DEVELOPMENT OF OOCYTES FOLLOWING GRAFTING OF MARSUPIAL OVARIAN TISSUE TO A EUTHERIAN RECIPIENT

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Ovarian grafting to a host species is a novel technique for preserving female germ lines. Preliminary studies show that ovarian grafting can be used as an assisted reproductive technique for marsupial species. This study aims to determine the optimal hormonal environment of graft recipients and assess the developmental competence of marsupial oocytes retrieved from ovarian grafts. Ovaries were collected from tammar wallaby adult females and pouch young. Ovarian slices were grafted to the kidney capsule of intact (n = 10) or ovariectomised (n = 8) adult immuno-compromised female mice. The recipients were killed 8-12 weeks after grafting and processed for histology or oocyte culture. Antral follicles on grafts were carefully punctured with a 30 G needle to release oocytes for in vitro culture. These oocytes were incubated in maturation medium (9.875 ml EMEM, 0.100 ml FCS, 0.030 g BSA and 0.01 IU/ml FSH) at 37°C and 5% CO², and observed daily for changes. Follicles at all developmental stages were present in the transplanted ovaries, including corpora lutea. A total of seven mature oocytes from antral follicles were collected along with numerous oocytes from preantral follicles and primordial oocytes. Grafts from intact recipients were larger and contained more oocytes (n = 6) than those from ovariectomised recipients (n = 1). These results further clarify the hormonal environment required of graft recipients and highlight the potential for the use of this protocol in assisted reproductive techniques.

SEASONAL CHANGES IN THE EJACULATE OF THE MALE TAMMAR WALLABY, *MACROPUS EUGENII*: IMPLICATIONS FOR FERTILITY AND ASSISTED BREEDING

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The tammar wallaby, *Macropus eugenii* is a seasonally breeding macropodid marsupial. The seasonality observed in males is known to be driven by the reproductive state of the females. There is a significant increase in male prostate and Cowper's gland weights and testosterone concentration during the breeding season in January/February and again in October when the young females leave their mothers pouches and enter puberty. The dynamics of sperm production in the male tammar wallaby was assessed using changes in ejaculatory and sperm characteristics in and out of the breeding season in order to determine more accurately true seasonality in the male. Semen was collected from wild-caught adult males by electro-ejaculation at four times during the year (January, February, June/July and October). Ejaculates were assessed for semen volume, plug formation, sperm index, percentage and rating of motility, sperm and motile sperm concentration, and total sperm count. Increases were observed during the two breeding seasons in all traits assessed. Semen volume showed a steady increase from June/July (0.7 ml) to reach a peak in February (10.25 ml). In conclusion, we found a significant decrease in the size and coagulation properties of the ejaculate, and in sperm quality out of season. Implications for captive and assisted breeding programs are discussed.

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