

ually test vehicle-control animals simultaneously with the drug-treated animals. Scores are recorded by groups at the end of an appropriate time interval, converted to their logarithms, and averaged. The difference between these two means represents the response for that particular dose. Linear log dose-response curves have resulted from data recorded in this manner.

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References and Notes

1. Plastic containers suitable for animal enclosure construction are available as stock merchandise from Tri-State Plastic Molding Co., Henderson, Ky.
2. I thank J. G. Jackson for assistance in selecting this recording system. The relay unit was designed and constructed by G. F. Stoltz, South Central Broadcasting Corp., Evansville, Ind.
3. S. Irwin, *Rev. Can. Biol.* **20**, 239 (1961).

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Ureotelism of Echidna and Platypus

Abstract. Analyses of plasma and urine and the demonstration of arginase in the liver indicate that both the adult echidna (*Tachyglossus aculeatus*) and platypus (*Ornithorhynchus*) are ureotelic.

Data reviewed by Needham (1) show that the echidna excretes 80 to 90 percent of its nitrogen in the form of urea, and this animal is therefore classified as ureotelic. There is no published information on the platypus. The generalization has been made that animals with a closed egg are uricotelic and this raises interesting questions concerning the monotremes which are egg layers. The echidna lays a soft noncalcareous egg from which the young emerge in the pouch at an early stage and proceed to feed on the milk which exudes into the pouch. For these reasons the pouch has been regarded as "a uterus located in an unusual position," and the echidna may therefore be classified as viviparous. Thus, this exception to the generalization has been tentatively argued away.

As Needham writes, "It would be very interesting to investigate the nitrogen partition in the urine of the platypus which allows its eggs to develop outside the body." Our results indicate that both the adult echidna and platypus are ureotelic. Since we do not possess any information concerning the nitrogen metabolism and water relations of these animals in the egg stage, it cannot be determined how completely the generalization concerning the relationship between the closed egg and uricotelism will have to be modified. Smith (2) raised doubts on this proposition when reporting that turtles excrete most of their nitrogen as urea.

Table 1 shows the analyses of blood plasma and urine. The urea was determined by the method described by Hawk (3), allantoin by the method described by Young and Conway (4), and uric acid by the Folin method (5).

The results show that there is considerable excretion of nitrogen in the form of urea in both the platypus and the echidna. In the echidna, urea is present in the plasma at a concentration similar to that found with mammalian species. Unfortunately we do not have any determinations on the plasma of the platypus. Table 1 also shows that uric acid and allantoin excretion occurs in the monotremes, but in comparison with sheep, a known excretor of allantoin, and the chick, a known excretor of uric acid, the amounts in the specimens we collected are small.

To confirm these results, the presence of arginase was sought in the livers. A simple homogenate of fresh liver was prepared by dispersing 1 g of tissue in 25 ml of 0.1M Na₂HPO₄ with the aid of a Potter-Elvehjem homogenizer. The homogenate (1 ml) was mixed with a solution of arginine (40 mM) which had previously been adjusted to the same pH, and the mixture was incubated at 38°C. The disappearance of arginine and the appearance of ornithine were followed chromatographically by taking serial samples over a period of 1 hour. The solvent system was butanol, acetic acid, and water (80:10:10, by volume), and the chromatograms were run for 2

Table 1. Urea, uric acid, and allantoin per 100 ml of blood plasma (P) and urine (U) collected from two echidna (*Tachyglossus aculeatus*), a platypus (*Ornithorhynchus*), a sheep, and a chicken.

Urea		Uric acid		Allantoin
U (g)	P (mg)	U (mg)	P (mg)	U (mg)
<i>Echidna</i>				
3.43	65	*	0.1	13
	49	24	0.9	
<i>Platypus</i>				
3.4		18		8
<i>Sheep</i>				
		*	0.3	60
<i>Chicken</i>				
	2		5.6	

*Positive.

days. The chromatograms clearly indicated the presence of arginase in considerable amounts: the hydrolysis of 40 μ moles was complete in less than 40 minutes. Under the same conditions liver tissue from the rat completely hydrolyzed the arginine in 20 minutes, but liver tissue from the chick showed no trace of activity. Determination of stoichiometric amounts of urea formed confirmed that the enzymatic activity measured was that of arginase. We therefore conclude that both the echidna and the platypus use urea as the main pathway of nitrogen excretion.

Because of the difficulty of obtaining further animals we thought that these results were sufficiently conclusive at this stage to warrant publication (6).

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2. H. W. Smith, *From Fish to Philosopher*, (Little, Brown, Boston, 1953), p. 245.
3. P. B. Hawk, B. L. Oser, W. H. Summerson, *Practical Physiological Chemistry* (Blakiston, New York, ed. 13, 1957).
4. E. G. Young and C. F. Conway, *J. Biol. Chem.* **142**, 835 (1942).
5. O. Folin, *ibid.* **101**, 111 (1933).
6. We thank the director of the Sir Colin MacKenzie Sanctuary, Healsville, for his assistance with the collection of specimens.

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