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ANZSCPB

15th ANNUAL MEETING

December 4 to 6
1998

Australian and New Zealand Society for Comparative Physiology and Biochemistry
Perth, 1998

The University of Western Australia, Perth

Programme
&
Abstracts

ISBN 0 86422 880 5

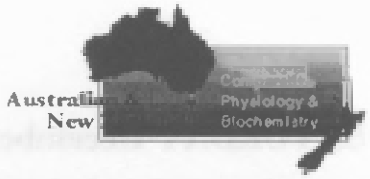
AUSTRALIA AND NEW ZEALAND SOCIETY FOR COMPARATIVE PHYSIOLOGY AND BIOCHEMISTRY
proudly sponsored by: The University of Western Australia, Physiological Zoology/University of Chicago Press, Journal of Comparative Physiology/Springer, Royal Society of Western Australia: affiliated with IUBS and Comparative Biochemistry and Physiology

The 15th annual meeting of the Australia and New Zealand Society for Comparative Biochemistry and Physiology has benefited substantially by generous contributions from the following:

The University of Western Australia, Vice-Chancellory
The University of Western Australia, Faculty of Science
The Royal Society of Western Australia
Springer Publishing (*Journal of Comparative Physiology*)
University of Chicago Press (*Physiological and Biochemical Zoology*)
Pergamon Press (*Comparative Biochemistry and Physiology*)

ANZSCPB Proceedings,
15th Annual Meeting, December 4-6, 1998
The University of Western Australia, Nedlands WA 6907
Organised by Philip C. Withers
ISBN 0 86422 880 5

PROGRAM



FRIDAY December 4

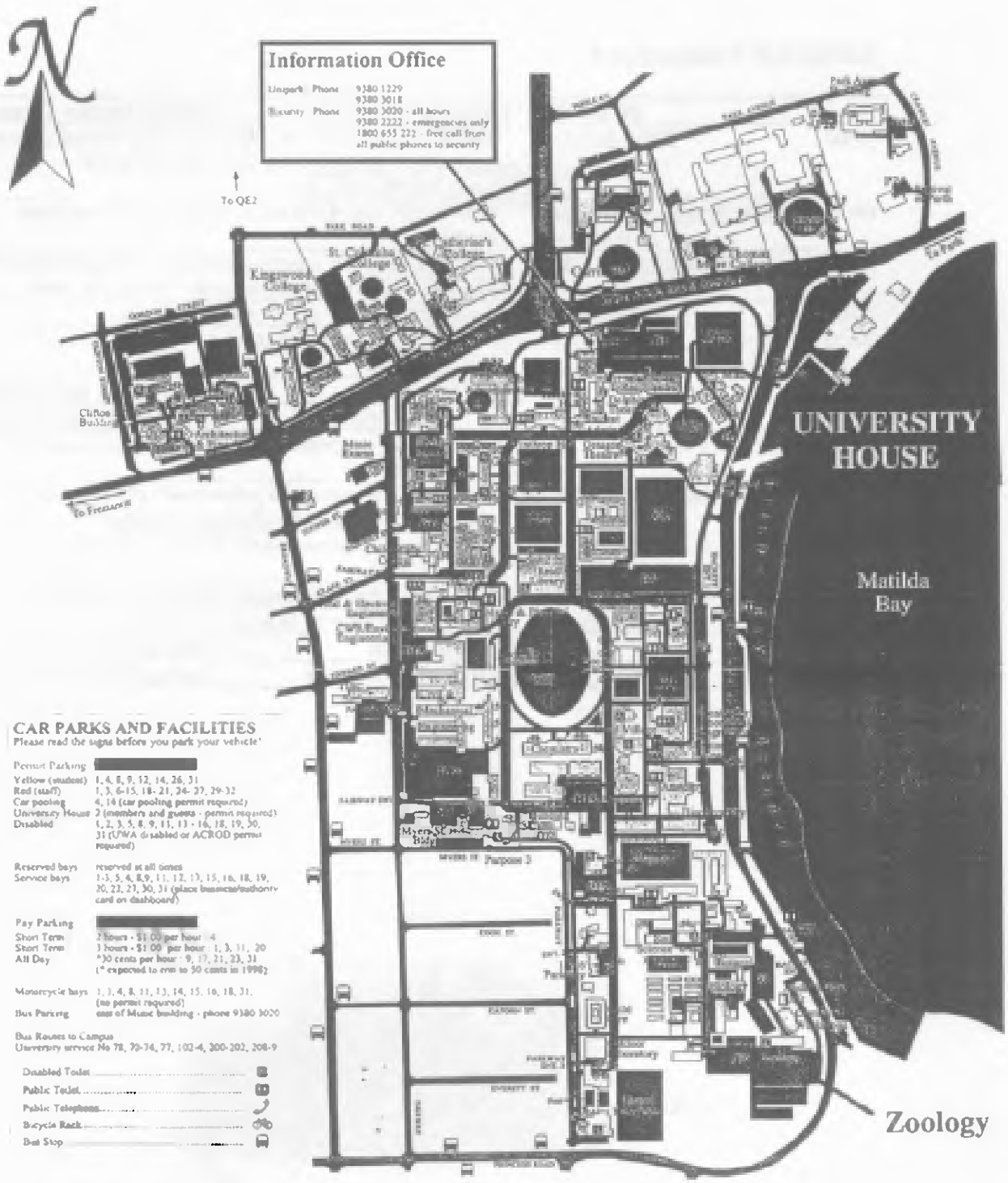
12:00		**REGISTRATION and BARBEQUE**	**University House**
14:10		**WELCOME**	**Zoology Lecture Theatre**
	SESSION 1 Metabolic Plasticity	**Chair: Mike Guppy**	**Zoology Lecture Theatre**
14:20	Philip Withers	Overview of metabolic depression in ectotherms and endotherms	
14:40	Fritz Geiser, G. Körtner & I. Schmidt	Effects of murine leptin on energetics and torpor of the dasyurid marsupial *Sminthopsis macroura*	
15:00	P. Hope, R. Baudinette, H. Turnbull, J. Kumaratilake, & Gary Wittert	Thermogenesis in marsupials: Uncoupling the evidence.	
15:20	Graham Thompson	Comparing the metabolism of reptiles with varying body mass	
15:40		**AFTERNOON TEA**	**Zoology quadrangle**
	SESSION 2 Metabolic Plasticity	**Chair: Fritz Geiser**	**Zoology Lecture Theatre**
16:00	Philip Bethge & S. Nicol	Energy requirements for different activities, including the cost of foraging, in the platypus (*Ornithorhynchus anatinus*)	
16:20	Julian Pakay, A. Hobbs, P. Withers & M. Guppy	The down-regulation of protein synthesis during metabolic depression in the land snail *Helix aspersa*	
16:40	Mark Chappell & K. Hammond	Organ system correlates with variation in aerobic performance in red junglefowl	
17:00		**POSTERS — CHEESE & WINE SOCIAL**	**Zoology 1st floor balcony**
	D. Adams, J. Cockrem & M. Potter	Corticosterone response of captive northern brown kiwi (*Apteryx australis mantelli*) to an acute stressor.	
	P. Bethge, H. Otley, S. Munks, & S. Nicol	The use of dataloggers to determine behavioural activity in the platypus (*Ornithorhynchus anatinus*)	
	P. Celi, D. Blache, G. Martin, P. Vercoe, R. Dynes & R. Tellam	Intracerebral infusion of recombinant leptin inhibits LH secretion in male sheep by decreasing feed intake	
	L. Cutler & A. Patak	Myofibrillar protein composition of regenerating and pristine claw closer muscle of the yabby, *Cherax albidus*	
	J. Elias & J. Baldwin	Anaerobic metabolism in the intertidal elephant snail, *Scutus antipodes* (Fissurellidae)	
	B. Gartrell, S. Jones, R. Brereton & L. Astheimer	An investigation of the factors causing the population decline of the swift parrot (*Lathamus discolor*)	
	F. Geiser & D. Coburn	Energy expenditure and water uptake in free-ranging and captive blossom-bats, *Syconycteris australis* (Megachiroptera)	
	L. Holm, I. Malecki & G. Martin	Effect of temperature and pH on motility and velocity of emu spermatozoa	
	T. Lamey & A. Patak	The effects of moulting on muscle fibre characteristics of the yabby, *Cherax albidus*	
	N. Leelapiyanart & H. Taylor	Respiratory responses of intertidal crab eggs to changes in oxygen tension	
	O. Lopatko, P. Wood, J. Joss, A. Smits & C. Daniels	Acetylcholine controls surfactant secretion by Type II pneumocytes isolated from Australian lungfish and American bullfrogs	
	S. Orgeig, S. Johnston, O. Lopatko & C. Daniels	Development of the pulmonary surfactant system in a bird and reptile	
	J. Van Cleeff, M. Blackberry & G. Martin	Hyperlipaemic emu serum is clarified for radioimmunoassay by polyethyleneglycol (PEG) precipitation	
	J. Wilde & P. Greenaway	Diet and assimilation in the robber crab, *Birgus latro* (L.)	

SATURDAY December 5

	SESSION 3	**Chair: Jamie O'Shea**	**Zoology Lecture Theatre**
09:00	John Donald, B. Davis & K. Minerds	The effect of total water deprivation on the natriuretic peptide system of the hopping mouse, *Notomys alexis*	
09:20	K. Bell & Sue Jones	Changes in plasma concentrations of corticosterone and testosterone in response to capture stress in the lizard *Egernia whitii*	
09:40	W. Callahan & Tes Toop	Fishing for mechanisms: the natriuretic peptide system and the gills of the hagfish	
10:00	Julie Roberts	Aldosterone and renal function in avian species	
10:20	Dennyse Newbound, J. O'Shea & P. Withers	Osmoregulation and ionoregulation in the Port Jackson shark, *Heterodontus portusjacksoni*	
10:40		**MORNING TEA**	**Zoology quadrangle**
	SESSION 4	**Chair: Tess Toop**	**Zoology Lecture Theatre**
11:00	Judith Van Cleeff, D.Blache, I. Malecki, and G. B. Martin	Gonadal hormones are proximate factors controlling seasonal fat metabolism in adult male emus	
11:20	Perdita Hope, W. Breed, H. Turnbull, J. Morley, M. Horowitz & G. Wittert	Gonadal steroids and alterations in photoperiod affect metabolic efficiency in female *S. crassicaudata*	
11:40	Colleen Veitch, C. Sernia & R. Gemmell1	Prostaglandin F2α Receptors in the brushtail possum, *Trichosurus vulpecula* and the short-nosed handicoot, *Isoodon macrourus* at birth	
12:00	Bronwyn McAllan & J. Joss	Effects of oral administration of melatonin on the reproductive cycle of a small marsupial, *Antechinus stuartii*	
12:20	Agung Riono, D. Blache, A. Dharmarajan & G. Martin	Ingestion of extract of leaves of mangosteen (*Garcinia mangostana* Linn) induces germ cell apoptosis in rat testis	
12:40		**LUNCH**	**Zoology quadrangle**
	SESSION 5	**Chair: Peter Greenaway**	**Zoology Lecture Theatre**
14:00	Robert Gemmell & B. Harmon	The function of the vaginal caecae of the marsupial bandicoot, *Isoodon macrourus*	
14:20	Michel Beal & N. Scott	Plasma concentrations of calcium, magnesium and phosphate during development of tammar pouch-young (PY)	
14:40	Ashley Edwards & S M Jones	Steroid biosynthesis in a viviparous reptile, *Tiliqua nigrolutea* (Scincidae)	
15:00	Peta Edwards, B. Tuch & M. Beal	Abundance of endocrine cells in the pancreatic islets of macropods	
15:20	K. McClelland & Ian Hume	Selective retention of digesta in the caecum of bandicoots	
15:40		**AFTERNOON TEA**	**Zoology quadrangle**
	SESSION 6	**Chair: Ian Hume**	**Zoology Lecture Theatre**
16:00	Jamie O'Shea	The innervation of the heart of the fat-tailed dunnart, *Sminthopsis crassicaudata*	
16:20	Suzanne Munns & B. Evans	Hypercapnia induced depression of ventilation in the Gippsland water dragon	
16:40	Anton Thompson & J. O'Shea	A cholinergically mediated adrenergic response in the heart of the horned shark, *Heterodontus portusjacksoni*	
17:00	Alan Roberts	Ecological correlates of locomotor performance in geckos	
18:30		**Pre-dinner drinks**	**Universlty House**
19:00		**1998 ANNUAL MEETING DINNER**	**University House**

SUNDAY December 6

	SESSION 3	**Chair: Don Bradshaw**	**Zoology Lecture Theatre**
09:00	Willie van Aardt	The MO_2 and metabolic release of CO_2 from labeled glucose and lactate in the exercising ghost crab, *Ocypode ceratopththalmus*	
09:20	Philip Withers, G. Thompson & R. Seymour	Metabolic physiology of the marsupial mole (*Notoryctes caurinus*)	
09:40	John Baldwin, A. Gupta & X. Inglesias	Scaling of anaerobic energy metabolism during tail flipping behaviour in the freshwater crayfish, *Cherax destructor*	
10:00	Michael Guppy	A comparative study of fuel choices by cells	
10:20	Joseph Hoh, Y. Kim, L. Sieber & C. Lucas	Macropodids express alpha cardiac myosin in their jaw-closing muscles	
10:40	**MORNING TEA**		**Zoology quadrangle**
	SESSION 4	**Chair: Phil Withers**	**Zoology Lecture Theatre**
11:00	Trevor Anderson & S. Smith	The relationship between histidase activity and trophic habit in surgeon fishes	
11:20	Don Bradshaw & F. Bradshaw	Measurement of nectar and pollen intake of free-ranging marsupial Honey possums (*Tarsipes rostratus*)	
11:40	Patrick Spanoghe, G. Davidson & B. Paterson	Effect of aerial exposure on the western rock lobster, *Panulirus cygnus*	
12:00	Stewart Nicol & N. Andersen	Using dataloggers to investigate hibernation in echidnas	
12:20	James Pearson	Physiological precocity in an altricial parrot	
12:40	**Award of Student Prizes**		**Zoology Lecture Theatre**
13:00	**DISCUSSION SESSION - 1999 Meeting**		**Zoology Lecture Theatre**



THE UNIVERSITY OF WESTERN AUSTRALIA

ABSTRACTS OF TALKS

The relationship between histidase activity and trophic habit in surgeon fishes

Trevor A. Anderson and Shawn Smith

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Herbivorous animals survive on a diet which is relatively poor in protein and essential amino acids (EAA) and may therefore possess mechanisms that protect the EAA from degradation by modifications in the K_m and V_{max} of the key degradative enzymes. We expected that the initial enzyme in the degradative pathway of an essential amino acid would have a lower K_m and a higher V_{max} in animals that receive an adequate supply of essential amino acids (carnivores). Our initial test of this hypothesis focussed on histidase, the primary enzyme in histidine degradation, as histidine levels are generally tightly regulated in fish. Individuals of five closely related species of tropical surgeon fish with a range of trophic habits were captured by spear, dissected and liver and muscle tissue stored in liquid nitrogen until assay. Homogenates of tissue were prepared and assayed for histidase using the method of Chiu *et al.* (1984).

The carnivorous *Naso hexacanthus* and *N. vlamingii* and the fermenting herbivore *N. unicornus* had lower K_m and V_{max} for liver histidase than the detritivorous *Ctenochaetus striatus* which had lower values than the non-fermenting herbivore *Acanthurus lineatus*. These data support the hypothesis with regard to K_m but not with regard to V_{max}.

Chiu, Y.N., Austic, R.E. & Rumsay, G.L. (1984) Comp. Biochem. Physiol. 78B, 777-783.

Scaling of anaerobic energy metabolism during tail flipping behaviour in the freshwater crayfish, *Cherax destructor*

J. Baldwin, A. Gupta and X. Inglesias

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Yabbies (*Cherax destructor*) display an escape behaviour in which rapid bursts of tail flipping propel the animal backwards. This is a classic example of anaerobic muscle work, initially powered by phosphagen hydrolysis then switching to anaerobic glycolysis once arginine phosphate reserves are depleted. We have investigated the effect of animal size on anaerobic scope (maximum rate of energy production) and anaerobic capacity (total energy used to reach exhaustion) during this escape behaviour.

The positive allometry of anaerobic scope, reflected in the activities of rate limiting glycolytic enzymes, scales with the increased power required by larger animals to overcome drag during locomotion through water. Exercise time and number of tail flips required to reach exhaustion showed poeitive allometry, as did anaerobic glycolytic capacity. However, the contribution of phosphagen hydrolysis to anaerobic capacity was independent of body mass. Limits to anaerobic capacity are not set by fuel stores, but may involve inhibition of glycolytic enzyme activity at the low pH values reached in tail muscle of exhausted animals. It is suggested that this positive allometry of anaerobic capacity observed during enforced exercise may not be utilised routinely in nature because of metabolic constraints imposed during recovery of large animals.

Plasma concentrations of calcium, magnesium and phosphate during development of Tammar pouch-young (PY)

A. M. Beal and N. A. Scott

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At birth, marsupial neonates lack calcified bones. Whether they are born with some capacity to regulate calcium and phosphorus or when this develops during pouch-life is unknown. We have measured plasma and milk concentrations of Ca, Mg, PO_4 and other ions from mothers and PY starting at 25 d post-birth. PY plasma [Ca] increased progressively from 25 to 150 d post-natum but remained lower than maternal plasma Ca ($P<0.001$) up to 100 d. In contrast, PY plasma was hypermagnesic to maternal plasma being near constant up to 100 d post-birth but rising between 100 and 200 d. Despite milk having substantially higher levels of both elements than either maternal or PY plasma, the concentrations of Ca and Mg in milk were not correlated with plasma levels of mothers ($P = 0.4$ and 0.93) or PY ($P = 0.18$ and 0.54). PY were hyperphosphataemic relative to the mothers throughout pouch-life with PY plasma concentrations increasing over the 100-250 d post-birth. Milk [PO_4] were much higher than that in plasma from the mothers or PY and was negatively correlated with maternal plasma ($P = 0.023$) and positively correlated with PY plasma ($P = 0.005$) throughout pouch life. The data suggest that PY gain control of Ca homeostasis earlier than that of Mg or PO_4.

Changes in plasma concentrations of corticosterone and testosterone in response to capture stress in the lizard *Egernia whitii*

K. Bell and S. M. Jones

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In vertebrates, exposure to a stressor generally stimulates the secretion of corticosteroids by the adrenal cortex. Often, an increase in plasma corticosteroids is followed by a decrease in plasma sex steroid concentrations; however, in reptiles, hese generalisations do not apply to all species. Males of the lizard *Egernia whitii* ere captured in the field and bled at 0, 10, 60 or 240 minutes after capture, each imal being sampled once only. The response to acute capture stress was reflected a rapid increase in plasma corticosterone concentrations, with a significant vation after ten minutes, and concentrations remaining high at 240 min. Plasma osterone concentrations also changed dramatically, but at a slower rate. enal-gonadal interactions in *E. whitii* were also investigated during long term vity, and through the reproductive cycle. There were no significant differences en males and females in plasma concentrations of corticosterone through the season, with lowest values occurring during the postpartum period. These were unexpected; the lack of sex-related differences in plasma corticosteroid trations may reflect the social nature of this species.

Energy requirements for different activities, including the cost of foraging, in the platypus (*Ornithorhynchus anatinus*)

Philip Bethge and Stewart Nicol

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We measured the energy requirements of freely-swimming platypuses in a 11 m^3 swim tank using respirometry. In the tank animals foraged voluntary while all activities and gaseous exchange were closely monitored. In addition we used a conventional treadmill to determine energy requirements for walking. Energy requirements of the active platypus were found to be higher than maximal metabolic rates for the animal reported in the past. Energy requirements for foraging averaged 8.6 W/kg (n=6) for dives lasting the normal dive duration of 20 to 40 s observed in the wild and increased slightly with decreasing dive duration. The metabolic rate for walking was found to be 8.7 W/kg (speed 0.2 m/s, n=8). Resting rates of the platypus were found to increase with decreasing air and water temperature respectively. Minimal energy requirements for resting under the water surface (wedging) amounted to 3.6 W/kg (water temp 14°C). Minimal metabolic rate of platypuses resting on the water surface was found to be 3.9 W/kg (water temp 20°C) while minimal resting rate on land was 2.1 W/kg (air temp 26°C).

Measurement of nectar and pollen intake of free-ranging marsupial honey possums (*Tarsipes rostratus*)

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Turnover rates of a combination of stable and radioactive isotopes (tritium, oxygen-18 and sodium-22) have been used to estimate daily intake of pollen and nectar in free-ranging honey possums in Scott National Park in the extreme southwest of Western Australia. The Field Metabolic Rate (FMR) is measured using doubly-labelled water and nectar intake is estimated independently from the turnover of both water and sodium. Nectar energy intake is then estimated from the sugar concentration of fresh nectar collected from the inflorescences on which the honey possums have been feeding. Pollen intake is finally estimated as the difference between the energy derived from nectar and the FMR. The method assumes that honey possums normally do not drink free-water in the field, that all the sodium in their diet is derived from nectar, and that individuals are in energetic balance over the measurement period. These assumptions have been tested and found to be reasonably robust, except during periods of heavy rain when some free-water intake may occur. Data from some 30 individuals show that nectar intake is reasonably constant at 6-7 $ml.day^{-1}$ for individuals with an average mass of 9 g. Daily pollen intake varies considerably between individuals, however, averaging 510 $mg.day^{-1}$ but ranging from 0 to as much as 2 $g.day^{-1}$. Laboratory studies with a captive colony of honey possums show that the minimum nitrogen requirement for the maintenance of balance in this species is approximately 115 $mg.kg^{-0.75}.day^{-1}$, well below that recorded for macropodid marsupials but very similar to that of the sugar glider, *Petaurus breviceps*, which also feeds on a high carbohydrate diet.

Fishing for mechanisms: the natriuretic peptide system and the gills of the hagfish

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Previous studies have identified a natriuretic peptide (NP) system in the hagfish. This earlier research focussed on the Atlantic hagfish, *Myxine glutinosa*. Recently, whole gill perfusion studies and myography of afferent and efferent branchial arteries have implicated NPs in the control of blood flow through the gills of the New Zealand hagfish, *Eptatretus cirrhatus* (Forster, Glover and Simpson, XIII Int. Congress Comp. Endocrinol. 1997). However, the NP receptors that would mediate this regulation have not been identified in this species. The current study demonstrates the presence of NP binding sites in the gills of *E. cirrhatus* and examines whether NP actions in the gills of this hagfish could be mediated by guanylate cyclase receptors, similar to those found in jawed vertebrates.

Organ system correlates with variation in aerobic performance in red junglefowl

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Limits to whole-animal performance may be set by peripheral effectors, central organs, or by all components in synchrony ("symmorphosis"). We examined these concepts using aerobic performance in red junglefowl as a model system. We measured individual variation in the masses of peripheral (pectoralis and leg muscles) and central organs (heart, lungs, gonads, liver, spleen, and gut), and searched for correlations with minimal and maximal aerobic metabolism (BMR and VO_{2max}). We found substantial variance within and between sexes, but few consistent relationships between performance and organ size. On average, males had lower BMR and higher VO_{2max} than females, and had smaller visceral organs (gut, liver) and larger cardiovascular organs (heart, lung, haematocrit) and muscles. BMR was not correlated with any single organ mass in either sex (there was no relationship between female reproductive status and either BMR or VO_{2max}, although gonadal and oviduct mass varied more than 12-fold). In males only, BMR was correlated to combined gut mass. Male VO_{2max} residuals were correlated to heart and muscle mass; female VO_{2max} residuals were correlated to haematocrit. These results suggest that attaining high aerobic performance requires increased investment in muscles and in organs involved in oxygen delivery, but not in digestive organs.

The effect of total water deprivation on the natriuretic peptide system of the hopping mouse, *Notomys alexis*

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The natriuretic peptide system counters hypervolaemia and concomitant hypertension by reoal diuresis and natriuresis, and vasodilation. However, the importance of hypervolaemic regulation in mammals has been questioned, and in particular, the role of natriuretic peptides in the maintenance of normal fluid balance. We are using the hopping mouse, *Notomys alexis*, to investigate the function of natriuretic peptides in a desert-adapted mammal that can survive without free access to water. Atrial natriuretic peptide (ANP) cDNA was cloned from N. alexis, and the sequence of ANP (1-28) was identical to rat ANP. This information enabled a radioimmunoassay for rat ANP to be used to quantify ANP levels in the heart and plasma of N. alexis. N. alexis, of either sex, were divided into an experimental group (n=12) subject to seventeen days of total water deprivation (TWD), and a control group (n=12) that had free access to water. No significant difference in haematocrit, skeletal muscle and liver water content, and cardiac ANP levels was observed between control and experimental groups. However, plasma ANP and the density of ANP binding on renal glomeruli were significantly lower in TWD animals. This data suggests that the ANP system is down-regulated during TWD, probably to conserve salt and water.

Steroid biosynthesis in a viviparous reptile, *Tiliqua nigrolutea* (Scincidae)

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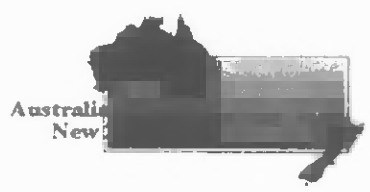
Predominance of either the delta-4 or delta-5 pathway for gonadal steroid biosynthesis varies between vertebrate classes. More recent information suggests that variation between both sex and reproductive condition can also occur. We examined *in vitro* biosynthesis of progesterone (P4), androstendione (AD), dehydroepiandrosterone (DHA), testosterone (T) and 17β-oestradiol (E2) from pregnenolone (P5). Late pre-ovulatory (Oct) and post-partum (Mar) ovaries and final-spermatogenesis (Sep) and quiescent (Feb) teates of a viviparous skink, Tiliqua nigrolutea were used. Steroids were identified using various thin layer chromatography (TLC) solvent systems and HPLC (attached to a radiometric detector) elution times. A strong preference for the delta-4 pathway was apparent in both sexes and at both reproductive and quiescent times of year. However, for several key steroids, the relative proportions synthesised during incubation differed between reproductive conditions. Interestingly, we have demonstrated that ovarian follicles do not produce E2 from P5 *in vitro* at either time of year. The end product of P5 metabolism is instead another, more polar steroid, which is currently unidsntified.



Abundance of endocrine cells in the pancreatic islets of macropods

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For more than twenty years, it has been believed that the islets of Langerhans from the macropod pancreas were essentially similar to those from the sheep pancreas in having a preponderance of α cells (glucagon-secreting) and few β cells (insulin-secreting) in comparison with islets from monogastric animals. This was argued to be an adaptation to foregut fermentation. On finding that the islets of one tammar wallaby had high β and low α cell numbers we have reassessed the proportions of the 4 cell types in islets from red and eastern grey kangaroos, tammars and sheep using immunohistological techniques. The results confirm that sheep islets have high α cell (53.9 ± 3.20 %) and low β cell numbers (29.1 ± 1.61 %) with the remainder being somatostatin-secreting δ cells (6.2 ± 1.96 %) and pancreatic polypeptide secreting ϕ cells (10.8 ± 4.04 %). In contrast, reds, greys and tammars were similar in having nearly twice as many β cells as α cells averaging 52.4 ± 4.36 % β-cells, 28.9 ± 2.27 % α-cells, 9.2 ± 1.33 % δ-cells and 10.5 ± 2.58 % ϕ-cells. Apparently, high α cell numbers are not an essential adjunct to being a forgut.

Effects of murine leptin on energetics and torpor of the dasyurid marsupial *Sminthopsis macroura*

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Leptin plays an important role in regulating body fat stores of placental mammals, but the contribution of changes in energy uptake and expenditure to this adjustment remains controversial. We were interested in how recombinant murine leptin would affect metabolic rate and body temperature of a marsupial mammal (*Sminthopsis macroura*, 25 g) known to display daily torpor. In a group of 8 animals, food-deprived for 1 day at 18 °C, leptin treatment (5 μg/g body mass in Tris buffer) halved the duration of torpor bouts, and raised the average daily minimum body temperature by 4.5 °C and the minimum metabolic rate by 2.2-fold in comparison to control treatment with Tris buffer. Leptin treatment thus increased daily energy expenditure by 9% although during the activity phase metabolic rates and body temperatures were not raised. Body mass was also not affected. These findings suggest that leptin affects the adjustment of thermoregulatory energy expenditure during the rest phase. They show that the hormone has strong physiological effects despite the lack of thermogenetically active brown fat in marsupials and some structural differences between murine and marsupial leptin.

The function of the vaginal caecae of the marsupial bandicoot, *Isoodon macrourus*

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In the bandicoot the lateral vaginae are straight and about 50mm long and there are large lateral anterior expanaions at the diatal ends, called the vaginal caecae. After coitus the caecae are dilated and filled with a clear viscid semifluid containing abundant numbers of sperm. No direct function other than a store for sperm has been suggested for the caecae. In the present study the structure and function of the vaginal caecae, the longevity of sperm in the vaginal caecum and the conception rate of the bandicoot Isoodon macrourus will be examined. Material from the lumen of the vaginal caecae obtained using a polyeththylene tube was placed on a microscope alide and the presence of motile aperm noted. Motile sperm were detected in the vaginal caecae approximately 12 hours after mating and non motile sperm were present after I8 hours. Sperm without heads were observed 6 to 8 days after conception. The vaginal caecae of the bandicoot may act as a short-term storage area that allows viable sperm to be released over several hours. These sperm fertilise the ova aa they pass through the uterine tube and allow the bandicoot a high rate of conception. The conception rate for *I. macrourus* is 100% (N=8).

A comparative study of fuel choices by cells

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Modern biochemiatry and physiology increasingly rely on the use of isolated cell preparations. But despite this, there are few data on what fuels cells use in an in vitro physiological situation to produce the ATP required to support energy-consuming processes, and no data on the comparative aspect of fuel choices by cells. This deficiency has been recently redressed by Guppy *et al.* (1997) who took advantage of the fact that human platelets can be incubated *in vitro* in their *in vivo* medium (human plasma). The data showed that lactate production and lipid oxidation accounts for 62% of the ATP turnover with minor contributions from the oxidation of glucose, ketones and glutamine, leaving 26% of the ATP turnover as yet unaccounted for. In a similar experiment using trout platelets in an artificial plasma, lactate production and fatty acid oxidation only accounted for 17% of ATP turnover, with again trivial contributions by the oxidation of glucose and glutamine. Mammalian and fish cells therefore show some marked differences in terms of fuel choices. This is interesting in itself, but also needs to be taken into account when designing culture media, if physiologically meaningful data are to be produced.

Guppy M, Abas L, Neylon C, Whisson M E, Pethick D W Niu X W. (1997) Fuel choices by human platelets in human plasma. Eur. J. Biochem. 244: 161 167.

Macropodids express alpha cardiac myosin in their jaw-closing muscles

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Jaw-closing muscles of vertebrates are functionally extremely diverse, having to adapt to different types of food they handle. Carnivores, primates and chiropterans express superfast myosin, rodents express fast myosins, while eutherian herbivores express slow myosin (= cardiac V3) in their jaw muscles. We investigated whether the macropodids, being herbivores, also express V3 myosin. Pyrophosphate gel electrophoresis revealed that ventricular myosin from the tammar wallaby and the red kangaroo had three ventricular myosins, V1, V2 and V3. Masseter myosin from the macropodids was homogeneous and comigrated with V1, the high ATPase cardiac isoform. An antibody raised in rabbits against red kangaroo masseter myosin reacted immunohistochemically specifically with macropodid jaw fibres, as well as atrial and ventricular muscles, but did not react with limb muscle fibres. In pyrophosphate Western blots, this antibody reacted with V1(myosin heavy chain composition: αα) and V2 (αβ), but not with V3 (ββ). These results confirm that macropodid jaw muscle express αMyHC rather than βMyHC. This difference in jaw myosin expression between eutherian herbivores and macropodids may be related to the fact that macropodids do not ruminate, and need faster and more powerful jaw muscles to comminute food only once to the correct particle size for gastric fermentation.

Thermogenesis is marsupials: Uncoupling the evidence.

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We have previously reported the presence of UCP1 in the dasyurid marsupial *S. crassicaudata*. Furthermore *S. crassicaudata* increases thermogenesis in response to both catecholamines and food intake, although UCP1 may not be involved in this process. In the 180 day old macropod *Macropus eugenii* (tammar wallaby), electronmicroscopy of interscapular fat showed mostly cells typical of white adipocytes; a few multilocular cells with a higher proportion of mitochondria were also seen. By contrast in perirenal fat there was a much higher proportion of these cells. While we did not detect a 32kd band consistent with UCP1 on an immunoblot, we found a much smaller band (~18kd) in the 200, but not 180 day old wallabies. Expression was greatest in perirenal fat and lowest in subcutaneous fat. Both GDP binding and thermogenesis were greater at 200 as compared to 180 days. Interestingly a ~1.2 kb mRNA band was seen in perirenal fat but not cortex, muscle or liver on an RNA blot hybridised with an oligoprobe specific for UCP1.

Recently other members of the uncoupling protein family (UCP2,3,4) have been cloned. We have identified the expression of UCP2 mRNA in both *S. crassicaudata* and *M. eugenii*, where expression is greater in perirenal than subcutaneous fat. In the new born wallaby, UCP2 expression is very low in perirenal fat and barely detectable in muscle. By 150 days expression is markedly increased in both perirenal fat and muscle, but not subcutaneous fat. No further increase in expression was seen at 200 days.

These data suggest that uncoupling proteins play a role in marsupial thermogenesis, even in the absence of clearly defined collections of brown adipose tissue.

Gonadal steroids and alterations in photoperiod affect metabolic efficiency in female *S. crassicaudata*

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In the marsupial *S. crassicaudata*, adiposity increases in animals housed under short day photoperiods (SD), as compared to long day photoperiods (LD). This effect is seen in both sexes, and occurs without an increase in energy intake, suggesting increased metabolic efficiency. To determine the influence of gonadal steroids on the response to photoperiod, female animals were either gonadectomised or sham operated and maintained under LD or SD for 103 days. Alterations in photoperiod had no effect on body weight in the sham controls, however SD housed gonadectomised animals gained weight over the first 60 days and thereafter body weight returned to baseline. Tail width (a reflection of body fat stores) increased in both sham ($P<0.05$) and gonadectomised ($P<0.01$) animals under SD. However on LD, tail width increased only in the gonadectomised animals and this occurred in the first 20 days ($P<0.01$) with no further increase thereafter. By contrast in the SD gonadectomised animals tail width increased only after day 20 ($P<0.01$). Gonadectomy resulted in a decrease in UCP2 expression in muscle, but not fat. There was no effect of photoperiod on UCP2 expression in any tissue. These data suggest that (i) body fat stores in *S. crassicaudata* are affected by gonadectomy and alterations in photoperiod (ii) both gonadal steroid dependent and independent mechanisms mediate the effect of photoperiod (iii) alterations in UCP2 expression may account for the effect of gondectomy, but not photoperiod and (iv) expression of UCP2 is differentially regulated in muscle and fat.

Effects of oral administration of melatonin on the reproductive cycle of a small marsupial, *Antechinus stuartii*

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Antechinus stuartii is a small marsupial with a brief, highly synchronised mating period believed to be controlled by the rate of change of photoperiod. Photoperiodic response in mammals is co-ordinated by the pineal gland and its hormone melatonin. The pineal hormone, melatonin, administered in the drinking water from the winter solstice, was found to affect the normal response of *A. stuartii* to increasing rate of change of photoperiod. Melatonin shifted the induction of estrus in the females from the first week of August (controls) to an earlier time of mid-July and the consequent pouch changes associated with pregnancy and psuedopregnancy were also shifted by the same length of time. The post-mating decline and consequent death in the males were also accelerated. The results were significantly different between control and melatonin treated groups ($p<0.05$). The hormone melatonin appears to be important in the timing of reproduction in this marsupial.

Osmoregulation and ionoregulation in the Port Jackson shark, *Heterodontus portusjacksoni*

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A comprehensive anatomical and physiological study of the Port Jackson shark, *Heterodontus portusjacksoni*, which migrates into estuarine environments to lay eggs showed relationships between the structure of the three organs involved in osmoregulation in elasmobranchs, the gills, kidneys and rectal salt gland, and how their function at lowered salinity may be achieved. Experimental acclimation to 67% seawater was undertaken over a period of six days.

The plasma of the Port Jackson shark is slightly-hyperosmotic to the environment at 100% and 67% seawater with high elevated levels of urea. The rectal salt gland has a complex structure, with the flow of secretory tubules oppposite to the capillaries, in a counter-current arrangement. The counter-current organisation and the lobulate arrangment of the gland function to concentrate rectal gland fluid which is predominantly sodium, chloride and magnesium. The structure of the glomeruli in the kidney suggests an important ultrafilration role for the nephron. The significance of this process may be replaced by secretion in lowered salinity. Chloride cells in the gills are likely to function in the elimination of ions.

The osmotic strategy used by the Port Jackson shark when adjusting to dilute media was to maintain urea concentrations at the expense of other electrolytes (especially NaCl). This strategy is suggested to be economical, as electolytes can rapidly be elevated by the passive diffusion influx of ions when sharks return to a marine environment. It is in contrast to the general pattern in elasmobranchs that acclimation to dilute external environment is achieved by a reduction in urea concentration, with minor reductions in electrolyte concentrations.

Using dataloggers to investigate hibernation in echidnas.

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Grigg, Beard & Augee used implanted temperature sensitive radio transmitters in free ranging animals to demonstrate that echidnas are true hibernators, but temperature transmitter based systems are prone to problems, such as loss of data when animals move out of range. We have used two different types of implanted datalogger to study echidnas throughout the year: a one channel logger that recorded body temperature, and a two channel logger that recorded body temperature and activity. The activity logger had the capacity to provide detailed information on activity patterns, and while it produced some useful data it proved unreliable. The temperature loggers produced more insights into echidna physiology than had been expected: as well as demonstrating an unexpectedly early entrance into hibernation by male echidnas the temperature records could be used to clearly identify reproductive activity of females.

Grigg G C, Beard L A & Augee M L (1989). Hibernation in a monotreme, the echidna (Tachyglossus aculeatus). Comp. Biochem. Physiol. 92A: 609-612.

The innervation of the heart of the fat-tailed dunnart *Sminthopsis crassicaudata*

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The innervation pattern of the heart of the dasyurid marsupial, *Sminthopsis crassicaudata*, by autonomic nerves was studied using isolated cardiac preparations. The atrial cardiac chambers and strips of the ventricular muscle were maintained under controlled conditions and allowed to beat spontaneously or were electrically paced. The presence or absence of autonomic nerves was demonstrated by recording any changes in the contractile force of the cardiac preparations in response to electrical stimulation across their walls. Nerve-mediated responses to the stimulation of the intramural nerves were mimicked with applied neurotransmitters and challenged by applying known neuroreceptor blocking reagents. Stimulation of intramural nerves in the ventricles caused a substantial inhibition of contractile force mediated by cholinergic nerves and an excitation mediated by adrenergic nerves. The inhibition to ventricular contractile force which occurred in response to nerve stimulation and applied acetylcholine occurred in the absence of any adrenergically maintained tone. Although the innervation pattern of the dunnart atria ia typically mammalian that of the ventricles is not. The presence of a functional adrenergic innervation in the ventricles of a mammal that is able to enter and arouse from torpor is contrary to accounts for most hibernating mammals. This adrenergic ventricular innervation may be involved in the large increases in cardiac output associated with the massive adrenergic activation that mediates arousal from torpor. The occurrence of a cholinergic vagal innervation capable of inhibiting the basal force of ventricular contraction has been described in only one other group of mammals, the bats.

The down-regulation of protein synthesis during metabolic depression in the land snail *Helix aspersa*

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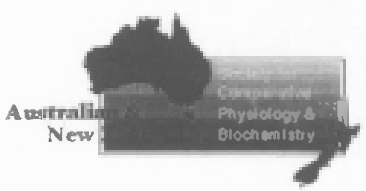
Metabolic depression represents a reduction in the basal metabolic rate of an animal in the order of 60-100% (1) and is employed as an adaptive mechanism in response to environmental stress. Metabolic depression is far from a rare phenomenon and has been reported in most animal phyla (2). The land snail *Helix aspersa* provides a convenient model for the study of metabolic depression as it readily enters a period of dormancy (aestivation) when deprived of food and water (in the absence of any temperature change). Associated with this dormancy is a high degree of metabolic depression, an 84% decrease in metabolic rate as measured by oxygen consumption (3). Protein synthesis is a major contributor to cell energy expenditure and thus ia thought to be one of the major energy utiliaing processes which must be down-regulated during metabolic depression. The aim of this study was to measure the rate of protein synthesis in active and aestivating snails in two major tissues *in vivo*, foot muscle and hepatopancreas (digestive gland). It was found that the rate of protein synthesis decreased by 86% and 82% respectively. Evidence from other systems suggests that the down-regulation of protein synthesis during metabolic depression is controlled at the level of initiation of translation. One of the most well characterised mechanisms of translational control is down-regulation by phosphorylation of the alpha subunit of eIF-2 (eukaryotic initiation factor 2). Therefore the phosphorylation state of eIF-2alpha is being investigated as well as kinase activity towards eIF-2alpha during both the active and aestivating states.

(1) Guppy et al. (1994). Comp. Biochem. Physiol. 109 B (2/3): 175-189; Guppy M. & Withers P. (1999) Metabolic depression in animals: Physiolgical perspectives and biochemical generalisations. Biol. Rev. in press; (3) Pedler et al. (1996). J. Comp. Physiol. B 166: 375-381.



Physiological precocity in an altricial parrot

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Parrots lay small eggs and hatch with the lowest degree of morphological and behavioural maturity. Previously one parrot species, *Agapornis roseicollis*, was shown to have a low degree of functional maturity during both the embryonic and posthatching periods. In this study the cockatiel, *Nymphicus hollandicus*, was found to have low functional maturity during incubation, *i.e.* a low hatchling dry mass fraction and low metabolic rates of embryos and hatchlings relative to that of predicted conspecific adults of similar masses, which is similar to most altricial birds. However, in spite of an absence of natal down, cockatiel chicks develop strong thermogenic powers within 4-5 d at small body masses, exceeding that of predicted adults at the same mass, much earlier in development than in other altricial species. The development of posthatching metabolism in this parrot resembles the patterns of small precocial species, emphasising that there is more diversity among altricial birds than recognised in even the most recent studies, and that there is continuous variation in developmental modes with regard to physiological maturity.

Ingestion of extract of leaves of mangosteen (*Garcinia mangostana* Linn) induces germ cell apoptosis in rat testis

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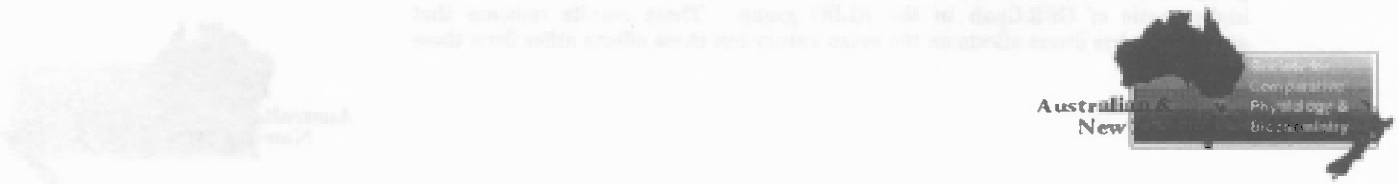
Indonesian women drink tea made from mangosteen as a contraceptive. This herbal medicine might also work in males because it decreases sperm production in rats (Riono *et al.*, unpublished). However, the mechanism is not clear. We tested whether mangosteen extract reduces germ cell numbers by promoting apoptosis, the controlled cell death that is characterized by cellular shrinkage, chromatin condensation, membrane blebbing and orderly DNA fragmentation. This process is thought to explain 75% of the normal loss of mature sperm cells in adult mammalian testis. Male rats were given either a control (vehicle) or treatment (a daily oral dose of a double methanol extract of mangosteen leaves, 100 mg/kg BWt) for 8 weeks. Five rats from each group were killed every 2 weeks and testicular tissue was removed and subjected to *in situ* 3'-end-labeling of internucleosomal DNA fragments. Apoptosis increased in extract-treated rats after 4 weeks of treatment, suggesting that reduction of sperm production by mangosteen extract is due to an increase in apoptosis, probably in the germ cells themselves. Further studies are needed to elucidate the active components of the extract and to test whether the extract directly induces apoptosis, or whether other factors are involved.



Ecological correlates of locomotor performance in geckos

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In many diurnal lizards, aspects of locomotor performance such as endurance and sprint speed, and their thermal dependencies, are often closely related to ecological or physiological traits such as foraging mode and the preferred body temperature for activity. In the few nocturnal lizards examined to-date, however, sprint performance seems to peak at body temperatures well above those experienced during routine activity and endurance capacities that are often assumed to be limiting. To further investigate these apparent differences, an examination of the inter-specific differences in endurance and sprint speed was undertaken for 5 species of sympatric nocturnal gecko and a diurnal agamid from the semi-arid region of Western Australia. Maximum sprint speeds and endurance times varied dramatically between the five species of gecko. Species thought to be exposed to the greatest risk of predation had the fastest sprint speeds, whereas those that moved most frequently while foraging had the greatest endurance. Body temperatures optimal for sprinting were higher in all geckos than those optimal for endurance. Peak sprint performance appeared to occur at body temperatures typically encountered during times of the greatest risk of predation i.e. during the daytime. Endurance peaked at the lower body temperatures typically experienced during nocturnal activity. In contrast to the geckos, sprint speed and endurance had similar thermal optima in the diurnal agamid, both coinciding with its daytime activity temperatures. In all species, sprint speed had a wider thermal performance breadth than endurance, possibly reflecting the importance of being able to escape from potential predators regardless of body temperature.

Aldosterone and renal function in avian species

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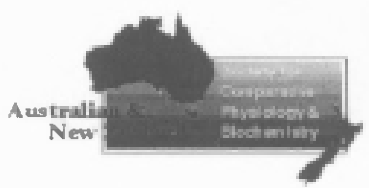
The role of aldosterone in avian kidney function has not been clearly demonstrated although the assumption has been that the action of aldosterone in avian species is similar to that in mammals. Ten adult roosters were used to investigate the effect of aldosterone infusion on plasma electrolyte composition and renal function. Birds were anaesthetised with DIAL and a carotid artery and superficial ulnar vein cannulated. The hindgut was blocked off and a tube inserted for collection of ureteral urine. The five birds comprising the control group (CONT) were infused at 0.2 mL/min/kg with a solution of isotonic saline containing 10 g/kg inulin and 10 g/kg PAH, for a total of 8.5 hours. The five roosters in the aldosterone treatmeat group (ALDO) were infused with the same solution except that aldosterone (125 ug/L) was added to the infusate after the initial 30 minutes of the experiment. Blood samples were collected hourly and urine samples every 15 minutes. Plasma and urine samples were analysed for inulin, PAH, osmolality, sodium (Na), potassium (K), chloride (Cl). Plasma samples were taken for aldosterone assay at four stages of the experiment. The plasma aldosterone concentrations achieved represented a high physiological level. Plasma K increased significantly, urine K was significantly lower and the urine:plasma ratio for K was lower in the ALDO group. An increased urine flow rate in the ALDO group was accompanied by lower urine osmolality and chloride and lower urine:plasma ratio for osmolality. Osmolar clearance and total osmolar output were significantly higher for the ALDO group. The total excretion rate and the fractional excretion of sodium were higher in the ALDO group throughout the experiment and there was a transient decrease about 45 minutes after the start of the aldosterone infusion. Glomerular filtration rate (GFR) was not significantly affected by aldosterone infusion whereas the clearance of PAH (Cpah - an indication of renal plasma flow) remained constant in the ALDO group at the same time as it increased in the CONT group. This resulted in a higher ratio of GFR:Cpah in the ALDO group. These results indicate that aldosterone has direct effects on the avian kidney but these effects differ from those in mammals.



Effect of aerial exposure on the western rock lobster, *Panulirus cygnus*

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Post-harvest operations play a critical role in determining the quality and value of live seafood products, such as the western rock lobster, *Panulirus cygnus*. Although lobsters should be kept in water as much as possible after harvest, lobsters do experience unavoidable episodes of emersion, as may occur during transport to processing facilities or to domestic or foreign markets. Given these considerations, this study sought to examine the effect of a 24 h period of emersion followed by a 48 h period of reimmersion, at 20°C, on the physiology of western rock lobsters.

The results revealed that, when exposed to air, both pre- and post-branchial PO_2 decreased and pre-branchial PCO_2 increased, indicating that gill function was compromised. A progressive increase in haemolymph lactate titre during emersion suggested an increased dependence on anaerobic metabolism when in air. The increase in lactate concentration and PCO_2 led to a mixed respiratory/metabolic acidosis and a fall in haemolymph pH. A classic hyperglycaemia was also observed. The results suggest that although a depression of behavioural responses to handling could be detected during emersion and following re-immersion, western rock lobsters can defend tissue energy levels anaerobically, with no change in adenylate energy charge (AEC) of abdominal muscle being recorded. Perturbations in the levels of potassium, magnesium and calcium in the haemolymph were recorded during both emersion and following re-immersion. Recovery of all parameters was complete within 8 h after re-immersion.

A cholinergically mediated adrenergic response in the heart of the horned shark, *Heterodontus portusjacksoni*

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The atypical excitatory effect of acetylcholine (Ach) on cardiac function was investigated in the horned shark, *Heterodontus portusjacksoni*. Electrically paced ventricular strips showed a striking 391 ± 26% increase on basal force of contraction as a response to exogenously applied Ach. The response was similar in nature to the response of electrically paced ventricular strips to adrenaline (Ad) which caused a 383 ± 72% increase. The response to Ad was faster than the response to Ach, especially over the first 10 mins after application. The response to Ach was blocked by the muscarinic antagonist, atropine and the β-adrenergic antagonist, propranolol, and was reduced by bretylium, an agent known to inhibit the release of catecholamine from adrenergic nerves. The evidence from this study suggests that Ach caused a localised release of a catecholamine via muscarinic receptors. A cholinergic-adreno complex has been proposed suggesting that elasmobranchs may be capable of finer control of cardiac output than was previously suspected. Investigations on the spontaneously beating atrium gave results in accordance with previous authors with no evidence of an excitatory response. Exogenously applied Ach caused the atrium to beat slower and less forcibly. Atropine blocked the response before and after it was initiated suggesting Ach was effecting the muscle directly via muscarinic receptors.

Comparing the metabolism of reptiles with varying body mass

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How useful are the generally accepted allometric exponents of 0.75 (interspecific) and 0.67 (intraspecific) when comparing the metabolism of reptiles of varying body mass? This paper will explore some of the problems and issues of comparing the standard metabolic rate of reptiles (specifically goannas and pythons) of different body mass. It will look at species which have an intraspecific mass exponent not approximately 0.67, genera where the interspecific mass exponent is not 0.75 and species where neonates don't appear to conform to the intraspecfic allometric relationship evident for adults. It will also address some related statistical issues.

The MO_2 and metabolic release of CO_2 from labeled glucose and lactate In the exercising ghost crab, *Ocypode ceratopththalmus*

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It is know that in ghost crabs the gills act as osmoregulatory organs and do not participate in the CO_2 and O_2 exchange process during respiration. To determine the efficiency of the relatively thin branchial lining as a functioning "lung", crabs were run on a treadmill for 20 minutes. During exercise the strong and weak points of the respiratory physiology of the crab could be determined. Before the run each crab was injected with radioactively labeled glucose or l-lactate. It was found that haemolymph l-lactic acid levels increased to 20 mmol L^{-1}; the heart rate increased from 100 beats per minute to nearly 400 beats per minute; MO_2 increased about five times above resting levels. The oxygen dissociation curves constructed for the blood from resting crabs and exercising individuals shows a large shift to the right. A linear increase of $^{14}CO_2$ from injected l-lactate indicates that this gas is nearly instantaneously released by the branchial lining or "lung". The same result was found when CO_2 was metabolically released from injected glucose. These findings, together with the MO_2 measurements gave evidence that the branchial lining has a gas exchange capacity similar to that of an ectothermic vertebrate

Gonadal hormones are proximate factors controlling seasonal fat metabolism in adult male emus.

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Mature male emus fed *ad libitum* show inversely related seasonal patterns of liveweight and sex hormones. Feed intake is lowest in winter but doubles by 5 weeks after the winter solstice, and reaches a maximum near the spring equinox. Liveweight is highest in summer, lowest 5 weeks after the winter solstice, but returns to summer values within 3 weeks of the spring equinox. Testosterone is detectable from 9 weeks before until 7 weeks after the winter solstice.

Mature male emus that had been castrated at 8 weeks of age ate similar amounts of feed to intact males in spring, summer and autumn. In winter, intake was maximum one week after the solstice and higher than for intact birds. Liveweight of castrated emus followed a similar pattern to intact birds, except that minimum values were observed in the week of the winter solstice. Castrates weighed less than intact birds, except in winter.

Early castration attenuated vernal fattening of male emus, while appetite was only slightly affected. The pattern of feed intake and fat gain and loss was shifted to earlier in the year for castrates. These results confirm for emus the interaction between photoperiod and gonadal steroids in the control of fat deposition and metabolism.

Prostaglandin $F_{2\alpha}$ receptors in the brushtail possum, *Trichosurus vulpecula* and the short-nosed bandicoot, *Isoodon macrourus*, at birth.

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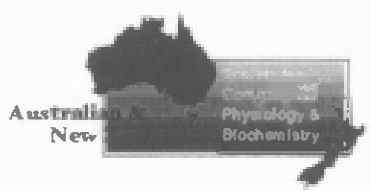
Prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) is involved with the regression of the corpus luteum (CL). Regression occurs at parturition in the possum but later in the bandicoot. The plasma concentration of 13, 14-dihydro-15-keto-prostglandin $F_{2\alpha}$ (PGFM), the major metabolite of $PGF_{2\alpha}$, is briefly elevated at birth in the possum and elevated for several days around birth in the bandicoot. In both species, $PGF_{2\alpha}$ injection induces adoption of the birth position. This study investigates $PGF_{2\alpha}$ receptors in the uterus and CL of possums and bandicoots at parturition. $PGF_{2\alpha}$ receptors are not present in the bandicoot and possum uteri and the bandicoot CL. $PGF_{2\alpha}$ does not appear to be involved with uterine contractions in the possum or the bandicoot at parturition. However, low concentrations of receptors were found in the possum CL, suggesting the involvement of $PGF_{2\alpha}$ in CL regression. In the bandicoot, there were no $PGF_{2\alpha}$ receptors in the CL at birth. This provides an explanation for the failure of $PGF_{2\alpha}$ administration to regress the bandicoot CL at birth. However, $PGF_{2\alpha}$ injections will cause regression of the CL at day 30 of lactation and therefore future studies will be directed to this post-natal period.



Overview of metabolic depression in ectotherms and endotherms

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Metabolic depression has been recorded for virtually all major animal phyla in response to environmental stress. An allometric analysis suggests three general patterns of metabolic depression. Firstly, a common pattern is metabolic depression to approximately 0.05-0.4 of rest *e.g.* molluscs, earthworms, crustaceans, fishes, amphibians, reptiles. This "intrinsic" depression is in anticipation of adverse environmental conditions but without substantial changes to their state of body water. Some animals survive anoxia for limited periods, with a similar depression to 0.05-0.4. The profound decrease in metabolic rate of torpid mammals and birds is mainly due to readjustment of thermoregulatory control and a decrease in body temperature with a concomitant Q_{10} effect, although there may be a modest intrinsic metabolic depression for deep torpor (to approximately 0.6). Secondly, a more extreme pattern of metabolic depression to <0.05 of rest is evident for cryptobiotic (anhydrobiotic, osmobiotic, or cryptobiotic) animals with a profound change in their ionic/osmotic balance or state of body water. A few normally-aerobic animals can have an anaerobic metabolic rate than 0.005 of resting. The metabolic rate of anhydrobiotic animals can be unmeasurable, if not zero. Thirdly, a pattern of indefinite absence of metabolism when anoxybiotic but normally hydrated is apparently unique to diapaused eggs of the brine-shrimp (*Artemia* spp., an anostracan crustacean).

Metabolic physiology of the marsupial mole (*Notoryctes caurinus*)

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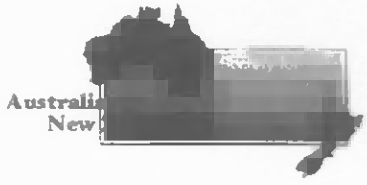
The marsupial mole (*Notoryctes*) is the most unusual of all Australian marsupials, essentially living entirely underground. It closely resembles the golden moles (placental family Chrysochloridae) such as the Namib Desert golden mole (*Eremitalpa grantii namibensis*), being an excellent example of convergence in morphology. Not only is the marsupial mole unusual, it is extremely rare and nothing is known of ita physiology, not even ita body temperature. The essentially completely fossorial habit of the marsupial mole, in contrast with the nocturnal surface-feeding of the Namib Desert golden mole, makes the marsupial mole extremely difficult to locate for study.

We report that the marsupial mole (*N. caurinus*) has a low and labile body temperature, as expected for a fossorial mammal, but does not have a substantially lower-than-predicted BMR. Thermal conductance is high, and variable. The metabolic cost of burrowing through dry sand is very high, as expected, compared to the metabolic cost of terrestrial (running) locomotion. The kidney of the marsupial mole has only a moderate medullary area, suggesting a mesic- rather than arid-adapted water balance, reflecting the high water content of the food. With respect to ita physiology, the marsupial mole is also remarkably convergent with the Namib Desert golden mole.



POSTERS

Corticosterone response of captive northern brown kiwi (*Apteryx australis mantelli*) to an acute stressor

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Corticosterone is the main adrenocortical hormone released during stress in birds, and plasma corticosterone levels usually rise rapidly after a bird experiences a stressor. In this study we examined the corticosterone response of six captive northern brown kiwi to an acute stressor (handling) by collecting blood samples from kiwi 3 times over one hour and then once more, either 3, 5 or 7 hours after the kiwi had initially been handled. All of the kiwi responded to the acute stressor with a rise in corticosterone levels. Plasma corticosterone levels increased markedly to a peak 60 min after handling then declined and remained above initial levels from 3 to 7 hours after handling. Corticosterone levels were higher than usual for birds. Results from this preliminary study also suggest that there may be a difference between the corticosterone responses of nocturnal housed and outdoor housed captive kiwi.

The use of dataloggers to determine behavioural activity in the platypus (*Ornithorhynchus anatinus*)

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Studies into the behavioural activity pattern of the platypus have been restricted in the past on visual observation or radio tracking techniques. To examine platypus behaviour in the wild more closely, in this study animals were equipped with dataloggers (Minimitter Co., Inc., USA) measuring activity in epoch lengths from 2 sec to 5 min by using an accelerometer sensitive to <.05 g, integrating degree and intensity of motion. Over the last 9 months 18 individuals have been equipped with combined datalogger-transmitter packages (weight 40 g) in a subalpine lake in northwest Tasmania. Animals were recaptured and data retrieved after three to five weeks. Preliminary results indicate a high variability in activity. About 40% of animals foraged preferentially over the day while 60% preferred night time. 50% of all platypuses showed irregular activity patterns with some animals changing start and end time of their active period within days and foraging up to 20 hours without resting. On average foraging trips lasted for 13 hours daily. Mean dive times of three individuals were between 12 and 66 seconds with 47% of all dives lasting between 30 to 40 seconds. Animals performed up to 800 dives per day.

Intracerebral infusion of recombinant leptin inhibits LH secretion in male sheep by decreasing feed intake

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We have previously observed that infusion of leptin reduces both LH pulse frequency and feed intake in mature Merino rams. We therefore infused bovine leptin into the third cerebral ventricle of rams fed *ad libitum* to investigate whether the inhibition of LH secretion is a direct effect of leptin or is due to a decrease in the availability of energy substrates. Seventeen rams were randomly allocated to treatment with artificial cerebrospinal fluid (aCSF) or 0.4 μg/h bovine leptin in

aCSF. The effect of decreased food intake on LH secretion was controlled by pair-feeding a third group to the leptin-infused group. The infusions were continuous for 24 h for five days. Blood plasma sampled every 20 min for 24 h on days 1 and 5 of the treatment period was assayed for LH. Leptin infusion decreased feed intake compared to the control group ($P<0.05$). Five days of icv leptin infusion and pair-feeding decreased LH pulse frequency compared to control values ($P<0.01$). Leptin decrease feed intake and thus energy substrates, and this reduces LH secretion. It seems unlikely that leptin directly inhibits reproduction.

Myofibrillar protein composition of regenerating and pristine claw closer muscle of the yabby, *Cherax albidus*.

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Yabbies have the ability to regenerate lost limbs. This involves many developing stages until the new limb becomes morphologically and physiologically virtually indistinguishable from its predecessor. In the yabby, the claw becomes functional after the first moult when the dactyl becomes moveable. However, the muscle fibres appear to be at least partly functional already before this (West *et al.*, 1995). Crustacean muscles are composed of two major fibre types that differ in ultrastructure, contractile properties and protein composition. Fast fibres have short sarcomeres and low fatigue resistance, whereas slow fibres have long sarcomeres and high fatigue resistance. In addition, each fibre type has a unique myofibrillar protein profile. Muscle fibres from regenerating claws of *Cherax destructor* have previously been characterised by ultrastructure, sarcomere length and activation properties (West *et al.*, 1995). Muscle fibres isolated from regenerating claws with a fused dactyl had ultrastructural and activation properties characteristic of fast fibres with a sarcomere length typical of slow fibres. In the present study, the myofibrillar protein compositions of pristine and regenerating claw muscle fibres of the yabby were used to document muscle fibre regeneration and to compare pristine with regenerated muscle fibres.

West J M, Humphries D C, & Stephenson G (1995) Characterisation of ultrastructural and contractile activation properties of crustacean (Cherax destructor) muscle fibres during claw regeneration and moulting. J. Muscle Res. Cell. Motil. 16, 267-284.

Anaerobic metabolism in the intertidal elephant snail, *Scutus antipodes* (Fissurellidae)

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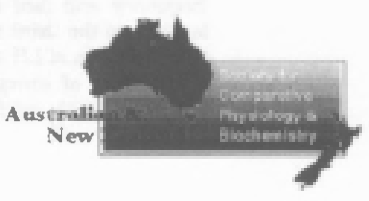
The elephant snail, *Scutus antipodes*, is a common gastropod of the intertidal zone on south-eastern Australian rocky shores. It occupies a similar ecological niche to the abalone (Haliotidae) with which it is grouped in the Archaeogastropoda. Abalone display well developed capacities for anaerobic metabolism during both exercise induced and environmental hypoxia, producing D-Lactate and the unusual pyruvate reductase end product tauropine. Given the phylogenetic, ecological and behavioural similarities of *S. antipodes* and abalone it was considered of interest to compare their anaerobic capabilities.

Tauropine dehydrogenase was the only pyruvate reductase enzyme detected in *S. antipodes* foot muscle, and tauropine accumulated during both air exposure (2-6 h) and enforced exercise (shell righting behaviour). These hypoxic stresses were accompanied by decreased arginine phosphate reserves. In comparison with abalone, the lower pyruvate reductase activities and intracellular pH buffering of *S. antipodes* foot musculature imply a more limited capacity for anaerobic metabolism in these hypoxic situations



An investigation of the factors causing the population decline of the swift parrot

B. D. Gartrell[1], S. M. Jones[1], R. N. Brereton[2] and L. B. Astheimer[3]
[1]School of Zoology, University of Tasmania, GPO Box 252-05, Hobart TAS 7001; [2]Department of Primary Industries, Water and Environment, Hobart TAS 7001; [3]Department of Biomedical Science, University of Wollongong, Wollongong, NSW 2522
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The swift parrot (*Lathamus discolor*) is a small migratory parrot, which breeds in the eucalypt forests of Southeast Tasmania. Its status has been recommended to be upgraded to "endangered" as there are less than 1000 breeding pairs in the wild. The main reasons for its decline include fragmentation and loss of foraging and nesting habitat, fluctuations in breeding success associated with years of poor flowering of the blue gum (*Eucalyptus globulus*) and finally, increased collision mortality.

This poster outlines a doctoral project with the aim to investigate those factors, which are suspected to be responsible for the swift parrot population decline. The importance of understanding the mechanisms of population decline in regard to conservation efforts and the rationale of the research plan as it relates to these mechanisms are outlined. At the conclusion of the project, the goal is to have the first detailed study of the reproductive biology and endocrinology of a threatened parrot in Australia. This will support the goals of the Swift Parrot Recovery Plan, to increase the breeding population and to preserve appropriate habitat of the swift parrot. In a broader context the project will develop investigative techniques with importance to other avian conservation programmes.

Energy expenditure and water uptake in free-ranging and captive blossom-bats, *Syconycteris australis* (Megachiroptera)

Fritz Geiser and Dionne K. Coburn
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Blossom-bats, *Syconycteris australis* (18 g) are known to be extremely active throughout the night. Since they frequently enter daily torpor, we wondered whether this may be related to high energy expenditure in the field. We measured field metabolic rates (FMR) of bats at a subtropical site using the doubly labelled water method. Captive bats at constant ambient temperature (T_a) and with food ad libitum were measured for comparison. The FMR of *S. australis* of 8.55 ml CO_2/g.h or 76.87 kJ/d (7.04-fold of basal metabolic rate, BMR) was one of the highest values measured in endotherms to date and may be why these bats frequently enter torpor when at rest. Daily water uptake in the field was about 1.8-fold of body mass. Use of energy in captive bats was about $^2/_3$ that of FMR. This suggests that foraging times in the field and laboratory were similar, and that daily energy expenditure was not strongly affected by T_a or food availability. Our study shows that *S. australis* has a FMR that is about 2-fold of that predicted for its size although their BMR is lower than predicted. Thus, extrapolations from BMR about energy requirements in free-ranging animals must be made with caution.

Effect of temperature and pH on motility and velocity of emu spermatozoa

Lena Holm[1], Irek A. Malecki[2] and Graeme B. Martin[2]
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imalecki@agric.uwa.edu.au

Chicken spermatozoa need to be motile to reach the sperm storage tubules in the oviduct and to leave the tubules to fertilise the eggs. During storage, however, the spermatozoa are immotile. The mechanisms responsible for inactivation and

reactivation are not clear but a change in pH has been suggested, since only small changes in pH are required to stimulate or inhibit the motility of spermatozoa from several avian species. The effect of pH on emu spermatozoa was studied in semen from 4 males. Velocity (μm/s) and % motile spermatozoa were measured using digitised image analysis (about 100 sperm per experimental condition) in four media: MES (pH 6.0), TES (pH 7.0), TRIZMA (pH 8.0), and CAPSO (pH 9.0). At 20°C, motility was high in all media (pH 6.0-pH9.0) but the velocity was low. At 40°C, spermatozoa showed high motility and high velocity at pH 6.0 - pH 8.0. At pH 9.0, however, both measures were markedly reduced, although the sperm remained motile. These observations contrast with those for spermatozoa from the chicken, turkey and quail, which are inhibited at 40°C and pH 6.0-7.0 but stimulated at pH 9.0. The optimal pH range for spermatozoa may differ between the emu and other species.

The effects of moulting on muscle fibre characteristics of the yabby, *Cherax albidus*

T. Lamey and A.E. Patak
School of Natural Sciences, Edith Cowan University, 2 Bradford Street, Joondalup, WA 6050
a.patak@cowan.edu.au

Moulting has a profound effect on the growth of crustaceans. Growth is cyclic with relatively long times of physiological stasis punctuated by short periods of rapid growth at moulting. The claw muscle is particularly affected. It undergoes extensive atrophy (up to 60% of muscle mass) to enable withdrawal through the narrow base whilst remaining functional. In contrast, the large abdominal muscle does not undergo atrophy during moulting. Crustacean muscles have two major fibre types that have previously been identified in *Cherax destructor* claw muscle; slow contracting fibres with long sarcomeres, and fast contracting fibres with short sarcomeres. All muscle fibre types are involved in moult-induced atrophy as well as moulting-associated fibre elongation, although different mechanisms appear to operate in each fibre type. In the present study, histochemical staining techniques have been developed to demonstrate contractile speed, fatigue resistance and fibre size of yabby muscle fibres. These techniques were used to determine the occurrence and distribution of fibre types and sub-types in the claw and abdomen during intermoult and to compare the fibre characteristics of moult-induced atrophic claw muscle with non-atrophic abdominal muscle over the moult cycle.

Respiratory responses of intertidal crab eggs to changes in oxygen tensions

N. Leelapiyanart[1] and H. H. Taylor[2]
[1]*Department of Aquatic Science, Burapha University, Chonburi, Thailand;*
[2]*Department of Zoology, University of Canterbury, Christchurch , New Zealand*
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In response to declining oxygen tension, pre-hatching eggs of *H. rotundifrons* and *C. lavauxi* exhibited characteristics of oxygen conformers and regulators respectively. The P_{crit} value for late stage eggs of *C. lavauxi* was approximately 6 kPa (45 mm Hg). The lactate concentration in eggs of *H. rotundifrons* increased from about 0.602 mmol.l^{-1} in normoxic eggs to about 10.20 mmol.l^{-1} after 3h hypoxic exposure. The rate of lactate accumulation during hypoxia suggests that the eggs are not sustaining metabolism by anaerobiosis, but are allowing metabolic depression. Pre-hatching eggs of *H. rotundifrons* and *C. lavauxi* exhibit no ventilatory or locomotory movements, and limited circulatory function. We hypothesize that the greater dependence of oxygen uptake on ambient PO_2 in the larger *H. rotundifrons* eggs reflects a greater diffusion limitation. This interpretation is supported by calculations of PO_2 gradients in a simplified model. This model showed that the centre of *Heterozius* eggs becomes anoxic at quite moderate external hypoxia, thus limiting total oxygen uptake. For *Cyclograpsus*, the centre does not become anoxic until severe external hypoxia prevails.

An investigation of the factors causing the population decline of the swift parrot

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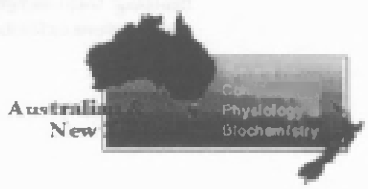

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Acetylcholine controls surfactant secretion by type II pneumocytes isolated from Australian lungfish and American bullfrogs

Olga V. Lopatko[1], Philip G. Wood[1], Jean M. P. Joss[2], Allan W. Smits[3] and Christopher B. Daniels[1].

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The factors that control the release of pulmonary surfactant (PS) in non-mammalian vertebrates are largely unknown. In mammals, changes in ventilatory pattern and input from the sympathetic nervous system predominantly control surfactant secretion, while the parasympathetic nervous system has no effect. However, we have found that alterations in ventilatory pattern have no affect on surfactant release in lizards, but both the sympathetic and parasympathetic nervous systems stimulate surfactant phospholipid (PL) release. We examined the effects of the major autonomic neurotransmitters on PL release by type II pneumocytes isolated from lungs of the Australian lungfish, *Neoceratodus forsteri*, and the North American bullfrog, *Rana catesbiana*. Type II cells were isolated from each species by 'panning' whole lung cell suspensions over IgG coated bacteriologic plates. Cells were cultured for 15h in phosphate-free DMEM and PL secretion monitored in response to the application of adrenaline (AD, 100 μM) or acetylcholine (ACh, 100 μM). Only ACh stimulated PL release from lungfish type II cells (an increase of 14%), but both Ad and ACh stimulated PL release from isolated bullfrog type II cells (AD: 108% increase; ACh: 106% increase). Application of atropine blocked the ACh - elicited release of PL in both species. The Ad-stimulated release of PL from bullfrog pneumocytes was abolished by the presence of alprenolol. ACh also triggered the differential release of disaturated PL from lungfish and bullfrog type II cells. It appears that, *in vivo*, acetylcholine, and therefore the parasympathetic nervous system, may regulate the surfactant system in non-mammalian vertebrates, controlling surfactant release and composition. Adrenergic influences on the surfactant system may have developed subsequent to the radiation of the tetrapods.

Funded by the Australian Research Council and University of Adelaide and George Murray Overseas Travel Grants.

Development of the pulmonary surfactant system in a bird and reptile

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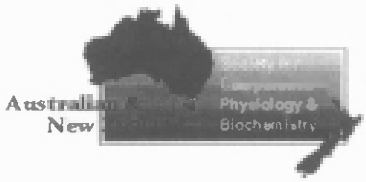
Little is known about the maturation of the pulmonary surfactant system in non-mammalian vertebrates. Unlike mammals, birds and reptiles hatch from eggs; often a lengthy and exhausting process whereby the animal commences lung clearance and pulmonary ventilation by "pipping" through the shell membranes. We examined the composition of pulmonary surfactant in the developing lungs of the chicken, *Gallus gallus*, and the bearded dragon, *Pogona vitticeps*. Lung tissue was collected from chicken embryos at day 14, 16, 18 (pre-pip) and 20 (post-pip) of incubation and from 1 day post-hatch, 3 week and adult animals. SPA mRNA was detected in lung tissue from chickens of all stages using Northern blot analysis. Chickens were lavaged at days 16, 18 (pre-pipped) and 20 (post-pipped) of incubation and 1 day post-hatch. Bearded dragons were lavaged at day 55 (pre-pipped), days 57-60 (post-pipped) and days 58-61 (post hatched). Total phospholipid (PL), disaturated phospholipid (DSP) and free cholesterol (CHOL) were measured in lavage fluid of both species. Total PL increased throughout incubation in both species (day 16 chick: 0.26± 0.09 μg/mg dry lung weight (dlwt), n=4 to hatch: 0.93 ± 0.24 μg/mg dlwt, n=9) and (day 55 bearded dragon: 0.39± 0.08 μg/mg dlwt, n=4 to hatch: 4.42 ± 0.84 μg/mg dlwt, n=7). DSP was not measurable at day 16 of incubation in the chick embryo. However the percentage of DSP/PL increased from day 18 (pre-pipped) to day 20 (post-pipped). As both PL and DSP increased with development, but CHOL remained unchanged during incubation in the chicken embryo, the CHOL to PL and CHOL to DSP ratios decreased in chick embryos respectively: (day 16: 39.53 ± 6.72%, n=4 to hatch: 8.61 ± 0.55% n=9) and (day 18: 146.90 ± 50.04%, n=8 to hatch 35.64 ± 4.91%, n=9). This indicates that the CHOL and PL components are differentially regulated. The CHOL to PL ratio is higher in embryonic chickens than any other animal studied to date. The changes in lipid composition during development of the pulmonary surfactant system in the chicken and bearded dragon mimic those of the mammal despite the difference in birth strategy.

Supported by the Australian Research Council



Hyperlipaemic emu serum is clarified for radioimmunoassay by polyethyleneglycol (PEG) precipitation

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Emu serum collected during spring is often hyperlipaemic, and females laying in winter have high circulating concentrations of yolk proteins. The lipid or vitellogenin content in serum prevents accurate radioimmunoassay. Serum lipids are sequestered within lipoproteins, so we have adopted a method developed for analysis of lipoproteins to clarify samples. A solution of PEG (45% in water) is added to samples to precipitate large molecular weight proteins by steric exclusion. Centrifugation separates clarified serum from the lipoprotein precipitate. PEG treatment clarified 97% of emu serum samples (n=500) collected from May to October. The nature of the precipitate varied according to sex and season. We identified four types of precipitate: 1) thin, cohesive (48%); 2) floating, fatty (16%; 67% male); 3) large volume, loose (26%; 96% from breeding-season females); 4) floating fatty layer and thin precipitate (5%). Samples which were not clarified (4%) were from egg-laying females. Of type 2 precipitates, 71% were found in October (non-breeding season). Residuals after PEG precipitation of emu serum spiked with tracer-conjugated hormones were 87, 89, 93, 100, and 100% for insulin, LH, glucagon, prolactin, and testosterone, respectively. Hyperlipaemic samples being prepared for radioimmunoassay can be clarified with minimal hormone loss using PEG recipitation.

Diet and assimilation in the robber crab, *Birgus latro* (L.)

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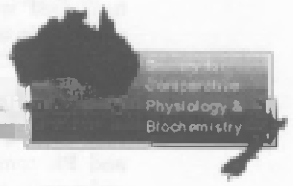
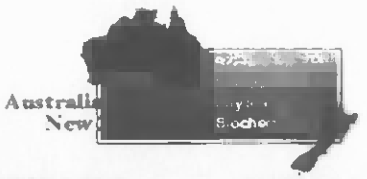
From observations of the natural diet on Christmas Island, it was clear that *Birgus latro* principally ate fruits and seeds rich in storage carbohydrates (*Pandanus*, *Ochrosia ackeringae*) and lipids (*Aleurites moluccana*, *Calophyllum inophyllum*, *Arenga listeri*) but also predated on other crabs and scavenged animal carcases. The capacity to assimilate the major components of these diets was assessed in the laboratory using 3 artificial diets rich in plant polysaccharides, plant lipids and crab tissue respectively. Assimilation of dry matter, carbohydrate and lipids were high on all diets. Assimilation efficiencies (AE's) of dry matter were similar for the three diets (71.7 ± 0.7%, 75.5 ± 1.3% and 64.7 ± 5.1%, for carbohydrate, fat and creb diets respectively), but AE's of lipid were significantly different (87.4 ± 1.7%, 96.03 ± 0.7% and 70.8 ± 3.7%, respectively). Near complete assimilation of polysaccharides occurred in the carbohydrate and fat diets (98.9 ± 0.2% and 98.06 ± .07%, respectively) but assimilation was significantly lower in the crab diet (89.4 ± 4.5%). Dry matter intake was affected inversely by the N concentration of the diets and was lowest (0.86 ± 0.08 $g.kg^{-1}.day^{-1}$) on the crab diet (5.4 mmol $N.g^{-1}$), intermediate (99 ± 0.61 $g.kg^{-1}.day^{-1}$) on the fat diet (1.2 mmol $N.g^{-1}$) and highest (4.31 ± 0.52 $g^{-1}.day^{-1}$) on the carbohydrate diet (0.9 mmol $N.g^{-1}$). Fibre was a relatively small component of the carbohydrate and fat diets but assimilation of hemicellulose (49.5 ± 0% and 64.8 ± 2.9%, respectively) and cellulose (16.1 ± 4.3% and 53.9 ± 3.5%, respectively) was quite high. The rate of passage, measured with particulate markers, was slow with a mean retention time of 27.3 ± 7.22 h but as defecation was infrequent this value may be overestimated.





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Weekdays

Route No.		Cottesloe	Claremont	Crawley	Busport
72	am	5:45	5:59	6:06	6:12
72		6:20	6:34	6:41	6:47
71		6:40	6:54	7:01	7:07
71		-	-	-	7:27
71		7:03	7:18	7:26	7:37
72		-	-	-	7:47
70		7:15	7:31	7:39	7:52G
72		-	-	-	7:57
70		7:25	7:41	7:49	8:02G
72		-	-	-	8:07
70		7:35	7:51	7:59	8:12G
72		-	-	-	8:17
70		7:47	8:03	8:11	8:24G
71		7:59	8:15	8:23	8:36
72		8:11	8:28	8:36	8:49
71		8:33	8:48	8:56	9:09
71		8:53	9:08	9:16	9:29
73		-	-	-	9:40
71		9:25	9:40	9:48	9:59
73		-	-	-	10:20
71		9:55	10:10	10:18	10:29
71		10:25	10:40	10:48	10:59
73		-	-	-	11:00
72		10:55	11:10	11:18	11:29
73		-	-	-	11:40
71		11:25	11:40	11:48	11:59
73		-	-	-	12:20
71	pm	11:55	12:10	12:18	12:29
71		12:25	12:40	12:48	12:59
73		-	-	-	1:10
71		12:55	1:10	1:18	1:29
72		1:25	1:40	1:48	1:59
73		-	-	-	2:15
71		1:55	2:10	2:18	2:29
72		2:24	2:40	2:48	2:59
73		-	-	-	3:12
71		2:54	3:10	3:18	3:29
73		-	-	-	3:42
72		3:19	3:35	3:43	3:54
72		3:29B	3:45	3:53	4:04
71		-	-	4:08J	4:19
73		-	-	-	4:22
71		3:59	4:15	4:23	4:34
71		-	-	4:38J	4:49
73		-	-	-	4:52
72		4:30	4:45	4:53	5:04
71		4:40	4:55	5:03	5:14
73		-	-	-	5:22
72		4:52	5:07	5:15	5:26
72		5:10	5:25	5:33	5:44
73		-	-	-	5:57
72		5:40	5:55	6:03	6:12
72		5:55	6:10	6:18	6:27
72		6:15	6:30	6:38	6:47
72		6:50	7:04	7:11	7:17
72		7:20	7:34	7:41	7:47
72		8:00	8:14	8:21	8:27
72		-	-	9:06J	9:12
72		9:58	10:12	10:19	10:25
72		10:35	10:49	10:56	11:02
72		11:13	11:27	11:34	11:40

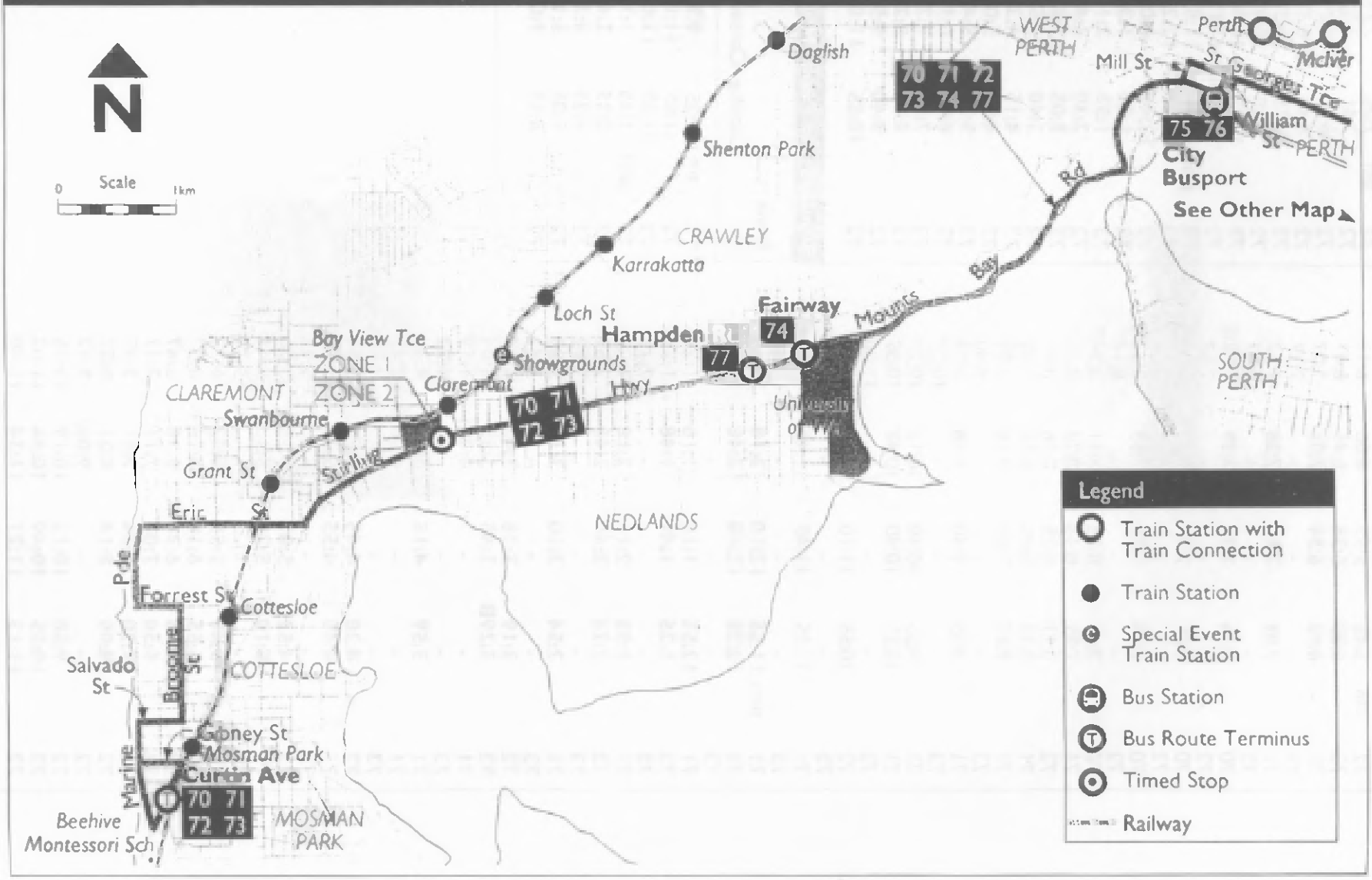
Saturdays

Route No.		Cottesloe	Claremont	Crawley	Busport
72	am	7:05	7:19	7:26	7:32
72		7:41	7:55	8:02	8:08
72		8:10	8:24	8:31	8:37
72		8:40	8:55	9:03	9:12
73		9:15	9:30	9:38	9:47
72		9:50	10:05	10:13	10:22
73		10:25	10:40	10:48	10:57
72		11:00	11:15	11:23	11:32
73	pm	11:35	11:50	11:58	12:07
72		12:10	12:25	12:33	12:42
73		12:45	1:00	1:08	1:17
72		1:20	1:35	1:43	1:52
73		1:55	2:10	2:18	2:27
72		2:30	2:45	2:53	3:02
73		3:05	3:20	3:28	3:37
72		3:40	3:55	4:03	4:12
73		4:15	4:30	4:38	4:47
72		4:50	5:05	5:13	5:22
72		5:25	5:40	5:48	5:57
72		6:00	6:15	6:23	6:32
72		7:10	7:24	7:31	7:37
72		8:15	8:29	8:36	8:42
72		9:40	9:54	10:01	10:07
72		10:55	11:09	11:16	11:22

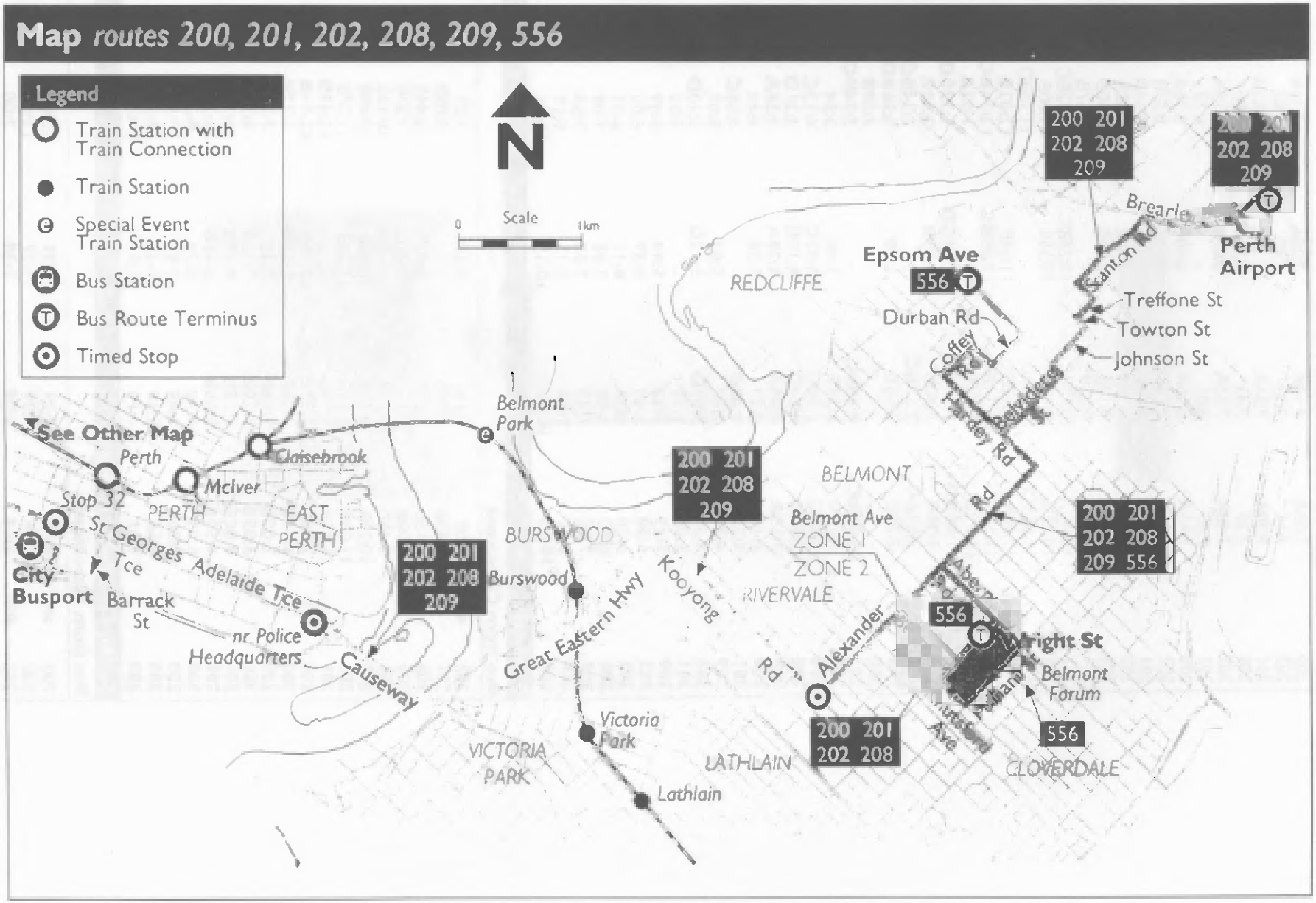
Sundays and Public Holidays

Route No.		Cottesloe	Claremont	Crawley	Busport
72	am	9:25	9:39	9:46	10:00
72		11:05	11:19	11:26	11:32
72		11:50	12:04	12:11	12:17
72	pm	1:25	1:39	1:46	2:00
72		2:35	2:49	2:56	3:02
72		4:25	4:39	4:46	5:00
72		5:05	5:19	5:26	5:32
72		6:55	7:09	7:16	7:30

Map *routes 70, 71, 72, 73, 74, 75, 76, 77*



Map *routes 200, 201, 202, 208, 209, 556*



Bus from Claremont

Route No.		Claremont	Nedlands	West Perth	Perth	Domestic Airport
209		7:00A	7:10A	7:20A	7:36A	-
202		7:15	7:33	7:42	7:47	8:28
201		7:20A	7:36A	-	7:53A	-
209		7:30	7:43	7:55	8:00	8:41
202		7:40A	7:58A	8.07A	8:19A	-
201		7:45	8:01	-	8:17	9:02
209		7:55A	8:08A	8:20A	8:38A	-
202		8:05B	8:23B	8:32B	8:37B	9:19
201		8:15A	8:31A	-	8:53A	-
202		8:30B	8:45B	8:56B	9:02B	9:42
208		8:55B	9:10B	9:25B	9:30B	10:12
201		9:25B	9:41B	-	9:57B	10:42
202		10:00BC	10:15BC	10:26BC	10:32BC	11:18
208		10:25B	10:40B	10:55B	11:00B	11:42
201		11:02BD	11:18BD	-	11:34BD	12:23
202		11:30B	11:45B	11:56B	12:02B	12:42
208		11:59BC	12:14BC	12:29BC	12:34BC	1:20
201	pm	12:30B	12:46B	-	1:02B	1:47
202		1:00BD	1:15BD	1:26BD	1:32BD	2:18
208		1:25B	1:40B	1:55B	2:00B	2:42
201		2:02BC	2:18BC	-	2:34BC	3:23
202		2:20E	2:35E	2:50	2:56BD	3:42
200		-	-	-	3:20B	4:04
201		3:02BC	3:18BC	-	3:34BC	4:25
202		3:18E	3:33E	3:48	3:54	4:33
208		3:35C	3:50C	4:05C	4:12C	4:57
202		3:49D	4:04D	4:15D	4:21D	5:06
209		3:55A	4:08A	4:20A	4:36A	-
202		4:12	4:27	4:38	4:44	5:25
201		4:26C	4:42C	-	4:58C	5:46
209		4:39	4:52	5:04	5:09	5:50
202		4:46D	5:01D	5:12D	5:18D	6:03
201		5:00	5:16	-	5:32	6:14
209		5:19	5:32	5:44	5:49	6:28
202		5:30	5:48	5:57	6:02	6:41
202		6:08	6:23	6:31	6:35	7:10
200		6:40	6:55	7:05	7:09	7:44
200		7:25	7:40	7:50	7:54	8:29
200		8:50	9:05	9:15	9:19	9:54
200		10:10	10:25	10:35	10:39	11:14

Saturdays

Bus from Claremont

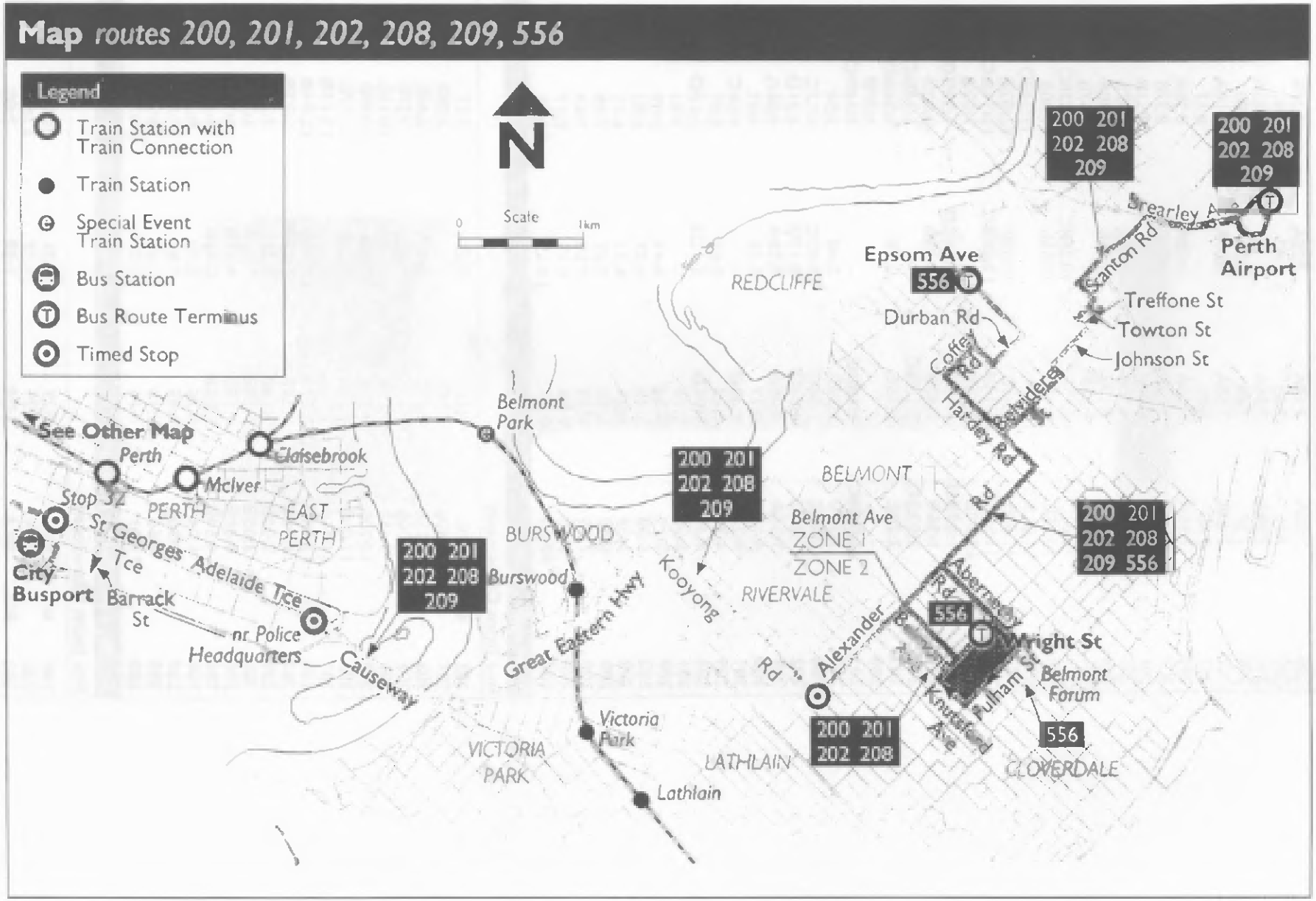
202	am	7:00	7:15	7:23	7:27	8:02
201		7:40B	7:54B	-	8:08B	8:47
208		8:15B	8:27B	8:40B	8:44B	9:18
202		9:00B	9:15B	9:23B	9:27B	10:05
201		9:45B	9:59B	-	10:13B	10:52
208		10:20B	10:32B	10:45B	10:49B	11:23
202		11:00B	11:15B	11:23B	11:27B	12:05
201		11:45B	11:59B	-	12:13B	12:52
208	pm	12:25B	12:37B	12:50B	12:54B	1:28
202		1:10B	1:25B	1:33B	1:37B	2:15
200		2:00B	2:15B	2:25B	2:29B	3:07
200		3:05B	3:20B	3:30B	3:34B	4:12
200		4:00B	4:15B	4:25B	4:29B	5:07
200		4:50B	5:05B	5:15B	5:19B	5:57
200		6:05	6:20	6:30	6:34	7:09
200		7:30	7:45	7:55	7:59	8:34
200		9:55	10:10	10:20	10:24	10:59
200		11:01	11:16	11:26	11:30	12:05

Sundays and Public Holidays

Bus from Claremont

200	am	9:14	9:29	9:39	10:00	10:35
200		11:14	11:29	11:39	12:00	12:35
200	pm	1:14	1:29	1:39	2:00	2:35
200		2:49	3:04	3:14	3:18	3:53
200		4:14	4:29	4:39	5:00	5:35
200		-	-	-	6:02	6:37
200		6:44	6:59	7:09	7:30	8:05

Map *routes 200, 201, 202, 208, 209, 556*



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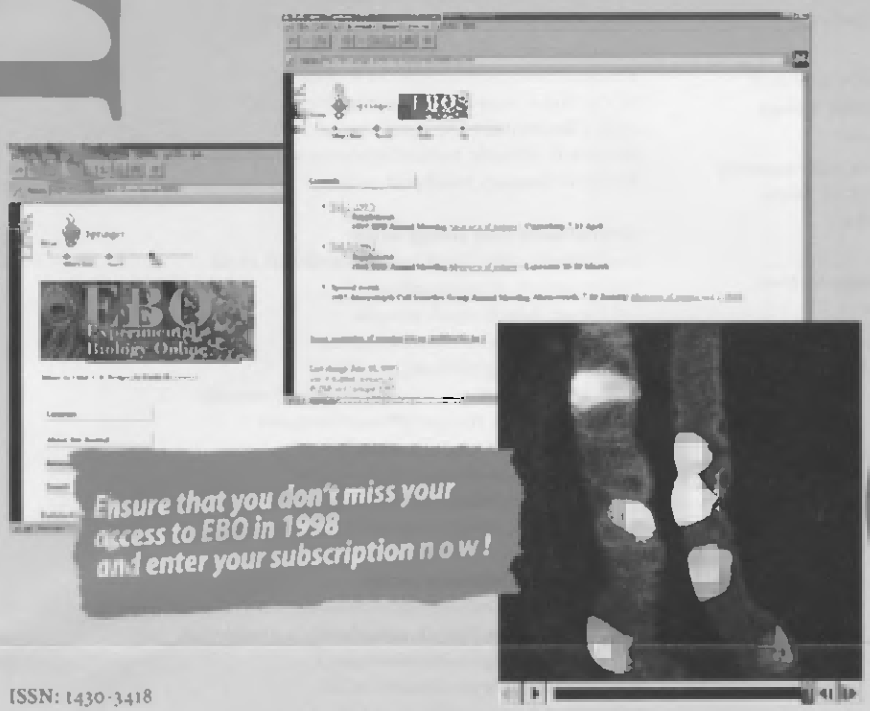

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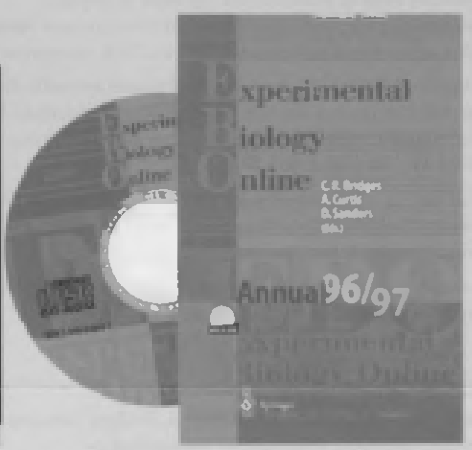

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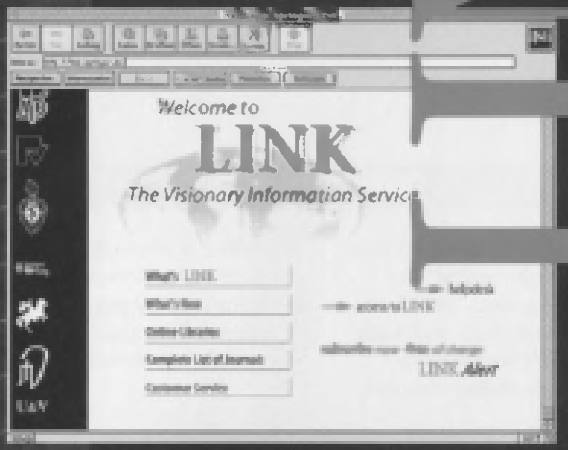

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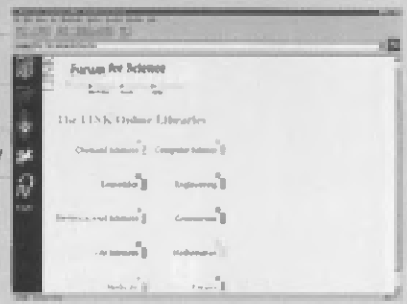

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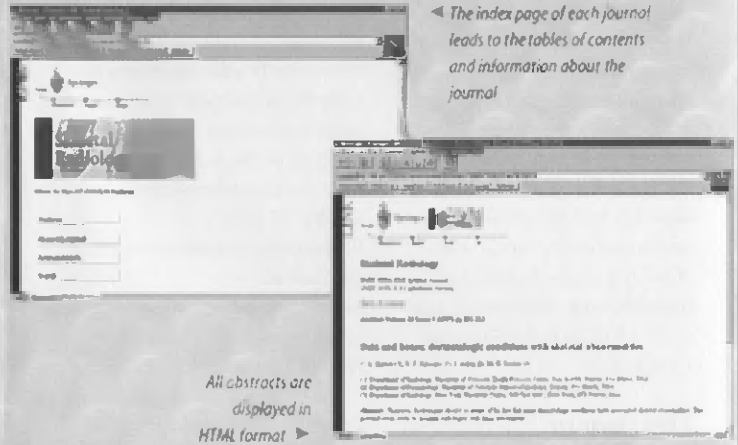

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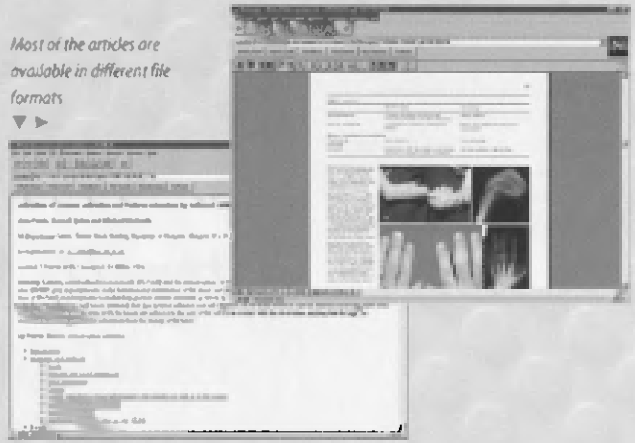

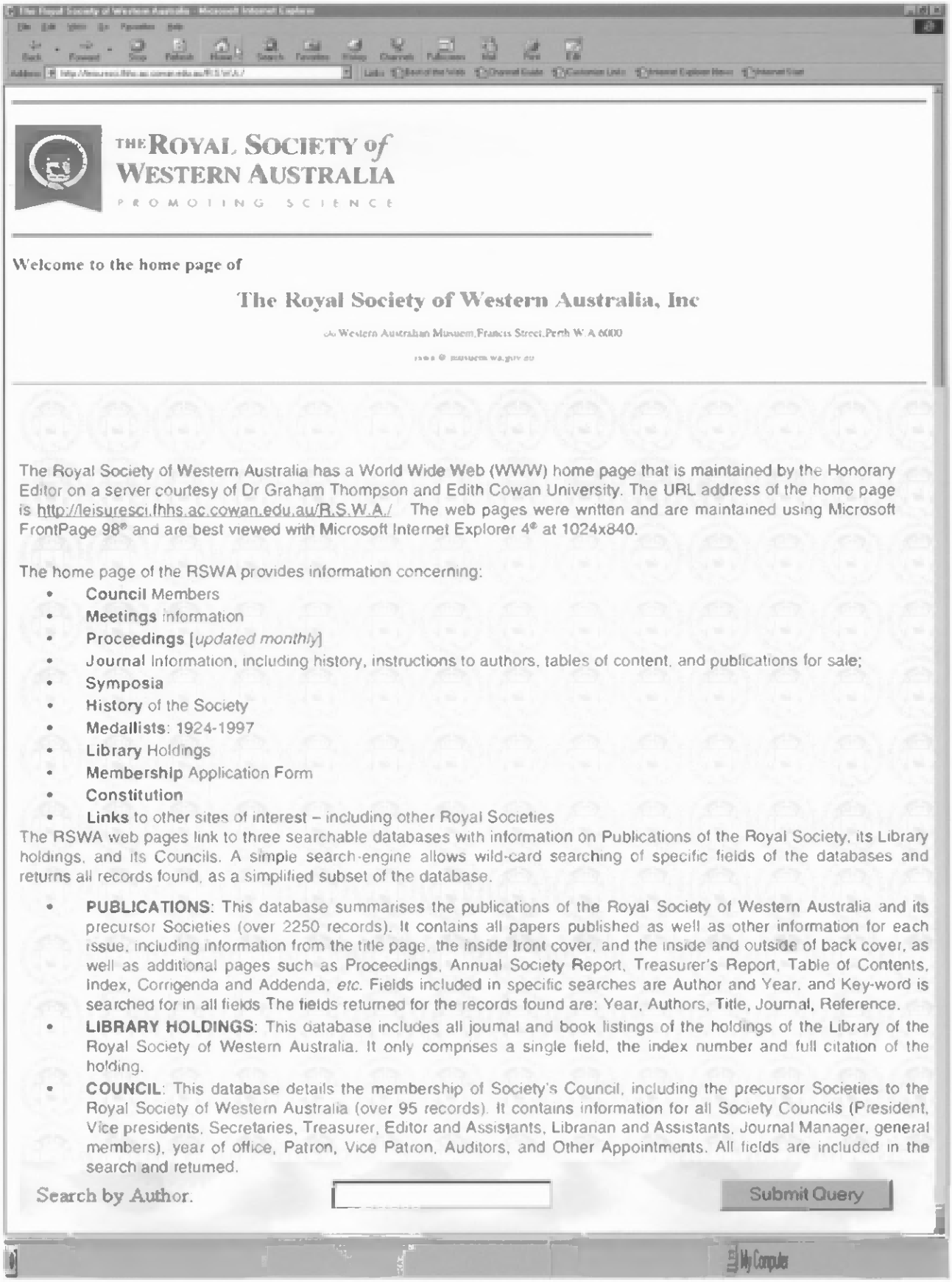

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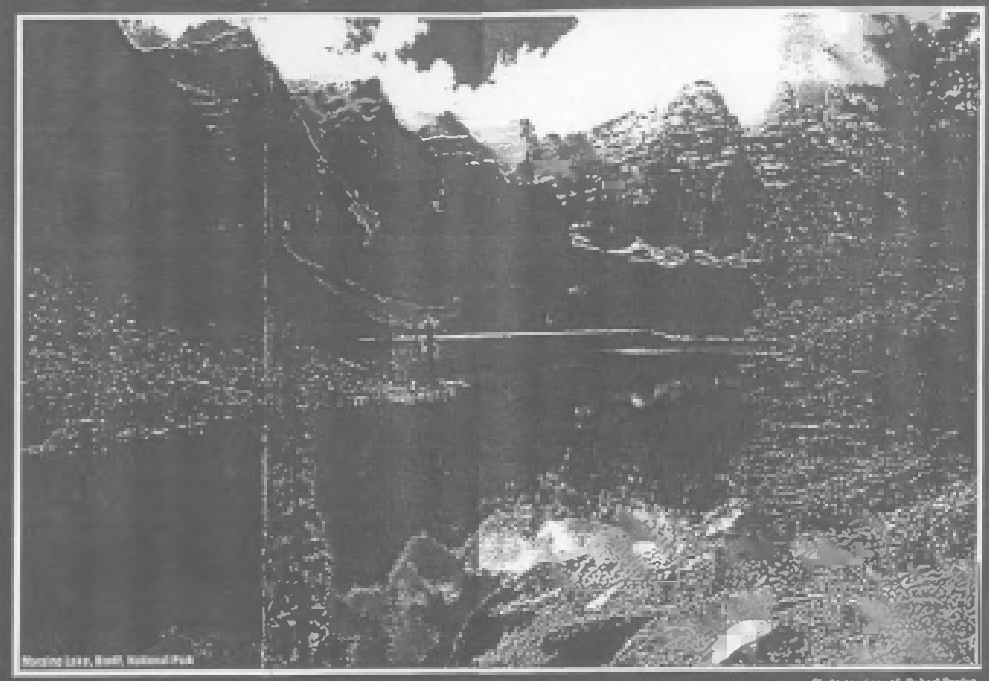

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