeks that an increase in plasma progesterone was obtained. Teat morphology would therefore support the view that a non-pregnant or oestrous cycle had occurred in September in this female. Photorefractoriness in long-day breeding mammals involves spontaneous reactivation of the gonads despite continued exposure to short photoperiod or melatonin (Goldman, 1983). Hamsters do not seem to respond to melatonin while they are in a photorefractory state and long-term treatment with melatonin of long-day-housed Syrian hamsters induced testicular regression followed by reactivation (Bittman, 1978). A possible explanation for the change in hormonal concentration in Bandicoot 12 could be that it entered a refractory state in which melatonin could no longer suppress ovulation.

The breeding season in this semi-captive colony of bandicoots varies annually (Gemmell, 1986b), but the start of the breeding season in 1985 was similar to that observed during 1981 and 1984. There were no births during the months March to June; the first births occurred late in July and continued until 2 September. Since the onset of the breeding season in the bandicoot appears as a gradual build up of births, photoperiod is unlikely to be the only cue influencing breeding activity. As suggested previously, photoperiod may be the basic factor controlling seasonality in reproduction, although other factors, such as temperature and rainfall, probably also have some influence.

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References


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