

Gross renal morphology of the numbat *Myrmecobius fasciatus*
(Marsupialia: Myrmecobiidae)

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Abstract

There is a strong correlation between the structure of the mammalian kidney and its urinary concentrating ability. We examine here the kidney of an endangered termitivorous marsupial, *Myrmecobius fasciatus*, and use the measured kidney morphometrics to calculate maximal urinary concentration. The relative medullary area (1.34) of the kidney of *M. fasciatus* is typical of other dasyuromorph marsupials, as is its predicted maximal urinary concentration of 3617 mOsm kg⁻¹ H₂O, despite its historically semi-arid/arid distribution. The termitivorous diet of *M. fasciatus* presumably provides it with sufficient water to limit selection for a high urinary concentrating capacity.

The mammalian kidney can produce urine that is significantly more concentrated than the blood plasma (Gottshalk 1987). There is a strong correlation between the structure of the mammalian kidney and its urinary concentrating ability (Sperber 1944; Schmidt-Nielsen and O'Dell 1961; Brownfield and Wunder 1976; Beuchat 1990, 1996). Mammalian kidneys consist of three major regions, the cortex, the inner medulla and the outer medulla. The inner and outer medullae contain the loops of Henle, which are responsible for the counter-current multiplication of solute concentration. The maximum urine concentrating capacity (U_{\max}) of a mammal is proportional to the relative length of its loops of Henle, so there is a strong relationship between relative size of the medulla and U_{\max} for mammals (Sperber 1944; Schmidt-Nielsen and O'Dell 1961; Heisinger and Breitenbach 1969; Brownfield and Wunder 1976; Beuchat 1990). Body mass also contributes to variability in U_{\max} (Beuchat 1990, 1996), reflecting mass-specific metabolic rate, hence the metabolic activity of the kidney (Greenwald and Stetson 1988). For arid-habitat mammals in particular, the production of concentrated urine is important for water conservation, as nitrogenous wastes and ions can be eliminated with a minimum of water loss. Therefore, kidneys of arid-habitat mammals are characterised by a relatively larger medulla compared to those of mesic habitat mammals (Sperber 1944; Beuchat 1996; Diaz and Ojeda 1999; Al-Khatani et al. 2004).

Here we determine the relative medullary area (RMA = area of renal medulla/area of renal cortex) and predict U_{\max} for the numbat, *Myrmecobius fasciatus*, a 550g dasyuromorph marsupial belonging to the monospecific family Myrmecobiidae (Friend 1989). Amongst marsupials, *M. fasciatus* is unique as it is the only strictly diurnal and exclusively termitivorous species. It feeds on subterranean termites that it extracts from shallow sub-surface soil galleries by day and shelters in hollow logs, burrows and tree hollows at night (Friend 1995). Although it was once found throughout southern and central Australia, the geographical distribution of *M. fasciatus* has contracted dramatically since European settlement and it is now restricted to a few remnant populations in Western Australia (Friend 1990). We examine here the kidney of *M. fasciatus* to determine if it shows renal adaptation to its historically arid and semi-arid habitat.

Kidneys from four deceased numbats (two male and two female) were removed within approximately an hour of death, and fixed in 10 % buffered formol saline. One animal was a wild male from Dryandra Woodland (approximately 170 km east of Perth, Western Australia 31° 46' S 117° 1' E) and the other three animals were captive individuals from Perth Zoo. Three right kidneys and one left kidney were available. We

recognise the limitations of using tissue obtained post-mortem, but for an endangered species such as the numbat it is not possible or ethical to obtain fresh kidney tissue from purpose-killed animals, or to directly measure maximal urine concentration. We use RMA as our index of kidney structure for *M. fasciatus*, as Brownfield and Wunder (1976) suggest that it is the best method of predicting urine concentrating ability for mammals.

The kidneys were processed through graded ethanol, chloroform and embedded in paraffin wax. They were then orientated to obtain a midline sagittal section and 6 μm serial sections were cut until the greatest cross-sectional area and papillary length was located. Sections were stained with Harris' haematoxylin and eosin or Heidenhain's azan, then photographed using an Olympus BX 50 light microscope and Olympus DP11 camera. The total, cortical, inner, and outer medullary areas of each section were determined using Image Pro Plus software (Media Cybernetics).

A representative cross-sectional area of a numbat kidney is presented in Figure 1. The mean total area of the mid-sagittal kidney section of $156.4 \pm 11.67 \text{ mm}^2$ consisted of $67.1 \pm 5.59 \text{ mm}^2$ of cortex and $89.3 \pm 6.39 \text{ mm}^2$ of medulla (outer medulla $61.5 \pm 4.37 \text{ mm}^2$ and inner medulla $27.8 \pm 2.03 \text{ mm}^2$). Consequently, the mean RMA for the four numbat kidneys was 1.34 ± 0.054 (Table 1).

The kidney of *M. fasciatus* is typical of that of a generalised marsupial. The RMA for marsupials varies from the particularly low value of 0.3 for the nectarivorous honey possum (*Tarsipes rostratus*; Slaven and Richardson 1988) to 1.42 for the arid-zone fat-tailed false-antechinus (*Pseudantechinus macdonnellensis*; Brooker and Withers 1994). As there was no correlation between body mass and RMA for dasyurids (Brooker and Withers 1994), it was not necessary to mass-correct the RMA of *M. fasciatus* for comparison with other species. The RMA of *M. fasciatus* (1.34) closely approximates the mean value of 1.32 for 29 species of marsupial (Slaven and Richardson 1988; Brooker and Withers 1994). Predicted maximum urine concentration (U_{max}) for *M. fasciatus*, from the U_{max} equation for mammals ($U_{\text{max}} \text{ mOsm kg}^{-1} \text{ H}_2\text{O} = 837 + 2106 \text{ RMA}$) of Brownfield and Wunder (1976) is $3617 \text{ mOsm kg}^{-1} \text{ H}_2\text{O}$. This is similar to that measured for 5 other species of dasyurid marsupial, which ranged from 2951 to 3958 $\text{mOsm kg}^{-1} \text{ H}_2\text{O}$ (mean $3505 \pm 161 \text{ mOsm kg}^{-1} \text{ H}_2\text{O}$; Schmidt-Nielsen and Newsome 1962; Morton 1980; Brooker 1992).

Although no correlation was found between RMA and climatic variables for dasyurids (Brooker and Withers 1994), it is likely that RMA is a function of overall water availability (and consequently the need to produce concentrated urine), which is

determined not only by climate but also by diet, habitat and behaviour. For example, desert woodrats (*Neotoma spp*) feeding on succulent plants produce urine less concentrated than predicted by their arid habitat, due to their high dietary water intake (Lee 1963). The exclusively termitivorous diet of *M. fasciatus* would contribute to a favourable water balance. Termites have a relatively high water content (78%) compared to many foods, and this combined with their relatively low digestibility of 58-81% results in a high water economy index (ratio of water turnover to energy turnover) of 0.20-0.29 (Cooper and Withers 2004). Therefore, energy rather than water is limiting for *M. fasciatus*, which is unlikely to require access to drinking water. Indeed, the field water turnover rate of free-living *M. fasciatus* during summer, without access to free water, conformed to field water turnover rates for other marsupials (100.5% of the allometrically predicted value; Cooper et al. 2003). Presumably there has been little selection pressure on *M. fasciatus* for a high RMA and production of highly concentrated urine, due to high dietary water gain, despite its historically arid and semi-arid habitat distribution.

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References

- Al-Kahtani MA, Zuleta C, Caviedes-Vidal E, Garland T, 2004. Kidney mass and relative medullary thickness of rodents in relation to habitat, body size, and phylogeny. *Physiological and Biochemical Zoology* **77**, 346–365.
- Beuchat CA, 1990. Body size, medullary thickness, and urine concentrating ability in mammals. *American Journal of Physiology* **258**, R298-R308.
- Beuchat CA, 1996. Structure and concentrating ability of the mammalian kidney: correlations with habitat. *American Journal of Physiology* **271**, R157-R179.
- Brooker B and Withers PC, 1994. Kidney structure and renal indices of dasyurid marsupials. *Australian Journal of Zoology* **42**, 163-176.
- Brooker BM, 1992. Kidney structure and urine concentrating ability of dasyurid marsupials. BSc Honours Thesis, Department of Zoology, University of Western Australia, Perth.
- Brownfield MS and Wunder BA, 1976 Relative medullary area: A new structural index for estimating urinary concentrating capacity of mammals. *Comparative Biochemistry and Physiology A* **55**, 69-75.
- Cooper CE and Withers PC, 2004. Termite digestion by the numbat (*Myrmecobius fasciatus*): the inter-relationship between diet, digestibility, and energy and water turnover for myrmecophages. *Physiological and Biochemical Zoology* **77**, 641-650.
- Cooper CE, Withers PC and Bradshaw DS, 2003. The field metabolic rate of the numbat (*Myrmecobius fasciatus*). *Journal of Comparative Physiology B* **173**, 687-693.
- Diaz GB and Ojeda RA, 1999. Kidney structure and allometry of Argentine desert rodents. *Journal of Arid Environments* **41**, 453–461.
- Friend JA, 1989. Myrmecobiidae. Pp. 583-590 In: *Fauna of Australia Mammalia Vol 1B* (eds. DW Walton and BJ Richardson). Australian Government Publishing Service, Canberra.
- Friend JA, 1990. The numbat *Myrmecobius fasciatus* (Myrmecobiidae): history of decline and potential for recovery. *Proceedings of the Ecological Society of Australia* **16**, 369-377.
- Friend JA, 1995. Numbat. Pp. 160-162 In *The Mammals of Australia*. (ed. R Strahan). Reed Books, Chatswood.
- Greenwald L and Stetson D, 1988. Urine concentration and the length of the renal papilla. *News in Physiological Sciences* **3**, 46-49.

- Gottschalk CW, 1987. History of the urinary concentrating mechanism. *Kidney International* **31**, 507-511.
- Heisinger JF and Breitenbach RP, 1969. Renal structural characteristics as indexes of renal adaptation for water conservation in the genus *Sylvilagus*. *Physiological Zoology* **42**, 160-172.
- Lee AK, 1963. The adaptations to arid environments in woodrats of the genus *Neotoma*. *University of California Publications in Zoology* **64**, 57-96.
- Morton SR, 1980. Field and laboratory studies of water metabolism in *Sminthopsis crassicaudata* (Marsupialia: Dasyuridae). *Australian Journal of Zoology* **28**, 213-227.
- Schmidt-Nielsen B and Newsome AE, 1962. Water balance in the mulgara (*Dasyercus cristicauda*) a carnivorous desert marsupial. *Australian Journal of Biological Sciences* **15**, 683-689.
- Schmidt-Nielsen B and O'Dell R, 1961. Structure and concentrating mechanism in the mammalian kidney. *American Journal of Physiology* **200**, 1119-1124.
- Slaven MR and Richardson KC, 1988. Aspects of the form and function of the kidney of the honey possum *Tarsipes rostratus*. *Australian Journal of Zoology* **36**, 465-471.
- Sperber I. 1944. Studies on the mammalian kidney. *Zoologiska Bidrag fran Uppsala* **22**, 249-432.

Table 1: Areas of the cortex, inner medulla and outer medulla (mm²) for four *Myrmecobius fasciatus* kidneys. Values are absolute areas for *M. fasciatus* 1-4, with mean \pm S.E.

Numbat	Cortex	Inner Medulla	Outer Medulla	Total Medulla	Total Area	RMA
1	74.1	32.1	71.3	103.3	177.5	1.39
2	69.6	26.2	58.6	84.8	154.4	1.22
3	74.0	30.3	65.2	95.2	169.3	1.29
4	50.6	22.9	50.9	73.9	124.5	1.46
Mean	67.1 \pm 5.59	27.8 \pm 2.03	61.5 \pm 4.37	89.3 \pm 6.39	156.4 \pm 11.67	1.34 \pm 0.054

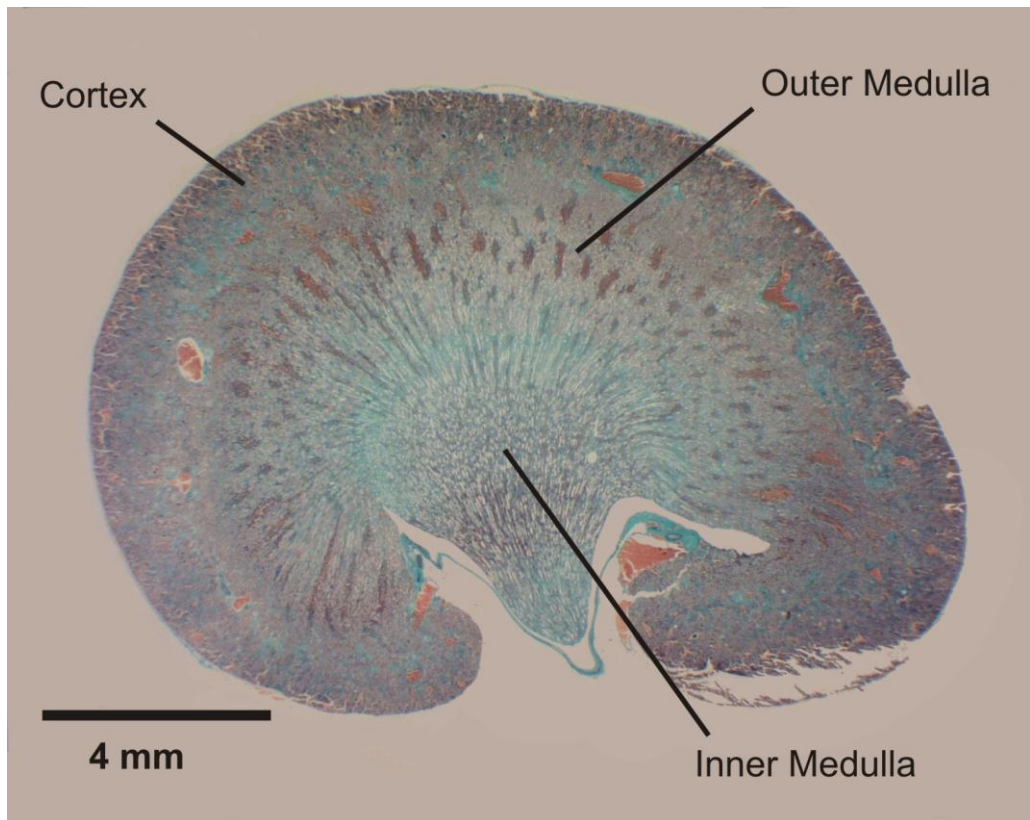


Figure 1: Mid-sagittal longitudinal kidney section of *Myrmecobius fasciatus*, showing the cortex, inner medulla and outer medulla.