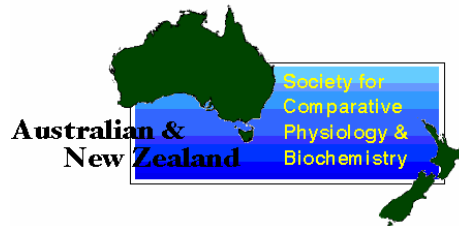




ANZSCP **2001**
Adelaide University



ANZSCP B

Proceedings, Volume 18

**Australian and New Zealand Society for Comparative
Physiology and Biochemistry**

18th Annual Meeting
6-9th December 2001

Organized by:

Dr Sandra Orgeig, Convenor

and

Dr Suzanne Munns

with support from

**Assoc. Prof. Chris Daniels,
Prof. Russ Baudinette and Prof. Roger Seymour**

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Venues:

The **main venue** for the conference is the Benham Building (map reference C9). All talks, registration, lunches, morning and afternoon teas, and the cheese and wine social will be held in or on the lawns surrounding the Benham.

The **poster session** on Friday evening will be held in the Level 2 Tearoom of the Darling Building (map reference G6).

The **conference BBQ** will be held on the Barr Smith Lawns (map reference E7).

The **conference dinner** will be a sumptuous 6-course banquet, held in the Henry Ayers Room, Ayers House. Ayers House is located at 288 North Terrace, east of Frome Road, opposite the Royal Adelaide Hospital.

Useful Information and Phone Numbers:

Parking on Campus

Weekend parking is available on campus for \$5.00 per day. Enter via gate 13 Kintore Ave, or gate 22 North Terrace.

Adelaide University Information and Security ph 8303 5990 (24hrs)

Banks

Bank SA Flexi tellers (accepting most cards) are located on the ground floor of the Union Building (map reference E5), and next to Security Office in Hughes Plaza (map reference H8).

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18 King William St (near North Terrace railway station).

Adelaide Zoo

Frome Rd, just south of the Torrens River. Opening hours 9:30-5:00pm.

Note:

The city of Adelaide is a dry zone. Consumption outside licensed venues (including conference venues) is illegal under South Australian law.

Program at a Glance

Time	Thursday 6 th Dec	Friday 7 th Dec	Saturday 8 th Dec	Sunday 9 th Dec
8.00		Registration		
8.50		Welcome		
9.00		Torpor & Hibernation	Cardiovascular Physiology	Aquaculture
9.15		Torpor & Hibernation	Cardiovascular Physiology	Aquaculture
9.30		Torpor & Hibernation	Cardiovascular Physiology	Endocrinology
9.45		Torpor & Hibernation	Cardiovascular Physiology	Endocrinology
10.00		Torpor & Hibernation	Morning Tea	Endocrinology
10.15		Torpor & Hibernation	Morning Tea	Morning Tea
10.30		Morning Tea	Cardiovascular & Lymphatic Regulation	Morning Tea
10.45		Morning Tea	CV & Lymph Regulation	Reproduction & Development
11.00		Thermal Biology	CV & Lymph Regulation	Reprod & Develop
11.15		Thermal Biology	CV & Lymph Regulation	Reprod & Develop
11.30		Thermal Biology	CV & Lymph Regulation	Reprod & Develop
11.45		Thermal Biology	CV & Lymph Regulation	Reprod & Develop
12.00		Thermal Biology	Lunch	Reprod & Develop
12.15		Thermal Biology	Lunch	Prizes / Closing
12.30		Photo	Lunch	Lunch
12.45		Bio-Rad Lunch	Lunch	Lunch
1.00		Bio-Rad Lunch	Lunch	Lunch
1.15		Bio-Rad Lunch	Lunch	Lunch
1.30		Bio-Rad Lunch	Keynote Address	Lunch
1.45		Bio-Rad Lunch	John Baldwin	Lunch
2.00		Metabolism & Scaling		Lunch
2.15		Metabolism & Scaling	Respiratory Physiology	
2.30		Metabolism & Scaling	Respiratory Physiology	
2.45		Metabolism & Scaling	Respiratory Physiology	
3.00		Metabolism & Scaling	Respiratory Physiology	
3.15		Metabolism & Scaling	Respiratory Physiology	
3.30		Afternoon Tea	Afternoon Tea	
3.45		Afternoon Tea	Afternoon Tea	
4.00	Student Chair Meeting	Environmental Adaptation	Marsupial Physiology	
4.15		Environ Adaptation	Marsupial Physiology	
4.30	Registration & Welcome Function	Environ Adaptation	Marsupial Physiology	
4.45		Environ Adaptation	Marsupial Physiology	
5.00		Environ Adaptation	AGM	
5.15		Posters & Refreshments		
5.30		Refreshments		
5.45				
6.00				
7.00		BBQ	Dinner	

Thursday, December 6th

4.30-7.00	Registration and Welcome in the Benham Building
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Friday, December 7th

8.00-8.50	Registration: Foyer of the Benham Building
8.50-9.00	Welcome
	Session 1 - Torpor and Hibernation, Chair: Kylie Leonard
9.00-9.15	Interpreting the geographic variation observed in torpor patterns of echidnas <i>Lyn Beard and Gordon Grigg</i>
9.15-9.30	Torpor, hibernation, echidnas and the evolution of endothermy <i>Gordon Grigg and Lyn Beard</i>
9.30-9.45	The timing of hibernation in Tasmanian echidnas: why do they do it when they do? <i>Stewart Nicol and Niels Andersen</i>
9.45-10.00	The devil's crucible: non-shivering thermogenesis in the Tasmanian devil, <i>Sarcophilus harrisii</i> <i>Alexander Kabat, Randy Rose, and Adrian West</i>
10.00-10.15	Daily torpor in <i>Sminthopsis macroura</i> : effects of sex and photoperiod <i>Nicole Feay and Fritz Geiser</i>
10.15-10.30	Does the glucose inhibitor 2-deoxy-D-glucose induce torpor in the Eastern pygmy-possum, <i>Cercartetus nanus</i> ? <i>Wendy Westman and Fritz Geiser</i>
10.30-11.00	Morning Tea
	Session 2 – Thermal Biology, Chair: Alexander Kabat
11.00-11.15	Thermal biology of free-ranging sugar gliders (<i>Petaurus breviceps</i>) <i>Nereda Christian and Fritz Geiser</i>
11.15-11.30	Effects of captive breeding on behaviour, morphology, and physiology of feathertail gliders (<i>Acrobates pygmaeus</i>) <i>Fritz Geiser and Caroline Ferguson</i>
11.30-11.45	The metabolic physiology of two species of Australian pigeon: the Crested Pigeon (<i>Ocyphaps lophotes</i>) and the Brush Bronzewing (<i>Phaps elegans</i>) <i>Alexander Larcombe</i>

Friday, December 7th continued

11.45-12.00	Body temperature and metabolism of a termite-eating marsupial, the numbat (<i>Myrmecobius fasciatus</i>): a comparison with termite-eating eutherian mammals Christine Cooper
12.00-12.15	Seasonal changes in gut morphology and function in a large herbivorous hibernator, the alpine marmot Ian Hume
12.15-12.30	Antifreeze proteins in the European eelpout <i>Zoarces viviparous</i> Thomas Flarup Sørensen and Hans Ramløv
12.30-2.00	Conference Photo – Steps of the Barr-Smith Library Bio-Rad Lunch: Benham Building Gardens
	Session 3 – Metabolism and Scaling, Chair: Wendy Westman
2.00-2.15	Acyl composition of skeletal muscle membranes varies with body size in birds Tony Hulbert, Sally Faulks, William Buttemer and Paul Else
2.15-2.30	The metabolism of hepatocytes in birds of varying body size Paul Else, Nigel Turner, M. Brand and Tony Hulbert
2.30-2.45	An allometric comparison of the sodium pump in the hearts of mammals and birds Nigel Turner, Kurt Haga, Tony Hulbert and Paul Else
2.45-3.00	Chasing pink mouse-to-elephants: will we ever know the ‘true’ allometric exponent relating BMR and body mass? Craig White
3.00-3.15	Do passerine birds have a higher SMR than non-passerine birds? – a revisit by autoregression Philip Withers and Elke Schleucher
3.15-3.30	Specific Dynamic Action (SDA) in the lizard <i>Eulamprus quoyi</i> Sebastian Iglesias, Frank Seebacher and Michael Thompson
3.30-4.00	Aftersnoon Tea
	Session 4 – Environmental Adaptation, Chair: Norman Ragg
4.00-4.15	What are the effects of light and temperature on the <i>in vitro</i> pineal melatonin rhythm of the Sleepy lizard, <i>Tiliqua rugosa</i> ? Kylie Leonard, Bruce Firth, Robert Moyer, David Kennaway, Mark Hutchinson

Friday December 7th, continued

4.15-4.30	A functional explanation of spectral matching between green tree frogs and green leaves <i>William Buttemer and Stephen Dain</i>
4.30-4.45	Cloning and expression of guanylin and uroguanylin peptides in the Spinifex hopping mouse, <i>Notomys alexis</i> <i>Ray Bartolo, Rachel Heimeier, and John Donald</i>
4.45-5.00	Snake kidney morphology - does the difference mean anything? <i>Naomi Bentley, Renée Firman, Danielle Philippe, Adrian Rakimov, Karen Wills, Graeme Zosky and James O'Shea</i>
5.15-7.00	Poster Presentations and Refreshments – Darling Building, Tea Room
7.00-	Conference BBQ – Barr Smith Lawns

5.15-7.00 Poster Presentations

P1	Does cholesterol facilitate low surface tensions in surfactant during torpor? <i>Jonathan Codd, Samuel Schürch, Christopher Daniels and Sandra Orgeig</i>
P2	Effects of feeding on energy metabolism in the radula muscles of the Blacklip abalone, <i>Haliotis rubra</i> <i>John Elias</i>
P3	Respiratory adaptations in terrestrial hermit crabs: an anatomical study <i>C. Farrelly and Peter Greenaway</i>
P4	Multiple embryonic α -type globin chains in a marsupial, the Tammar wallaby (<i>Macropus eugenii</i>) <i>Katherine Gill, R. Holland, A.A. Gooley, G. Dolman, R. Hope, S. Cooper</i>
P5	Expression of the natriuretic peptide receptors in the kidney of water deprived Spinifex hopping-mice <i>Rachel Heimeier and John Donald</i>
P6	Preparing to breathe: Ultrastructural changes in a lizard lung <i>Sonya Johnston and Christopher Daniels</i>
P7	Tachykinin receptors and actions in the chicken intestine <i>Lu Liu and Elizabeth Burcher</i>
P8	Isolation and contractile activities of a substance P-related peptide from the gut of the Australian lungfish, <i>Neoceratodus forsteri</i> <i>Lu Liu, Michael Conlon, Jean Joss and Elizabeth Burcher</i>

Friday December 7th, posters continued

P9	Postnatal development and control of the pulmonary surfactant system in the Tammar wallaby, <i>Macropus eugenii</i> <i>Natalie Miller, Sandra Orgeig, Christopher Daniels and Russell Baudinette</i>
P10	Expressional analysis of the natriuretic peptide system in the cane toad, <i>Bufo marinus</i> , in response to volume loading <i>Shane Riddell and John Donald</i>
P11	Possible maternal-fetal relations in antifreeze production in the eelpout <i>Zoarces viviparous</i> <i>Thomas Flarup Sørensen and Hans Ramløv</i>
P12	Trawl stress in Degens leatherjacket (<i>Thamnaconus degeni</i>) <i>B. Milic, Kelie Stead, Philip Thomas and I. Svane</i>
P13	Branchial reprocessing of the urine in the terrestrial Christmas Island red crab <i>Gecarcoidea natalis</i> <i>Harry Taylor and Peter Greenaway</i>
P14	Circulatory effects of clamping to the substratum in the paua, <i>Haliotis iris</i> <i>Harry Taylor, Norman Ragg and David Just</i>
P15	The potential uses of an antimicrobial, silver coated mesh (X-static™) in aquaculture <i>Ryan Wilkinson, John Carragher and Amanda Goodman</i>

Saturday, December 8th

	Session 5 – Cardiovascular Physiology, Chair: Rachel Heimeier
9.00-9.15	Does an increased haemoglobin content ameliorate the effect of hypoxia on heart rate in <i>Daphnia carinata</i> ? <i>Paul Wiggins</i>
9.15-9.30	Correlates of choroid rete development with the metabolic potential of tropical reef fish and the effect of strenuous exercise on the visual acuity threshold of <i>Lutjanus carponotatus</i> <i>Neill Herbert, Rufus Wells and John Baldwin</i>
9.30-9.45	Phylogeny of the systemic secondary vascular system in the teleostei <i>Peter Skov and Michael Bennett</i>
9.45-10.00	Nitric oxide regulation in the central arteries of the toad, <i>Bufo Marinus</i> <i>Brad Broughton and John Donald</i>
10.00-10.30	Morning Tea

Saturday, December 8th, continued

	Session 6 – Cardiovascular and Lymphatic Regulation, Chair: Peter Skov
10.30-10.45	Endocardial smooth muscle in mammalian and tortoise hearts <i>Bren Gannon, Graeme Campbell, Tony Thomas and Sue Murphy</i>
10.45-11.00	Brain and arterial blood temperatures of free-ranging oryx (<i>Oryx gazella</i>) <i>Shane Maloney, Andrea Fuller, Graham Mitchell, Duncan Mitchell</i>
11.00-11.15	The function of the autonomic innervation of the heart of the Fat-tailed dunnart (<i>Sminthopsis crassicaudata</i>) at low temperature <i>Graeme Zosky and Jamie O'Shea</i>
11.15-11.30	Control of heart rate during thermoregulation in the heliothermic lizard, <i>Pogona barbata</i> : importance of cholinergic and adrenergic mechanisms <i>Frank Seebacher and Craig Franklin</i>
11.30-11.45	Regenerating gecko tails: a story of lymphatic restoration <i>Suzanne Munns, Chris Tsopelas, Benjamin Lewis, Christopher Daniels, Rodney Cooter, Barry Chatterton</i>
11.45-12.00	Lymphangiogenesis and the presence of Vascular Endothelial Growth Factor-C (VEGF-C) in regenerating gecko tails <i>Benjamin Lewis, Christopher Daniels, Suzanne Munns, Sandra Orgeig, Megan Baldwin, Steven Stacker, Marc Achen and Rodney Cooter</i>
12.00-1.30	Lunch: Benham Building Gardens
1.30-2.15	Keynote Address: Philosophical Phish Physiology <i>John Baldwin</i>
	Session 7 – Respiratory Physiology, Chair: Ben Smith
2.15-2.30	Abalone gills: inefficient evolutionary relics? <i>Norman Ragg and Harry Taylor</i>
2.30-2.45	Effects of freshwater exposure on marine Atlantic salmon (<i>Salmo salar</i>): impact of water hardness and disease status <i>Shane Roberts and Mark Powell</i>
2.45-3.00	The oxygen cascade in varanid lizards: determinants of O ₂ transfer <i>Peter Frappell</i>
3.00-3.15	Lung maturity at hatching in birds of different activity patterns <i>Sue Runciman, Roger Seymour, Russell Baudinette</i>
3.15-3.30	The mechanics of breathing in the newborn Tammar wallaby <i>Peter MacFarlane, Peter Frappell, and Jacopo Mortola</i>
3.30-4.00	Afteenoon Tea

Saturday, December 8th, continued

Session 8: Marsupial Physiology, Chair: Peter MacFarlane	
4.00-4.15	Comparison of wear pattern and mode of action of the teeth of four species of wallaby <i>Roger Lentle, I. Hume, K. Stafford, M. Kennedy, S. Haslett, B. Springett</i>
4.15-4.30	Control of jaw movements in marsupial herbivores; wombats and kangaroos <i>Fuzz Crompton, Russell Baudinette, D. Lieberman and T. Owerkowicz</i>
4.30-4.45	Energetic advantages conveyed to Southern hairy-nosed wombats through their complex burrow systems and stable environment <i>Glenn Shimmin, Jayne Skinner and Russell Baudinette</i>
4.45-5.00	The role of the tail in the hopping kangaroo <i>Russell Baudinette</i>
5.00-5.30	Annual General Meeting, Chair: Russell Baudinette
7.00-	Dinner – Henry Ayers Room, Ayers House

Sunday December 9th

Session 9 – Aquaculture, Chair: Lucy Sullivan	
9.00-9.15	Levels of stress indicators in the commercially harvested rock lobster <i>Jasus edwardsii</i> , of the Southern Zone Lobster Fishery, South Australia <i>Michael Roberts and John Carragher</i>
9.15-9.30	Lobsters in the toilet: acid-base effects of ammonia exposure in the Spiny rock lobster <i>Jasus edwardsi</i> <i>Mark Powell, Brad Crear and Grant Allen</i>
Session 10 – Endocrinology, Chair: Lucy Sullivan	
9.30-9.45	Plasma corticosterone concentrations through the activity cycle in an alpine lizard <i>Susan Jones, Corrine de Mestre and Jane Girling</i>
9.45-10.00	Influence of corticosterone on progesterone and estradiol production in vitro by placental tissues of the Southern snow skink (<i>Niveoscincus microlepidotus</i>) <i>Jane Girling and Susan Jones</i>
10.00-10.15	Glucocorticoids, iodothyronine deiodinases and free thyroid hormones during embryonic development of the saltwater crocodile <i>Caroline Shepherdley, Christopher Daniels, Sandra Orgeig, Samantha Richardson, Barbara Evans and Veerle Darras</i>
10.15-10.45	Morning Tea

Sunday December 9th, continued

Session 11 – Reproduction and Development, Chair: Caroline Shepherdley	
10.45-11.00	Two distinct reproductive strategies are correlated with an ovarian phenotype in co-existing parthenogenetic strains of a parasitic wasp <i>Otto Schmidt, Dongmei Li, H. Roberts, M. Beck, A. Reinecke, U. Theopold</i>
11.00-11.15	Effects of maternal diet on offspring size and locomotion performance in two viviparous skinks, <i>Pseudemoia pagenstecheri</i> and <i>Pseudemoia entrecasteauxii</i> <i>Jacque Herbert, Michael Thompson and Simon Hudson</i>
11.15-11.30	Effect of incubation temperature on yolk reserves and energy content of hatchling green turtles (<i>Chelonia mydas</i>) <i>David Booth and Elizabeth Burgess</i>
11.30-11.45	Maternally derived hormones in Australian pelican (<i>Pelecanus conspicillatus</i>) eggs: a possible mechanism for brood reduction by siblicide? <i>John Carragher, Catherine Smallridge and Jeremy Robertson</i>
11.45-12.00	Arctic breeding birds and their use of capital or income for egg production <i>Marcel Klaassen</i>
12.00-12.15	Birth in the marsupial quoll, <i>Dasyurus hallucatus</i> <i>John Nelson and Robert Gemmell</i>
12.15-12.30	Student Prize Presentations and Closing Remarks
12.30-2.00	Lunch: Benham Building Gardens

Philosophical Fish Physiology

John Baldwin

School of Biological Sciences, Monash University, Clayton, Vic, 3800.

If the discipline of comparative biochemistry and physiology has any unifying philosophy it is probably to be found in the 'August Krough Principle'. This principle can be summarised by stating that for most problems nature has invented an animal on which it can be most conveniently studied. Adopting this philosophy leads to the use of animal type as an experimental tool, both for identifying and investigating unique problems, and for solving more general ones. Looking back over my own scientific career reveals that a disproportionate number of studies involved the use of fish as experimental tools. On reflection, I can identify three main reasons for why fish have so often been the animals of choice. Firstly, there are simply so many different species, which provides a huge pool of genetic diversity on which natural selection can act. Secondly, fish inhabit a wide range of very different environments that supply the potential for selection at the biochemical and physiological level. Thirdly, fish occupy a useful evolutionary position relative to other vertebrate classes. The importance of these three attributes of fish is illustrated by their application to the following questions: 1. What determines the size of red blood cells? 2. How are proteins modified to function at high pressure? 3. What is the ancestral subunit type of vertebrate lactate dehydrogenase? While these examples demonstrate how fish can be good for science, an additional question that can be asked is whether this science is good for fish. Answering such questions involves quite a different philosophy.

The role of the tail in the hopping kangaroo

R.V. Baudinette

Dept of Environmental Biology, Adelaide University, Adelaide, S.A., 5005.

In bipedal running the legs move in opposing phases and angular momentum is effectively cancelled out, making the body position relatively stable. In hopping a different situation applies as the legs move in phase, thus tending to make the body pitch. However in kangaroos the tail represents about 7% of the body mass. As the legs move through a step cycle the tail swings up and down, thus counteracting the pitching effects. This counterbalance is not precise, but the head + trunk only moves through about 10° . This reduction is helped by the tail not moving as a rigid body. What would happen if the kangaroo did not have a tail? A young red kangaroo, found near Swan Reach, South Australia, was born without a tail. There were other apparent abnormalities in the animal but these were not related to locomotory performance. The animal was matched with an animal of the same body mass and the two were trained to hop on a motor-driven treadmill. High speed digital images were used to determine the arc of the hip to metatarsophalangeal joint and the angle through which the (head+trunk) pitches. From these observations the role of the tail was defined.

Interpreting the geographic variation observed in torpor patterns of echidnas

Lyn A. Beard and Gordon Grigg

Dept of Zoology & Entomology, The University of Queensland, Qld, 4072.

Echidnas in colder parts of Australia have been shown to undergo both short-term torpor and long-term hibernation akin to the pattern seen in “classic” eutherian hibernation. We have suggested previously that both may be a result of the same capacity, expressed to different extents. Further documentation of the daily and seasonal body temperature patterns expressed by echidnas in different geographical areas (eg Southern Downs in SE Qld, Brisbane, Sydney, Alice Springs) seems to support this hypothesis, with all combinations of torpor observed, from daily through to seasonal. We have also suggested previously that echidnas hibernating in mild climates (with food available) may be taking advantage of the cold, using torpor for energetic advantage rather than energetic necessity once enough ‘condition’ has been built up for the coming breeding season. Taken with the new data referred to above, our present concept is that echidnas are, in a sense, facultative endotherms, with a well-developed capacity for dropping metabolic rate, for either a short or a long period. They can employ it or not depending on a combination of external (e.g. climatic) and internal (e.g. body condition) factors. Thus echidnas in the colder areas of Australia, which exhibit a long hibernation, are taking advantage of the cold and echidnas in milder climates may hibernate/torpidate facultatively, depending on their fat reserves, how good the season is etc. Echidnas in different areas would be expected therefore to enter torpor to different extents in different seasons and possibly in different years. This interpretation would predict that echidnas in tropical (warm, humid - not hot) areas, without a source of ‘coolth’, would be unlikely to enter torpor, while echidnas in very hot areas would drop their metabolic rate in a summer torpor (“aestivation”) to reduce their ‘resting’ heat production.

Effect of incubation temperature on yolk reserves and energy content of hatchling green turtles (*Chelonia mydas*)

David T. Booth and Elizabeth Burgess

Dept of Zoology & Entomology, The University of Queensland, Qld, 4072.

All sea turtles have temperature dependent sex determination, but it is unknown if incubation temperature affects hatchling attributes other than sex. In this study we incubated green turtle eggs at 26°C (all male), 28°C (mixture of male and female) and 30°C (all female) and determined hatchling, yolk-free hatchling and residual yolk mass, as well as energy density of yolk-free hatchling tissue and residual yolk. A 52 g newly laid green turtle contains 355 kJ, 12 kJ in the shell, 7 kJ in the albumen and 336 kJ in the yolk. Incubation temperature affected hatchling attributes in a complicated manner. Hatchlings from 26°C were heavier than hatchlings from 28°C and 30°C, and had larger yolk-free bodies and smaller residual yolks than hatchlings from 28°C and 30°C. Water content of yolk-free hatchling tissue was greatest at 30°C but similar at 26°C and 28°C, while residual yolk water content was lowest at 28°C but similar at 26°C and 30°C. Incubation temperature did not affect ash content of yolk-free hatchling tissue or residual yolk. Yolk-free hatchling tissue energy density (dry mass basis) from 28°C was greater than 30°C while hatchlings from 26°C had an intermediate energy density. Residual yolk energy density (dry mass basis) at 30°C was lower than 26°C and 28°C. Total energy content of hatchlings was greatest at 28°C (232 kJ) but similar at 26°C (205 kJ) and 30°C (200 kJ). Hence from an embryo energy expenditure point of view incubation at 28°C is optimal.

Nitric oxide regulation in the central arteries of the Toad, *Bufo marinus*

Brad Broughton and John Donald

School of Biological and Chemical Sciences, Deakin University, Geelong, Vic, 3217.

In the mammalian vasculature, nitric oxide (NO) derived from nitric oxide synthase (NOS) in the endothelium is the predominant signalling pathway mediating vasodilation. Once released, NO induces vasodilation by activating a soluble guanylyl cyclase (sGC) located intracellularly in the vascular smooth muscle cells, which generates cGMP. In contrast, the mechanisms by which nitric oxide controls the vascular smooth muscle of amphibians are not well understood; however it is assumed that they are similar to mammals. This study was designed to examine these mechanisms in the central vasculature of the cane toad, *Bufo marinus*. NADPH diaphorase histochemistry and immunohistochemistry found no evidence for eNOS in the endothelium of toad aortae, but it could be readily demonstrated in rat blood vessels, which were used as a control. Interestingly, NADPH histochemistry and immunohistochemistry using an antibody to neural NOS revealed the presence of NOS in the perivascular nerve fibres. The anatomical data was supported by *in vitro* organ bath physiology, which demonstrated that acetylcholine (10^{-5} M) (NOS dependent vasodilator) mediated vasodilation was not dependent on the presence of the vascular endothelium; however it was significantly reduced in the presence of a nNOS inhibitor, vinyl-L-NIO (10^{-4} M). In addition, atropine (10^{-5} M) (a muscarinic receptor inhibitor), L-NA (10^{-4} M) (a NOS inhibitor) and ODQ (10^{-5} M) (a sGC inhibitor) abolished the vasodilatory effect of acetylcholine. The presence of a sGC was supported via the use of SNP, a NO donor, whose vasodilatory effect was abolished in the presence of ODQ (10^{-5} M). In conclusion, we propose that an endothelial NO system is absent in toad aortae and that a NO system generated in perivascular nerves mediates vasodilation.

A functional explanation of spectral matching between green tree frogs and green leaves

William A. Buttemer¹ and Stephen J. Dain²

¹Dept of Biological Sciences, University of Wollongong, Wollongong, N.S.W., 2522, ²School of Optometry and Vision Science, University of New South Wales, Kensington, N.S.W., 2033.

Green leaves absorb light strongly at red and blue wavelengths but reflect light at both green (550 nm) and near-infrared (near-IR) wavelengths (700-1200 nm). Because over half of the solar heat load is associated with near-IR radiation, high near-IR reflectance greatly reduces leaf temperatures and subsequent transpiration. It is also known that some green, leaf-sitting tree frogs have high near-IR reflectance, but explanations for how such reflectance is achieved are lacking. We have characterised the spectral reflectance of a number of Australian tree frogs and have found some of these frogs (e.g., *Litoria caerulea*, *L. chloris*, *L. infrafrenata*, & *L. xanthomera*) to match the spectral reflectance of green leaves both at visible (400-700 nm) and at near-infrared (near-IR; 700-900 nm) wavelengths. Furthermore, they also show a sharp rise in spectral reflectance at 650 to 700 nm as seen in green leaves. Such extended and precise colour matching provides effective crypsis from avian and reptilian predators that are able to discriminate near IR radiation. By contrast, this near-IR spectral matching it is not found in all green *Litoria*, nor is it found in any of the brown-skinned species. Interestingly, all of the green Australian tree frogs that show high near-IR reflectance lack melanin in their melanophores. The melanophores of these frogs instead contain pterorhodin, which is nearly transparent to wavelengths above 650 nm. Melanin, by contrast, strongly absorbs light from UV through near-IR wavelengths. We believe that the different light absorbing properties of these darkening pigments is responsible for these intraspecific differences in spectral reflectivity.

Maternally derived hormones in Australian pelican (*Pelecanus conspicillatus*) eggs: a possible mechanism for brood reduction by siblicide?

John F. Carragher, Catherine J. Smallridge and Jeremy G. Robertson

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Studies in a wide range of oviparous vertebrates show that eggs can contain significant amounts of maternally derived hormones. It is suggested that these hormones may have multiple effects on a wide range of morphological, physiological and behavioural traits of the offspring. Thus, in some species of bird the mother deposits progressively more androgens (eg testosterone) in the second and subsequent laid eggs in a particular clutch. The hypothesized function of the extra androgen is to make the later hatchlings more aggressive than their older and larger siblings when it comes to begging for food – so more chicks survive to fledge. In contrast, in bird species where 2 eggs are laid but only one chick survives (usually the elder), the mother deposits more androgens in the first laid egg to facilitate the aggressive behaviour that results in siblicide. The Australian pelican is a species where brood reduction by siblicide does occur, however, as far as we can determine, it appears to be unique in that the siblicide is *facultative* and not *obligate* (Robertson & Johnson, unpublished obs.). There are several hypotheses that could explain why siblicide occurs in some Australian pelican nests but not in others, and the maternally derived hormone mechanism is one of these. We have sampled first and second laid eggs in pelican nests and measured testosterone concentrations in the yolk. There were significant differences in testosterone levels both within a single yolk (ie core, middle and outer yolk layers) and between eggs. Interestingly, there was a significant difference in the total amount of testosterone present in first and second laid eggs. However, because the majority of eggs were sampled destructively, we cannot yet answer the question whether the difference in maternally derived testosterone was correlated to the expression of siblicidal behaviour in a particular nest. Non-destructive sampling of yolk was successfully trialed in a small number of pelican eggs.

Thermal biology of free-ranging sugar gliders (*Petaurus breviceps*)

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Few studies on daily torpor have been conducted in the field and there is no information on whether differences in habitat affect torpor patterns and torpor occurrence. Therefore, we conducted a field study on the use of torpor by 10 sugar gliders of varying body mass from a mature woodland habitat by using temperature telemetry and radio tracking and performing behavioural observations from May to August (late Autumn and Winter). We compared our data with published information from a field study conducted in a more marginal habitat and with laboratory studies. Activity patterns of free-ranging gliders were affected by photoperiod, ambient temperature and the duration and amount of rainfall. Daily torpor occurred in conjunction with low ambient temperatures and/or rainfall and torpor bouts often occurred over consecutive nights and were synchronised among gliders. Gliders from the mature woodland habitat employed daily torpor in 4% of all observations (34 torpor bouts observed in 10 animals over 823 animal nights). Although torpor in the mature woodland habitat was much more frequent than in the laboratory, torpor was employed less often than in the study conducted in a more marginal habitat (17% of all observations, 103 torpor bouts in 11 animals over 614 animal nights). Our study suggests that torpor was employed in response to energetic constraints imposed by inclement or adverse weather conditions that made foraging difficult. However, torpor frequency also appears to be strongly affected by the quality of the habitat. It is likely that gliders from the mature woodland habitat are less constrained by limited energy availability than gliders from a more marginal habitat that frequently employed torpor to overcome energetic bottlenecks.

Body temperature and metabolism of a termite-eating marsupial, the Numbat (*Myrmecobius fasciatus*): a comparison with termite-eating eutherian mammals

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Eutherian termite-eating mammals have low body temperatures and basal metabolic rates, associated with reduced energy expenditure. This may be essential for mammals that exist on a diet of termites that is low in energy and is of uncertain availability. The numbat is the only marsupial to feed exclusively on termites, and is an excellent comparison for eutherian termite-eaters. The body temperature and basal metabolic rate of the numbat are not significantly lower than that of other marsupials. However they are comparable to that of eutherian termite-eating mammals. The basal metabolic rates of these eutherian mammals fit the regression line relating log mass and log basal metabolism for marsupial species. As numbats are marsupials, their rates of basal metabolism are already reduced to a level comparable to that of other termite-eating mammals.

Control of jaw movements in marsupial herbivores; wombats and kangaroos

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Little is known about activity pattern of the muscles controlling jaw movement in marsupial compared to placental herbivores (e.g. goats, horses, etc.). In placental herbivores, differential activity of the jaw muscles on the working and balancing sides controls transverse movements of the jaw during unilateral chewing, thereby transferring muscular force across the symphysis and increasing bite force. We report here on the electromyograms (EMGs) of the major adductor jaw muscles during mastication in 6 hairy nosed wombats and 3 red kangaroos while they were chewing on several different types of food. In contrast to placental herbivores, no significant activity in the balancing side jaw muscles was recorded during mastication in both these marsupials. Only active side jaw muscles apparently control jaw movement in marsupials so that no force is transferred via the massive and partially fused symphysis in wombats and the highly mobile symphysis in kangaroos. We argue that the different pattern of activity in the muscles of mastication in placentals and marsupials is related to the broad inflected angle to the jaw in marsupials and the deep vertical angle in placental herbivores. In marsupials, the superficial masseter inserts on the inflected angle and tends to rotate the jaw around its horizontal axis. In wombats, the whole jaw rotates so that only the molars on the working side occlude. In kangaroos the mobile symphysis permits only the working side hemimandible to rotate and break down food between the bilophodont molars.

**Cloning and expression of guanylin and uroguanylin peptides
in the Spinifex hopping mouse, *Notomys alexis***

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The guanylin peptides regulate renal and intestinal fluid and electrolyte epithelial transport by binding to and activating guanylyl cyclase C receptors to stimulate the intracellular production of cGMP. However, the role of guanylin peptides in fluid volume homeostasis is yet to be fully clarified. This study was designed to deprive Spinifex hopping mice (*Notomys alexis*), of water for a period of seven days, and then examine the expression of guanylin and uroguanylin mRNA in the kidney, and proximal and distal colon. *Notomys* is a desert rodent that can survive for extended periods without access to free water whilst maintaining plasma volume and osmolarity. Because signalling molecules that generate cGMP promote the excretion of water, it was hypothesised that guanylin and uroguanylin synthesis would be down-regulated in water-deprived *Notomys* to prevent the loss of water in the urine and faeces. The full open reading frame of guanylin and uroguanylin cDNAs from *Notomys* were cloned and sequenced to enable the expression of guanylin and uroguanylin mRNA to be determined using a semi-quantitative PCR method. Water deprivation caused a significant increase in guanylin and uroguanylin mRNA expression in the distal colon, but there was no significant difference in the expression of guanylin and uroguanylin mRNA in the kidney and proximal colon. The up-regulation of guanylin and uroguanylin mRNA in the distal colon of water-deprived mice was surprising and may be linked to the hydration and secretion of mucous that protects the colonic lining from abrasion from dehydrated faeces.

The metabolism of hepatocytes in birds of varying body size

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The metabolism of freshly isolated hepatocytes prepared from 8 species of birds ranging in body mass from 13 grams (zebra finch, *Poephila guttata*) to 35 kilogram (emu, *Dromaius novaehollandiae*) were measured. Parameters included; liver mass, cell viability and protein content, whole cell and functionally related oxygen consumption values were measured. Overall, isolated avian liver cells demonstrate a clear reduction in weight specific respiration with increasing body size. These relationships together with a compilation of values for both reptiles (16 species) and mammals (9 species) will be compared.

Daily torpor in *Sminthopsis macroura*: effects of sex and photoperiod

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Sex and photoperiod are known to affect torpor in several species of mammals and birds. As little is known about these interrelations in dasyurid marsupials, we examined how photoperiod affects the occurrence and patterns of torpor in female and male *Sminthopsis macroura* (25g) exposed to short (Feb.–Aug.) and long photoperiod (Aug.-Nov.). In both photoperiods, metabolic rates and body temperature (T_b) were measured in the absence of food (induced torpor); T_b and activity patterns were measured in the presence of food (spontaneous torpor). In both long and short photoperiods, induced torpor in males (100 & 96 %) was more frequent than in females (65 & 60%), but photoperiod did not affect torpor use. Spontaneous torpor in females (52%) was more common than in males (10%) during the short photoperiod; in long photoperiod torpor occurrence in both sexes was identical (8%). During induced torpor, metabolic rate, torpor bout length and the minimum T_b did not differ between the sexes nor between the photoperiods. However, during spontaneous torpor, females had significantly longer and deeper torpor bouts (bout length ~5.6-8.2 h; minimum T_b ~ 20 °C) than males (bout length ~2 h; minimum T_b ~27 °C) in both photoperiods. Our study shows that the use and pattern of torpor in response to photoperiod acclimation differs between male and female *S. macroura*. Males showed a high incidence of induced torpor under both photoperiods, whereas in females spontaneous torpor was common under short photoperiod. This suggests that in males, torpor is used predominantly as an emergency measure when food is limited whereas females may use spontaneous torpor to store energy during short photoperiod prior to their period of reproduction.

The oxygen cascade in varanid lizards: determinants of O_2 transfer

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Varanids in general exhibit greater aerobic capacities than other lizards. Despite a number of studies that have examined aspects of O_2 delivery in varanids no study has yet measured all of the oxygen transfer components in a single species. In a similar approach to the extensive investigations undertaken in mammals the respiratory system in three varanid species has now been examined in terms of oxygen transfer from the air to the blood during rest and sustained locomotory activity. The parameters controlling the transfer of O_2 through the various steps of the respiratory system are appropriate to meet the maximum demands for oxygen that occur at $\dot{V}O_2$ max with one possible exception, circulatory convection. Ventilatory convection is maintained during maximal aerobic locomotion ensuring adequate pulmonary ventilation and the protection of alveolar P_{O_2} . Little evidence exists to indicate a mechanically imposed constraint to breathe and the possibility of a gular pump acting to assist ventilation, as a general feature of varanids remains to be determined. Alterations in the relative contributions of $\dot{V}A/\dot{Q}_L$, pulmonary diffusion, diffusion equilibrium and R-L shunts preserved the alveolar-arterial P_{O_2} difference, ensuring that arterial oxygenation was maintained. In those species where increases in cardiac output were limited, $\dot{V}O_2$ max was achieved through increased extraction of oxygen at the tissues. Overall, the interrelationship of adjacent steps in the respiratory system ensures that one step cannot become limiting. Compensatory changes occur in various parameters to offset those parameters that are 'limited' the high aerobic activity of varanid lizards would not be achievable without a compensated circulatory convection.

Endocardial smooth muscle in mammalian & tortoise heart

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In 1887, Fano reported “tonus waves” (Tonusschwankungen) - slow cyclic baseline tone variations, in tortoise atria (*Emys europaea*). His and other pre-1900 studies suggested two distinct contraction types in heart muscle: (i) rapid repetitive cardiac twitches or beats, and (ii) slow partial contractions or changes in resting cardiac tone between individual beats. Interest in cardiac tonus waves was lost, however, when they were not seen in mammalian hearts, once these were first maintained *in-vitro* in the 1920's. Some smooth muscle was subsequently reported in tortoise heart lining (Shaner 1923), suggesting this was the origin of tonus waves. Human endocardial smooth muscle (ESM) was first reported by Nagayo (1909); subsequent mammalian studies are few, and its role in mammalian heart remains unknown. Gannon & Thomas, (1999) showed small ESM bundles were distributed in all human heart chambers, and found pig atrial ESM contracted to elevated K⁺ *in-vitro* (*unpublished*). Finding difficulties reliably isolating responsive mammalian ESM for *in-vitro* studies, we re-investigated the distribution and function of cardiac smooth muscle in Australian tortoises. By histology & immuno-histochemistry, in the Australian shortnecked tortoise [*Emydura macquarii*], ESM was restricted to the sinus venosus. “Tonus waves” were seen in electrically paced sinus venosus *in-vitro*, but not in Left or Right atria, nor ventricular muscle strips. Of many agonists tried, only Histamine gave any response (contraction) at physiological, or heroic doses; intramural nerve stimulation decreased ESM tone, and reduced tone-wave frequency. Tortoise ESM responses *in-vitro* may indicate likely responses of mammalian ESM *in-vitro* & *in-vivo*.

**Effects of captive breeding on behaviour, morphology, and physiology of feathertail gliders
(*Acrobates pygmaeus*)**

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Investigations on the physiology of mammals and birds are often conducted using captive-bred individuals and it is commonly assumed that the resulting data are representative of individuals living in the field. To investigate whether these assumptions are justified, we quantified morphological, behavioural, and physiological variables of the small marsupial feathertail glider (body mass approx. 12 g). We compared three populations: (i) individuals from a cool-temperate, montane area, (ii) individuals from a subtropical, coastal area, and (iii) captive-bred individuals. Captive-bred gliders differed from the montane field gliders in morphology (longer tails and snouts), behaviour (longer activity periods) and physiology (less frequent torpor, shorter torpor, shallower torpor, higher metabolic rates during rest and torpor, and slower rates of rewarming). Most of these differences were also apparent between the captive-bred and the coastal field gliders. Unlike both field populations, captive-bred gliders often became hypothermic and were unable to rewarm. The poor expression of torpor and thermal performance of the captive-bred gliders raises the question whether they possess the physiological capability for survival in the wild. In contrast to the other physiological variables, the minimum body temperature defended during torpor and the corresponding air temperature differed between the montane and coastal field gliders, but were similar in coastal field and captive-bred gliders. Our study shows that morphology, behaviour and physiology can be strongly affected by breeding in or acclimation to captivity. It demonstrates that large intraspecific differences can occur without long-term selection and that data from captive-bred individuals may not be representative of those living in the field.

Birth in the marsupial quoll, *Dasyurus hallucatus*

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Birth has been observed in a number of species of marsupial and in the studies to date, the newborn crawl up or across to the pouch. The method of birth in the quoll, a dasyurid, differs greatly from that observed in other marsupials. Births were recorded at normal speed using hand-held digital video cameras. Birth was heralded by a release of a watery fluid, about 1ml and then followed by a gelatinous, tubular mass, contained in either one or two tubes of material from the sinus. The newborn, still encased in their placental membranes, were within the gelatinous column. To exit this column, they had to grasp a hair and wriggle about 1 cm across to the pouch. In the pouch the newborn young had to compete for a teat. Although the quoll only possesses 8 teats, the number of young in the pouch immediately after birth was 17, 16, 6, 16, 13 and 11 for each of the 6 quolls filmed. Birth has been described previously in another two dasyurids. The observers did not describe birth as reported here for the quoll. Nevertheless the movement of the newborn from the sinus to the pouch is so quick that this could have been missed. It was only by filming birth from beneath and from the side that we were able to fully understand the birth process. It remains to be seen if this use of a gelatinous mass is part of the birth process in all dasyurids.

Influence of corticosterone on progesterone and estradiol production *in vitro* by placental tissues of the Southern snow skink (*Niveoscincus microlepidotus*)

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The reproductive cycle of the Southern snow skink (*Niveoscincus microlepidotus*) is characterised by an unusually prolonged gestation period. Gestation commences in spring and embryonic development is completed by early autumn; parturition, however, does not occur until the following spring. Plasma corticosterone in pregnant snow skinks exhibits a marked peak in autumn, presumably reflecting the metabolic requirements of these animals as they prepare for hibernation. We hypothesised that the corticosterone produced in autumn may also act indirectly by preventing parturition: plasma corticosterone concentrations are low in spring when parturition occurs. Therefore, we investigated the effect of corticosterone on *in vitro* production of progesterone and estradiol by placental tissues of *N. microlepidotus*. Both maternal (uterus) and embryonic (extraembryonic membranes) components of the placenta were incubated with and without corticosterone. The media were analysed for steroids using radioimmunoassay. Uterine and extraembryonic membranes exhibited similar patterns of steroid production. Estradiol production was inhibited in the presence of corticosterone, whereas progesterone production was stimulated. Thus, corticosterone influences the production of steroids by placental tissues *in vitro*. Further studies are required to determine whether modulation of placental steroid production by corticosterone influences the timing of parturition.

Torpor, hibernation, echidnas and the evolution of endothermy

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We propose that the evolution of endothermy must logically have been stepwise via facultative endothermy, similar to that preserved in some modern torpidators/hibernators. This proposal is in sharp contrast to other theories about the evolution of endothermy and is based on several lines of evidence and thought. These include; Tb patterns of 'classic' hibernation and shorter term, daily torpors occur in all the three mammalian classes, suggesting that these reductions in metabolic rate are plesiomorphic and, perhaps, an echo of the reptilian ancestry; a growing realisation that 'high and stable' is a very limited generalisation to describe mammalian and avian Tb, especially in smaller animals, because there are far too many exceptions; and a realisation, looking at the flexibility shown by echidnas in managing their thermal affairs for energy advantage in a diversity of habitats, that they are behaving in a way not far at all from the putative 'intermediate' facultative endotherm. The evolution of endothermy from a reptilian starting point was largely the evolution of insulation and heat production, particularly non-shivering thermogenesis. We present a model in which selection pressures acting on a medium-sized dominantly reptile-style thermoregulator favour the acquisition of insulation and a higher metabolic rate, augmenting heat retention at the end of a day. This would have allowed an ectotherm with some emerging 'endotherm' traits to remain active later into the night and so invade a new temporal habitat, or extend a geographic range to higher latitudes. We think that there is a need for a significant reappraisal of current thinking about the evolutionary context of torpor and hibernation in particular and, in general, about avian and mammalian endothermy and its origins.

Effects of maternal diet on offspring size and locomotion performance in two viviparous skinks, *Pseudemoia pagenstecheri* and *Pseudemoia entrecasteauxii*.

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Conditions during pregnancy have the potential to influence neonatal phenotype in reptiles. Additionally, the effects of external influences on the mother may increase with increasing placental complexity. The aims of this study were to: (1) determine the effect of maternal diet during pregnancy of viviparous skinks with complex placentae (*Pseudemoia pagenstecheri* and *P. entrecasteauxii*) on the size, mass and locomotor performance of offspring; and (2) determine whether such effects were greater in species with more complex placentae. Traits of offspring from females maintained in the laboratory during pregnancy, and from females that spent the majority of their pregnancy in the field, were compared for both species. Maternal conditions during pregnancy affected offspring size and mass in *P. pagenstecheri* (the more placentotrophic species), but not in *P. entrecasteauxii*. Offspring of *P. pagenstecheri* from laboratory pregnancies were significantly smaller (19.8 ± 0.3 mm SVL vs 22.4 ± 0.8 mm SVL) and lighter (0.15 ± 0.01 g vs 0.20 ± 0.02 g) at birth than offspring from mothers that spent the majority of their pregnancy in the field. These differences were not apparent in *P. entrecasteauxii*, indicating that *P. entrecasteauxii* was not significantly affected by the laboratory conditions. There was no clear effect of maternal diet on sprint speed for either pregnancy treatment in either species, or any effect on the relationship between offspring speed and body size. Offspring sprint speed did not increase as the offspring grew.

Correlates of choroid rete development with the metabolic potential of tropical reef fish and the effect of strenuous exercise on the visual acuity threshold of *Lutjanus carponotatus*

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It is hypothesised that the transport of oxygen to the retinal cells of fish with Root effect haemoglobins is impaired by strenuous exercise due to a proton load that drastically reduces arterial haemoglobin-oxygen affinity. Routinely active reef fishes have enhanced oxygen transport and anaerobic (i.e. blood lactate loading) potentials relative to inactive species. Surprisingly, the development of the choroid rete mirabile (employed as an oxygen concentrating apparatus in the eye) is directly correlated with post-exercise lactate loads rather than with the magnitude of the Root effect, and suggests that an increased development is adaptive for fish with high anaerobic potentials. The hypothesis that visual performance is reduced by strenuous exercise was tested in the stripey snapper, *Lutjanus carponotatus*, using the optomotor response. Moderate blood lactate loads (2 mmol l⁻¹ blood lactate) and red cell swelling responses were induced by exercise, but visual acuity thresholds (180 min of arc) were maintained. A moderate metabolic disturbance does not therefore appear to be a liability for the visual performance of a tropical fish in possession of Root effect haemoglobins.

Acyl composition of skeletal muscle membranes varies with body size in birds

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Phospholipids isolated from pectoral muscle of eight bird species were analyzed for their acyl composition by gas chromatography. The bird species examined ranged in body mass from 13g to 35kg and were: zebra finches (*Poephila guttata*), sparrows (*Passer domesticus*), starlings (*Sturnus vulgaris*), pied currawongs (*Strepera graculina*), pigeons (*Columba livia*), ducks (*Anas platyrhynchos*), geese (*Anser anser*) and emus (*Dromaius novaehollandiae*). Although the % unsaturates was relatively constant in birds of different body mass, there was a significant negative allometric trend in the unsaturation index of muscle phospholipids. The relative monounsaturate content of muscle phospholipids increased with increasing body mass of the bird species. The relative omega-6 polyunsaturate content did not vary with body mass whilst that of the omega-3 polyunsaturates was greater in the smaller species. The highly polyunsaturated omega-3 fatty acid, docosahexanoic acid (22:6), was more common in the muscle phospholipids from smaller bird species. The correlations between phospholipid acyl composition and body mass observed in birds are similar to those previously reported for mammals. The possible implications of these findings to understanding allometric variation in metabolic rate will be discussed.

Seasonal changes in gut morphology and function in a large herbivorous hibernator, the alpine marmot

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The gastrointestinal tracts of 76 free-living alpine marmots (*Marmota marmota*) shot during a population control program in Switzerland were collected and analysed for patterns of change in morphology and function over the period from emergence from hibernation in April to re-entry into hibernation in September. Between emergence and mid-summer (July) the fresh tissue mass of the small intestine increased by 259% (among the largest recorded for a mammal) versus 105% in the stomach and 138-185% in the hindgut. Mitotic activity was greatest in the small intestine, with mitotic indexes of 40% at emergence increasing to 60% in July, versus 4-8% in all other parts of the gut. Microbial activity in the caecum was significant even at emergence, increased between April and July, then declined to September. There were also significant declines in small intestinal tissue mass and mitotic activity between July and September. The small intestine responded to ingested food in spring later than did the stomach (in length) and caecum (in both length and fresh tissue mass), but declined in fresh tissue mass and fresh contents mass in autumn earlier than other sections of the gut. It is concluded that the gastrointestinal tract of alpine marmots probably continues to function, albeit at a low level, throughout hibernation. However, most of the increases in size and activity of the gut in spring are in response to ingested food rather than to some endogenous signal. The delay in up-regulation of the small intestine in spring, together with its early signs of down-regulation in autumn before re-entry into hibernation, are consistent with the high costs of maintaining this section of the digestive system.

Plasma corticosterone concentrations through the activity cycle in an alpine lizard

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In reptiles, it has been variously suggested that changes in plasma CORT reflect either reproductive activity, or the metabolic demands of feeding, growth and /or lipid deposition through the activity period; immune status may also be linked to adrenal function. In most species it is not possible to examine the effects of season independently of the effects of reproductive status. However the unusual biennial reproductive cycle of the viviparous southern snow skink, *Niveoscincus microlepidotus*, means that females in two different reproductive conditions are available simultaneously. To investigate the pattern of plasma CORT through the activity season, we made monthly collections of male and female southern snow skinks (in their vitellogenic or gestational year) from October to April. Blood samples were taken within 5 min of capture. Plasmas were assayed for CORT, and in whole blood smears we determined total white blood cell count, and the proportion of eosinophils, heterophils and lymphocytes, as indirect measures of immune function. Unexpectedly, we found that the profile of plasma CORT across months varied little with reproductive status or sex. All groups showed a peak of CORT in March/April, prior to hibernation. There were significant, complex, changes in white blood cell parameters in all groups: for example, total white cell counts were highest in January and in March. We conclude that in these alpine lizards, adrenal activity is more likely to reflect changing energy demands through the active season rather than reproductive status. Environmental factors also appear to influence white blood cell counts.

The devil's crucible: non-shivering thermogenesis in the Tasmanian devil, *Sarcophilus harrisii*

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The Tasmanian Devil (*Sarcophilus harrisii*; Boitard 1841) increases oxygen consumption by 12.8% in response to cold exposure (4° C for 14 days), which is further increased with an injection of norepinephrine by ≈37%. Thus alluding to Non-Shivering Thermogenesis in the devil, although unusually low compared to the majority of animals whom have been shown to possess NST. This study endeavoured to identify the mechanisms behind non-shivering thermogenesis in the Devil. We attempted to identify the presence of BAT/UCP1, UCP2 and UCP3 and their role in NST (if any). NST, indicated by resting VO₂, was measured under constant conditions at ≈20° C with various stimuli: long-term cold exposure (14 days at 2-3° C) and/or, Norepinephrine injection. Concurrent with the functional studies, western blots and RT-PCR were used to identify the expression of the BAT/UCP1, 2, and 3. Neither the expression of BAT /UCP1 nor UCP 3 was identified by the molecular techniques applied, in the devils pre- or post-cold acclimation. However UCP 2 was identified by molecular techniques in both pre-and post-cold acclimated Devils, suggesting that NST in the Devil is related to UCP 2. We suggest that it is this singular pathway that maybe the reason for the reduced NST capacity.

Arctic breeding birds and their use of capital or income for egg production

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Birds produce their eggs from recently ingested nutrients ('income breeders') or from body stores ('capital breeders'). Since summers are short at arctic latitudes, ecologists have long thought that arctic migrants need to bring the nutrients for egg production from afar, to be able to start breeding immediately upon arrival. However, using carbon stable isotope ratios of eggs, natal downs, and juvenile and adult feathers from ten wader species from 12 localities in Northeast Greenland and Arctic Canada, it was demonstrated that their eggs are produced from nutrients originating from tundra habitats. Thus, for these waders the fitness costs of transporting extra body stores to the breeding grounds outweigh the potential benefits. However, the capital strategy may still be used by large species of arctic breeding migrants, such as geese. It is likely that larger species need relatively smaller body stores for egg production. Also, the larger the species, the longer it needs to complete breeding, constrained by a fixed window opportunity. Indeed, tentative data using the same methodology of carbon stable isotope ratios in Lesser Snow Geese support the idea that, with respect to egg production, these larger species are capital rather than income breeders.

The metabolic physiology of two species of Australian pigeon: the Crested pigeon (*Ocyphaps lophotes*) and the Brush Bronzewing (*Phaps elegans*)

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The metabolic physiology of both the ubiquitous Crested Pigeon (*Ocyphaps lophotes*) and the mesic-dwelling Brush Bronzewing (*Phaps elegans*) was generally similar to that expected for birds of their size, although the Crested Pigeon possesses a number of characteristics which enhance its ability to survive in hot and dry regions. Body temperatures increased with ambient temperature similarly in both species, ranging from 38.8 to 43.1°C in the Crested Pigeon and from 39.3 to 42.4°C in the Brush Bronzewing. Both species became hyperthermic at high ambient temperatures, with the Crested Pigeon tolerating high body temperatures without obvious stress. The basal metabolic rate of the Crested Pigeon (0.64 ml O₂ g⁻¹ hr⁻¹ at 40°C) is low, ~64% of that predicted for a non-passerine, while that of the Brush Bronzewing (0.82 ml O₂ g⁻¹ hr⁻¹ at 30°C) is ~87% of that expected. The low metabolic heat production of the Crested Pigeon contributes to its high thermal tolerance. In both species, dry thermal conductance was low at T_a < 35°C. Cutaneous evaporative cooling (sweating) was identified in both species, with skin resistance decreasing as T_a increased. Cutaneous evaporative water loss in the Crested Pigeon and Brush Bronzewing was compared to other pigeons from a variety of habitats. The significance of the CECM was discussed. Both species were able to cool their expired air to temperatures considerably lower than T_b. This was most apparent at lower T_a.

Comparison of wear pattern and mode of action of the teeth of four species of wallaby

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Tooth morphology of two browsing (brush tailed and swamp wallabies), one grazing species. (Tammar wallaby) and one mixed feeder (parma wallaby) macropods are compared. The dental action of a single Tammar wallaby was studied by cinefluoroscopy. The teeth of the four species showed a number of morphological similarities along with differences that may represent adaptations for efficient acquisition and oral processing of browse or graze. Cinefluoroscopy confirmed independent rotation of each hemijaw on occlusion in the Tammar. The disposition of molar striae suggests a similar pattern of jaw movement in all four species. The narrower incisor arcade of the grazing species and coincidence of incisor action with a cutting action by the transverse loph during molar intercuspsation have implications for feeding efficiency and the functional response of macropod marsupials.

What are the effects of light and temperature on the *in vitro* pineal melatonin rhythm of the sleepy lizard, *Tiliqua rugosa*?

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The pineal organ has been shown to transmit environmental light and temperature conditions to the organism through its hormonal signal, melatonin. This signal has an effect on daily activity, reproductive cycles and thermoregulation. Previous studies on *Tiliqua rugosa* have shown that the rhythm of plasma melatonin content can be entrained to both light and temperature cycles and that the rhythm is a product of the interaction of these two cycles when they are presented concurrently. The aim of the present study was to determine whether the responses to light and temperature are intrinsic to the pineal organ or are the result of neural input from other brain structures. The pineal organs were isolated by culture in a dynamic perfusion system where two-hourly samples were collected for direct radioimmunoassay. The pineal melatonin rhythm entrained to both light and temperature cycles independently, and persisted in constant lighting and temperature conditions with reduced amplitude. Pineal melatonin rhythms were also present when light and temperature cycles were presented together such that the photophase and thermophase coincided or the photophase and cryophase coincided. In each instance the peak of the melatonin rhythm followed the cryophase. These results suggest that the pineal organ is able to work independently to generate melatonin rhythms in response to the environmental light and temperature conditions.

Lymphangiogenesis and the presence of Vascular Endothelial Growth Factor-C (VEGF-C) in regenerating gecko tails

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Lizard tail autotomy involves complete detachment of all tail tissues, including the lymphatics. In humans, when major lymphatic vessels are damaged or obstructed, drainage of the interstitial fluid is inhibited, resulting in lymphoedema. However, after reptilian tail autotomy the regenerate tail is not lymphoedematous. In humans, one of the protein growth factors thought to control the growth of lymphatic vessels (lymphangiogenesis) is vascular endothelial growth factor-C (VEGF-C). This study examined the process of lymphangiogenesis, and determined the presence of VEGF-C, in regenerating gecko tails. Histological analyses, over an eighteen-week period after autotomy, demonstrated that lymphangiogenesis occurred within the regenerating tails of *Christinus marmoratus*. Using a polyclonal antibody against human VEGF-C (C-20), reptilian VEGF-C polypeptides were identified by immunogenic blotting after six weeks of tail regeneration. The 58 kDa and 43 kDa VEGF-C immunopositive bands were absent in three-week old regenerates, suggesting that VEGF-C protein synthesis is not constitutive. Polymerase chain reaction confirmed that VEGF-C plays a role in lymphangiogenesis in the regenerating tails. While reptilian VEGF-C gene expression was absent during the first three weeks of tail regeneration it was dramatically up-regulated at six and nine weeks after autotomy. Gene expression was not detected at fifteen weeks of regeneration, and only low levels of expression occur thereafter. Northern blot analysis using a mouse VEGF-C probe confirmed that the vertebrate VEGF-C homology domain is highly conserved. There is, therefore, a strong correlation between the gene expression of reptilian VEGF-C and lymphangiogenesis.

The mechanics of breathing in the newborn Tammar wallaby

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We questioned whether or not the mechanical properties of the respiratory system may represent a major constraint to the spontaneous breathing act in the newborn tammar wallaby which is born after a very short gestation (≈ 28 days, birth weight ≈ 380 mg). The rate of oxygen consumption ($\dot{V}O_2$) through the skin was about 33% of the total $\dot{V}O_2$ at day 1, and about 14% at day 6. The resting minute ventilation ($\dot{V}E/kg$) and the ventilatory equivalent ($\dot{V}E/\dot{V}O_2$) were approximately the same at the two ages, with a breathing pattern significantly deeper and slower at day 1. The compliance of the respiratory system (Crs/kg) did not differ significantly between the two age groups, and was close to the values predicted from measurements in eutherian newborns. Respiratory system resistance (Rrs/kg) at day 1 was higher than at day 6, and also higher than in eutherian newborns. Chest distortion, quantified as the degree of abdominal motion during spontaneous breathing compared to that required to inflate the lungs passively, at day 1 was very large, whereas it was modest at day 6. We conclude that in the tammar wallaby at birth the high resistance of the respiratory system and the distortion of the chest wall greatly reduce the mechanical efficiency of breathing. At this age, gas exchange through the skin is therefore an important complement to pulmonary ventilation.

Brain and arterial blood temperatures of free-ranging oryx (*Oryx gazella*)

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We used implanted miniature data loggers to measure brain and arterial blood temperatures every 5 min for up to 15 days in four free-ranging oryx (*Oryx gazella*) in their natural habitat. Globe temperatures exceeded 45°C and average peak radiant heat load was $800 \text{ W}\cdot\text{m}^{-2}$. Arterial blood temperature exhibited a moderate amplitude nychthemeral rhythm of $1.8 \pm 0.3^\circ\text{C}$ (mean \pm SD). The amplitude of the nychthemeral rhythm was not influenced by variations in ambient heat load. Average brain temperature exceeded carotid arterial blood temperature by 0.29°C but there was a range of body temperatures over which the brain could be up to 0.4°C cooler or 0.5°C warmer than arterial blood. At high body temperatures ($>39.5^\circ\text{C}$) at rest, three of the animals tended to maintain the brain cooler than arterial blood. During exercise the brain was always warmer than arterial blood. The slope of the regression line relating brain temperature to carotid blood temperature was less than one. At short time scales of 5-20 min, brain temperature varied significantly more than did carotid blood temperature. We attribute part of the variability in brain temperature to transient stress responses and the influence of sympathetic activation attenuating selective brain cooling. We conclude that, contrary to the widely cited postulate, the carotid rete does not protect the brain during hyperthermia. Oryx also do not show adaptive heterothermy and, over short time intervals, have a brain temperature more variable than carotid blood temperature.

Regenerating gecko tails: a story of lymphatic restoration

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Lymphoedema is a chronic swelling, typically of one or more limbs, caused by the damage or malformation of lymphatic vessels and the associated insufficient removal of lymph fluid. Lymphatic vessels do not fully regenerate after injury, and there is still little effective treatment for patients with this disease. Caudal autotomy or “tail dropping” however, is common in the gecko *Christinus marmoratus*, where the regenerate tail shows no signs of lymphoedema. The caudal lymphatics of gecko tails were visualised and analysed following subdermal injections of the radiocolloidal agent ^{99m}Tc-antimony trisulphide (~1.5MBq/0.05ml). The lymphatic speed and the percentage activity migrated from the injection site were measured in geckos with non-regenerate tails, and at three week intervals (up to 18 weeks) post autotomy in geckos with regenerating tails. Results showed that fully regenerated tails (>6 months) had significantly slower lymph speed and migration of radiocolloid from the injection site than comparably sized geckos with non-regenerate tails. There was no measurable caudal lymphatic flow at 3 weeks post autotomy. At 6 weeks post autotomy a functional lymphatic system was established in regenerating tails, which did not differ in the lymph speed or in the radiocolloid migration characteristics from that measured at 9, 12, 15, or 18 weeks post autotomy. This study demonstrates that the size and number of caudal lymphatic vessels were indeed altered by autotomy, and that a functional lymphatic network was established in regenerating gecko tails between 3-6 wks post autotomy.

The timing of hibernation in Tasmanian echidnas: why do they do it when they do?

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We investigated the patterns of hibernation and arousals in 7 free-ranging echidnas *Tachyglossus aculeatus setosus* (2 male, 5 female) in Tasmania using implanted temperature data loggers. All echidnas showed a “classical” pattern of mammalian hibernation, with bouts of deep torpor interrupted by periodic arousals to euthermia (mean duration 1.03 ± 0.66 days, $n=147$). Torpor bout length increased as body temperature fell during the hibernation season, and became more variable as temperature rose again. Hibernation started in late summer (February 28 ± 5 days, $n=6$) and males aroused just before the winter solstice (June 15 ± 3 days, $n=3$), females that subsequently produced young aroused 40 days later (July 25 ± 3 , $n=4$) while females that did not produce young hibernated for a further two months (arousal Sept 27 ± 5 , $n=7$). We suggest that hibernation in Tasmanian echidnas can be divided into two phases, the first phase, marked by declining minimum body temperatures as ambient temperature falls, appears to be obligatory for all animals, while the second phase is “optional” and is utilised to varying amounts by females. We suggest that early arousal and breeding is the favoured option for females in good condition, and that the ability to completely omit breeding in some years, and hibernate through to spring is an adaptation to an uncertain climate.

Snake kidney morphology - does the difference mean anything?

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Snakes display extreme body elongation with consequent modifications and rearrangement to their abdominal contents; particularly the symmetry of paired organs. Some of the more obvious variations on the vertebrate body plan include features such as the loss of the left lung together with the attenuation and regional specialization of the right lung and the attenuation and staggering of the gonads and kidneys. The left kidney is typically located in front of the right kidney. Macroscopically, there appear to be differences in the relative length, shape and vasculature of the kidneys of various snakes. The snake kidneys facilitate osmoregulation and excretion. To achieve this end they are endowed with functional units called nephrons that allow filtration, selective secretion and selective reabsorption. Marine snakes usually possess lingual salt glands that are involved with secretion of NaCl and presumably take pressure off the kidneys for ionoregulation. At the commencement of this study it was predicted that marine and terrestrial snakes are subjected to different osmo- and ionoregulatory problems and may thus differ in their capacity to deal with such challenges - reflected in specialization of their renal morphology. We compared macro and micro anatomical organization of the kidneys of terrestrial and marine snakes using traditional dissection, histological techniques and scanning electronmicroscopy of resin (Mercox) filled corrosion castes of glomeruli and nephrons. We report that although there appear to be two different strategies for ionoregulation and excretion in snakes these do not appear to correlate with the environment of the snakes.

Lobsters in the toilet: acid-base effects of ammonia exposure in the spiny rock lobster, *Jasus edwardsi*

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Following harvest, rock lobsters (*Jasus edwardsi*) are often subjected to elevated ammonia concentration in holding facilities prior to and during transport to markets. Whereas the effects of air exposure has been well characterised for this species, little is known about the physiological and toxicological effects of environmental ammonia and deteriorated water quality. Lobsters were exposed in static tanks to a range of concentrations (0) of ammonium chloride (NH₄Cl) in normoxic seawater (n = 6) for 24 h. In a separate experiment animals were held at elevated ammonia concentrations (60 mg.L⁻¹ ammonium chloride) and sampled sequentially over a 24h period. Haemolymph pH was marginally elevated from 7.68 to 7.76 following exposure to 40 mg.L⁻¹ NH₄Cl but highest concentration of NH₄Cl tested (130 mg.L⁻¹) resulted in a significant acidosis (pH 7.61) after 24 h with a concentration dependant increase in CO₂ content, PCO₂ and haemolymph HCO₃⁻ concentrations. Increases in pH, total CO₂, PCO₂ and haemolymph HCO₃⁻ concentrations occurred within 5h of exposure to 60 mg.L⁻¹ NH₄Cl, however, these parameters remained stable over the ensuing 20 h of exposure. Following 24 h exposure to 60 mg.L⁻¹ NH₄Cl haemolymph pH, PCO₂ and total CO₂ concentrations were decreased and did not recover within 48h when lobsters were returned to NH₄Cl-free water. Similarly, exposure of lobsters to 60 mg.L⁻¹ NH₄Cl and 10 mg.L⁻¹ NaNO₂ also resulted in elevated haemolymph pH, and reduced PCO₂ and total CO₂ concentrations without recovery within 24 h following recovery in NH₄Cl and NaNO₂-free water.

Ammonia exposure (24h at 60 mg.L⁻¹) followed by 3 h of hypoxia (45% oxygen saturation) resulted in a similar increase in haemolymph pH with a reduction of haemolymph pH, PCO₂ and total CO₂ concentrations during recovery in NH₄Cl-free, normoxic water. These results suggest short-term physiological adjustments (within 5 h) in the acid-base status of the lobster due to environmental ammonia followed by longer term (5-24 h) metabolic effects. Recovery appears to be a lengthy process once CO₂ reserves (probably in the form of haemolymph HCO₃⁻) are depleted suggesting possible "metabolic tolerance" preferred over "mobilisation" of CO₂ from "hard reserves" such as the shell.

Abalone gills: inefficient evolutionary relics?

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A pair of bipectinate gills located either side of exhalent shell apertures represents an ancestral condition for gastropod molluscs. The presence of this arrangement in haliotids and a few other extant snails suggests that this may be a primitive and inefficient arrangement that has limited the lifestyle and success of species. Current dogma suggests that such animals are dependent upon external water movement to propagate ventilatory currents, in the absence of which the animal will be dependent upon supplementary oxygen diffusion across other body surfaces. Here we consider the gas exchange performance of the gills of the blackfoot abalone, *Haliotis iris*. Chronic indwelling cannulae allowed pre- and post-branchial haemolymph sampling for oxygen measurements and pulsed Doppler sonometry was used to quantify blood flow. Normoxic resting abalone maintained pre-branchial (P_{vO_2}) and post-branchial (P_{aO_2}) haemolymph P_{O_2} values of 35 and 70 Torr respectively. In the absence of either endogenous or exogenous ventilatory movements, P_{aO_2} frequently approached, or exceeded, seawater P_{O_2} values measured within the branchial chamber (P_{mO_2}), implying a level of diffusion efficiency usually seen only in advanced counter- or cross-current gas exchangers. By masking the shell apertures, a forced ventilatory current of 50 mL min⁻¹ was applied to the branchial chamber of 300 - 400 g abalone. Gill performance and P_{mO_2} changed little however, implying that the elaborate ciliary arrays found on the gills were sufficient to irrigate the lamellar surfaces under quiescent water conditions. The gills were shown to supply sufficient oxygen via haemolymph convection to support the resting aerobic metabolism of the whole organism in normoxic seawater. The roles of the gills, other potential gas exchange surfaces, and haemocyanin in accommodating the demands of acute exercise and hypoxia will be discussed.

**Levels of stress indicators in commercially harvested Rock lobster
Jasus edwardsii, of the Southern Zone Lobster Fishery, South Australia**

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Stress indicators were analysed in haemolymph samples taken from *Jasus edwardsii* at different handling stages in the commercial lobster fishing industry of South Australia's South East. These levels were compared with non-stressed (SCUBA caught and sampled) *J. edwardsii* samples. Variations in eight indicators were interpreted to determine factors associated with significant stress responses for this industry. Changes in levels of several stress indicators suggested that animals at all stages of the rock lobster industry tested (boat, jetty, transport and factory) were stressed, however stress indicator levels were highest at the jetty, transport and factory post harvest handling stages. Various factors such as the boat the lobsters were caught on and time held in factory tanks influenced the stress indicators measured. Recovery of lobster stress indicator levels was observed in factory holding tanks. All the stress indicators except calcium recovered to levels similar to those found in non-stressed samples during this period. However glucose, lactate and calcium levels increased dramatically within the first few hours in the holding tanks. The behaviour categories currently used by the industry appear to be good non-invasive indicators of the levels of stress responses experienced in rock lobster. Analysis of behaviour condition categories for each harvest and handling stage indicates that the majority of animals do not appear to be badly affected by stress. However, future research is needed to determine the economic significance (eg flesh quality and survival) of the stress responses identified in the Southern Zone Rock Lobster Industry of South Australia.

Effects of freshwater exposure on marine Atlantic salmon (*Salmo salar*): impact of water hardness and disease status

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Freshwater bathing is the most commonly used treatment for the alleviation of Amoebic Gill Disease (AGD) in sea-caged Atlantic salmon (*Salmo salar*). Water hardness ($[Ca^{2+}]$, $[Mg^{2+}]$) has been found to significantly differ between the freshwater sources used for such treatments. The physiological impact of exposing marine acclimated Atlantic salmon of different disease status to freshwater baths of differing hardness has yet to be documented. Here we examined the amoeba viability, branchial mucous cell histochemistry and mucus production, as well as ionoregulation of Atlantic salmon *in vivo* before and after 3 h bathing treatments. Experiments were conducted in tanks with groups of AGD affected fish (n=36), and in closed box respirometers with individual affected and non-affected fish being bathed (n=48). Treatments involved a control (seawater bathed fish), soft water bath ($37.35 \pm 5.35 \text{mg L}^{-1} \text{CaCO}_3$), hard water bath ($236.33 \pm 11.92 \text{mg L}^{-1} \text{CaCO}_3$) as well as a pre-treatment (fish sampled from holding tank). Results showed a significant increase ($P < 0.05$) in the kill rate of the AGD causative *Neoparamoeba* in soft freshwater (viable cells were reduced to 40.9%). Mucous cell glycology of diseased fish showed significant differences between healthy and lesioned lamellae ($> \text{PAS}^+$ cells in lesions, $P < 0.05$), but no significant differences, however, between treatments. No evidence of haemo-dilution occurred according to blood plasma protein and ion concentrations, thus differences found in net ion movement were attributed to an ionic flux. This study represents preliminary findings on freshwater exposure and disease status of marine Atlantic salmon and the respiratory physiological consequences involved, a first for this field.

Lung maturity at hatching in birds of different activity patterns

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Differences in maturity at hatching are bound to be related to patterns of development of pulmonary function and locomotor performance. We used morphometric techniques to investigate the maturational status of the lung in three species - the pelican an altricial, slow developing, swimmer and strong flyer, the domestic fowl (chicken), which is precocial and mainly terrestrial and the precocial and totally terrestrial emu. The lung of the chicken hatchling was more mature in appearance than that of the pelican and that of the emu. Parabronchi, were separated by well defined connective tissue septa. These septa were absent in the pelican and the emu lung. Air capillaries were smaller and more numerous in the chicken lung. Specific lung volume was highest in the pelican. The parenchymal fraction was highest in the lung of the emu but, within the parenchyma, relative volumes of airspace, tissue and blood capillaries were not significantly different. The surface area of the blood-gas tissue barrier per unit body mass ($S_t W^{-1}$) was significantly greater in the chicken but the difference between emu and pelican was not significant. These results suggest that the lung in the chicken has the highest diffusing capacity at the time of hatching.

Two distinct reproductive strategies are correlated with an ovarian phenotype in co-existing parthenogenetic strains of a parasitic wasp

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The question whether different organisms are able to compete for the same resource is of fundamental importance to evolutionary biology. Sympatric co-existence of similar species on a single resource has long been claimed to be unstable. However, indirect evidence suggests that parasitic wasps exhibit evolutionarily stable mixtures of life-history strategies. Here we describe genetically distinct strains of a parthenogenetic wasp *Venturia canescens* with different ovarian phenotypes that affect egg numbers in oviducts. Wasp females with large egg load search for caterpillars and deposit eggs immediately after host encounter, whereas females with fewer eggs delay parasitism. Since the outcome of interlarval competition within super-parasitised caterpillars depends on the age distribution of competing larvae, the two egg deposition strategies may co-exist under conditions that favour superparasitism.

Control of heart rate during thermoregulation in the heliothermic lizard, *Pogona barbata*: importance of cholinergic and adrenergic mechanisms

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During thermoregulation in the bearded dragon, *Pogona barbata*, heart rate during heating is significantly faster than during cooling at any given body temperature (heart rate hysteresis), resulting in faster rates of heating than cooling. However, the mechanisms which control heart rate during heating and cooling are unknown. The aim of this study was to test the hypothesis that changes in cholinergic and adrenergic tone on the heart are responsible for the heart rate hysteresis during heating and cooling in *P. barbata*. Heating and cooling trials were conducted before and after the administration of the muscarinic and β -adrenergic antagonists, atropine and sotalol, respectively. Cholinergic and β -adrenergic blockade did not abolish the heart rate hysteresis, and heart rate during heating was significantly faster than during cooling in all cases. Adrenergic tone was extremely high (92.3%) at the commencement of heating, and decreased to 30.7% at the end of the cooling period. Moreover, in four lizards there was an instantaneous drop in heart rate (up to 15 bpm) as the heat source was switched off, and this drop in heart rate coincided with either a drop in β -adrenergic tone or by an increase in cholinergic tone. Rates of heating were significantly faster during the cholinergic blockade, and rates of heating were least with a cholinergic + β -adrenergic blockade. The results showed that cholinergic and β -adrenergic systems are not the only control mechanisms acting on the heart during heating and cooling, however they do have a significant effect on heart rate and on rates of heating and cooling.

Glucocorticoids, iodothyronine deiodinases and free thyroid hormones during embryonic development of the Saltwater crocodile

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It is well recognised that thyroid hormones and glucocorticoids have important roles in development; however, there is little information about either of these hormones in developing reptiles. In many vertebrates studied to date, thyroid hormone levels in plasma peak late in development. An increase in plasma T_4 is mediated by the HPT axis, and a concurrent increase in T_3 is largely the result of an increase in conversion of T_4 to T_3 by deiodination. Late in development, glucocorticoids in plasma are also observed to rise. In embryonic mammals and birds, and metamorphosing amphibians, glucocorticoid treatment increases the outer ring deiodination (ORD) of T_4 to T_3 . Additionally, glucocorticoid treatment to embryonic birds and metamorphosing amphibians decreases the inner ring deiodination (IRD) of T_3 to T_2 . These relationships between glucocorticoids and deiodinase activities were examined in embryonic saltwater crocodiles by treating the embryos with dexamethasone, a synthetic glucocorticoid, at three time points during the last 20 days of incubation (days 60 to 80). Corticosterone and free thyroid hormone concentrations were measured in plasma, and high Km ORD, low Km ORD and IRD activities were measured in liver and kidney microsomes on days 60, 68 and 75 of incubation. Glucocorticoids decrease low Km ORD activity in liver and kidney microsomes on day 60 of incubation and may play a role in increasing high Km ORD late in development (around day 75). Moreover, glucocorticoids decreased T_3 IRD activity, which might have produced an increase in circulating total T_3 ; however, plasma free T_3 concentrations did not change, which suggests that free thyroid hormone levels could be tightly regulated during development.

Energetic advantages conveyed to Southern hairy-nosed wombats through their complex burrow systems and stable environment

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It is widely acknowledged that wombats, like other burrowing species, gain energetic advantages from adopting a subterranean lifestyle however neither the magnitude of these advantages nor the limitations or costs of warren construction have been explored. Furthermore, determining the environment in which wombats spend up to 70% of their lives could prove critical for the conservation of the endangered Northern Hairy Nosed Wombat. To determine wombat warren architecture in a non-destructive manner, which later allowed its environment to be sampled, we developed a porthole camera. Using this camera, warren complexes were sequentially portholed leaving in place sealed portholes for gas sampling. Logging of deep burrow temperatures showed daily ranges of less than 1° C over periods in which surface temperatures ranged up to 24 °C, however mean burrow temperatures varied 15.9 °C between winter and summer. Humidity gradients were maintained between the burrow and the surface and were depended on the complexity of the burrow. Oxygen and carbon dioxide levels were similar to those in free atmospheric air in unoccupied burrows, but in occupied tunnels reached 16.3 and 3% respectively. Wombat burrows ameliorate surface conditions in habitats of low humidity and high temperatures however humidity advantages promote the construction of tunnels beyond the requirements for temperature modulation.

Phylogeny of the systemic secondary vascular system in the Teleostei

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The presence of a systemic secondary vessel system has been documented in a number of representatives of the teleost division (2, 3, 5, 6, 7, 8 and others). Our knowledge of this vessel systems morphology shows that its points of origin, i.e. the interarterial anastomoses are similar for all species. A complete description of the secondary vessel systems distribution pattern is lacking, probably due to the significant variation between species, as does a satisfactory explanation of its physiological function. Ishimatsu et al. (1) suggested that this vessel system could be involved in acid-base regulation, although this suggestion was weakened by the observation of low fluid flow rates through this system (5). It has recently been shown that the flow of this system in the Atlantic cod (*G. morhua*) is in fact ten fold higher than previously assumed (4), a finding that re-opens discussion about its possible physiological functions. The points of origins of the secondary vascular system, and the tissues they supply, were examined in the silver trevally, *Pseudocaranx dentex* (Bloch & Schneider, 1801), collared sea bream, *Gymnocranius audleyii* (Ogilby, 1916), blackspot seaperch, *Lutjanus fulviflammus* (Forsskål, 1775), longfinned eel, *Anguilla reinhardtii* (Steindachner, 1867), black edged conger, *Conger cinereus* (Rüppell, 1830) and the Northern saratoga, *Scleropages jardinii* (Saville-Kent, 1892). These species are representatives of the Carangidae, Lethrinidae, Lutjanidae, Anguillidae, Congridae and Osteoglossidae respectively. The systemic secondary vascular system has not previously been described in any of these families. This poster presents information about the current knowledge of the phylogenetic distribution of systemic secondary vasculature. It also presents new data on the distribution of secondary vessel origins and the tissues that are invested with these vessels within individual species.

Antifreeze proteins in the European eelpout *Zoarces viviparus*

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Several fish species from polar and north temperate regions have evolved macromolecular antifreezes in order to avoid freezing at low water temperatures. The macromolecular antifreezes are believed to act by binding to the ice crystals and thereby preventing further growth. This action results in a non-colligative freezing point depression. Antifreeze activity, defined as the temperature difference (°C) between the melting point and the non-equilibrium freezing point of a solution, is also known as the thermal hysteresis. So far five different classes of macromolecular antifreezes have been identified (Ramløv, 2000). The European eelpout *Zoarces viviparus* is a common inhabitant in the shallow waters along the Danish coastline throughout the year. Locally *Z. viviparus* is subjected to seasonal fluctuation in water temperatures from > 20°C in summer to subzero temperatures in winter. Serum antifreeze activity was found and showed a seasonal variation with high levels (> 1.2°C) in winter and low levels (<0.1°C) during summer and autumn. Antifreeze activity seems unaffected by diminishing ice crystal fractions at ice fractions below 0.1 whereas ice fractions above 0.1 caused a decline in antifreeze activity (Sørensen & Ramløv, 2001). Purification of the antifreezes from *Z. viviparus* has revealed that up to 5 separate antifreezes are present in serum from adult specimens. Preliminary results indicate that at least 2 of the antifreeze proteins from *Z. viviparus* have molecular weights of app. 6 kDa. Mass Spectrometry analyses will elucidate the primary structure of these antifreeze proteins.

Specific dynamic action (SDA) in the lizard *Eulamprus quoyi*

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SDA refers to the rise in metabolic rate following feeding in animals, and may vary with time of day, different food type, size and feeding frequency. The circadian and feeding effects on the resting metabolic rate (RMR) of the Eastern water skink, *Eulamprus quoyii*, were measured using flow-through respirometry to determine the effect of: 1. circadian rhythms on RMR, 2 the energy and digestible protein content of a meal on SDA, and 3. repeated meals on SDA. The lizards showed strong circadian rhythm in RMR that was largely eliminated in conditions of constant light. Lizards were fed one of three diets, banana, banana supplemented with fat extract from mealworms, and mealworms. Both banana and banana plus fat extract elicited peaks in metabolic rate that matched the circadian fluctuations in RMR. Thus fat has little or no effect on SDA in *E. quoyii*. In contrast a significant SDA effect occurred in lizards fed mealworms, with a peak 15 hours after feeding. Sequential meals resulted in lizards maintaining an elevated RMR.

An allometric comparison of the sodium pump in the hearts of mammals and birds

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Metabolic intensity is allometrically related to body size in both mammals and birds. We have compared the allometry of the membrane bound sodium pump enzyme from the heart of mammals ranging in body mass from 40g to 500kg and birds ranging from 13g to 35kg. The species examined in the study were for mammals: mice (*Mus musculus*, Qs strain), rats (*Rattus norvegicus*, Wistar strain), rabbits (*Oryctolagus cuniculus*, N.Z. White strain), sheep (*Ovis aries*), pigs (*Sus scrofa*) and cattle (*Bos taurus*), and for birds: zebra finches (*Poephila guttata*), sparrows (*Passer domesticus*), starlings (*Sturnus vulgaris*), pied currawongs (*Strepera graculina*), pigeons (*Columba livia*), ducks (*Anas platyrhynchos*), geese (*Anser anser*) and emus (*Dromaius novaehollandiae*). Na⁺K⁺ATPase activity was measured in heart tissue homogenates. A significant negative allometric slope was found in both mammals and birds with the level of activity being similar between the two groups. Sodium pump density was determined in tissue biopsies. Constant pump density was found within mammals and birds of different mass, with the birds having slightly more pumps than mammals. These parameters were measured at 37°C for mammals and 41°C for birds, temperatures that approximately reflect *in vivo* conditions. Molecular activity, which is the turnover rate per individual sodium pump, was calculated by dividing the Na⁺K⁺ATPase activity by the sodium pump density to give molecules of ATP consumed per minute per pump. Both the mammals and the birds showed negative allometric exponents with the mammals having slightly higher activities than the birds. These results will be discussed relative to the membrane acyl composition of the heart.

Does the glucose inhibitor 2-deoxy-D-glucose induce torpor in the Eastern pygmy-possum, *Cercartetus nanus*?

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In heterothermic placental mammals the glucose inhibitor, 2-Deoxy-D-glucose (2DG) induces a torpor-like state, suggesting that decreased glucose availability triggers torpor onset. We tested whether glucoprivation by 2DG also induces torpor in a marsupial, the Eastern pygmy-possum, *Cercartetus nanus*, and whether 2DG-induced torpor differs from natural torpor. Oxygen consumption, carbon dioxide production and body temperature (T_b) were compared between animals on three doses of 2DG (800, 1600, and 2400 mg/kg), saline as a control, and natural torpor. Torpor frequency was similar for the two highest doses of 2DG and natural torpor (86-100%). The lowest 2DG dose resulted in less torpor (40%), no animals entered torpor on saline. Torpor bout length during natural torpor was two-three times longer than for the 2DG-induced state. Minimum T_b and metabolic rates were 20% and 50% lower for natural torpor than 2DG induced torpor, which was dose-dependent. Arousal rate was retarded by 2DG and was 3 times faster for natural torpor. Our study shows that 2DG induces a torpor-like state in *C. nanus* as in placental mammals. However, due to the pronounced differences in physiological variables between 2DG-induced torpor and natural torpor, the question arises whether decreased glucose availability triggers torpor onset or simply induces reversible non-regulated hypothermia.

Chasing pink mouse-to-elephants: will we ever know the ‘true’ allometric exponent relating BMR and body mass?

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The relationship between mammalian basal metabolic rate (BMR, ml O₂ min⁻¹) and body mass (*M*, g), specified by the equation $BMR = aM^b$, has been the subject of regular investigation over the past century, with metabolic data presently available for over 500 species. Arguments for and against geometric ($b = 2/3$) and quarter-power ($b = 3/4$) scaling have been made and rebutted throughout the literature, and a consensus has not yet been reached. The question could be resolved if measurements from all 4600 extant species were available, but this is unrealistic. Therefore the question becomes how many species covering what range of body masses are required to arrive at a reasonable estimate of the relationship between BMR and *M*. To answer these questions, I undertook an analysis of random subsamples of computer generated data for 4600 artificial species based on extant mammals to determine if calculation of a single ‘true’ allometric scaling exponent is currently possible. This analysis shows that sufficient BMR data are currently available to produce a reliable estimate of the scaling exponent ($BMR = 0.069 M^{0.69}$). However, reanalysis of recent compilations of mammalian BMR shows that the scaling exponent is significantly positively correlated with body mass ($b = 0.53 M^{0.042 \pm 0.001 [SE]}$) and varies from 0.57 at 10 g to 0.85 at 100 kg. This shows that there is no single, ‘true’ allometric scaling exponent relating mammalian BMR and *M*.

Does an increased haemoglobin content ameliorate the effect of hypoxia on heart rate in *Daphnia carinata*?

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Preliminary investigations have recently been carried out which reveal a great deal of basic information about the heart rate of the branchiopod crustacean *Daphnia carinata*: (1) at 18°C, animals taken directly from the wild ($T_a = 13^\circ\text{C}$) had a heart rate of 349.6 ± 12.7 bpm ($n = 20$); in acclimated animals (1-, 2-, and 3-days at 18°C; acclimation effect established in 24 hours) this rate was found to be 289.0 ± 6.6 bpm ($n = 60$); (2) a size effect was present – $HR = 200.84M^{-0.26}$ ($n = 60$) for acclimated animals; and, (3) that severe hypoxia has a dramatic effect on heart rate – moderate hypoxia (50% atmospheric) induced an 11 % increase in heart rate, whereas severe hypoxia (25 % atmospheric) caused heart rate to decrease to less than 40 % of the normoxic value. Furthermore, upon their return to normoxic water, the heart rates of animals exposed to severe hypoxia recovered to values equal to those observed during the initial period of normoxia. Thus, it is thought that there is a “breaking point” or P_c , below which these animals are unable to maintain their heart rate in hypoxia. Furthermore, more detailed experiments may show that the actual value of this P_c is dependent upon the animals’ haemoglobin content, a parameter which has been previously shown to play an important role in this species’ tolerance of hypoxia.

Do passerine birds have a higher SMR than non-passerine birds?– a revisit by autoregression

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Do passerine birds have a higher standard metabolic rates (SMR) than non-passerine birds? We re-examine SMR data for passerine and non-passerine birds using autoregression, being well aware of the problem with comparing the one order of passerines against the "non-passerines" that are not a uniform taxonomic group. Traditional analyses of the SMR of passerine and non-passerine birds (e.g. Lasiewski & Dawson, 1967; Zar, 1969) concluded that passerines have a higher SMR than non-passerines, but a phylogenetic analysis by independent contrasts (Reynolds & Lee, 1996) concluded that the SMR of passerines did not differ from that of non-passerines. Autoregression provides an alternative phylogenetic method to reinterpret the question, in view of the assumptions, limitations and shortcomings of independent contrasts, and has the advantage that phylogenetically-corrected BMR residuals are available for further analysis *e.g.* of the potential influence on SMR of climate or habitat. We conclude that passerine birds do indeed have a higher SMR than non-passerine birds and that the difference is phylogenetically based. However, the different orders subsumed into the "non-passerines" are as diverse with respect to each other each other as the passerines are compared to them. Some of this variability can be related to ecological and biogeographic factors, such as habitat, climate and feeding strategy.

The function of the autonomic innervation of the heart of the Fat-tailed dunnart (*Sminthopsis crassicaudata*) at low temperature

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The pattern of cardiac autonomic innervation in the Fat-tailed dunnart (*Sminthopsis crassicaudata*) was investigated at 32°C and 14°C using isolated cardiac preparations. It has been shown that *S. crassicaudata* has both a sympathetic and parasympathetic innervation of the ventricles which are both capable of altering the basal force of contraction; a pattern that has only been shown in one other mammal, the Bent-winged bat (*Miniopterus schreibersii*). As both these species are heterothermic, and given that a similar pattern of innervation is not present in mammalian homeotherms, it is possible that this pattern of cardiac autonomic innervation is related to the demands placed on the heart by the large alterations in body temperature associated with heterothermia. This study aimed to quantify the influence of the parasympathetic and sympathetic innervation of the Dunnart ventricle on the basal force of contraction at 32°C and determine whether these nerves remained functional at low temperature.

Does cholesterol facilitate low surface tensions in surfactant during torpor?

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Pulmonary surfactant reduces the surface tension created at the air-liquid interface in the lung. Surfactant, a complex mixture of lipids and proteins, is influenced by physiological parameters such as MR, T_b and breathing. In the microchiropteran bat *Chalinolobus gouldii* these parameters fluctuate throughout a 24h period. Here we examine the surface activity of surfactant from warm-active and torpid bats at both 24°C and 37°C to establish whether previously described alterations in surfactant composition correlate with changes in surface activity. Alterations in the lipid composition of surfactant occur with changes in the activity cycle. Most notable is an increase in surfactant cholesterol with decreases in body temperature (Codd et al. 2000). These alterations in the cholesterol content of bat surfactant appear to be crucial to allow it to achieve low surface tensions during torpor.

Effects of feeding on energy metabolism in the radula muscles of the Blacklip abalone, *Haliotis rubra*

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The effects of a feeding regime on energy metabolism of the radula muscle mass of blacklip abalone *Haliotis rubra* were examined. Abalone were induced to feed by either presenting them with 'drift' algae (*Ulva* sp., *Jeannerettia* sp., *Ecklonia* sp.) or allowing them to graze upon diatomaceous and micro-algal growth. Abalone browsing on drift algae displayed a preference for *Ulva* sp. Control/fasting animals showed little accumulation of either D-lactate or tauropine in the radula muscles, and these metabolites were not observed in the haemolymph. The metabolites did not change significantly in the radula muscles or haemolymph following browsing or grazing, and no significant differences were observed between the two feeding treatments. Enzyme profiles of the foot, shell adductor and radula muscles indicate a higher capacity for aerobic metabolism in the radula muscle. These results suggest a predominant contribution of aerobic metabolism to radula and buccal mass feeding movements. Whilst previous literature suggests an accumulation of D-lactate and opines following *in vitro* radula retractor muscle contraction, this does not appear to be the case *in vivo* during natural feeding behaviour of the blacklip abalone.

Multiple embryonic α -type globin chains in a marsupial, the Tammar wallaby (*Macropus eugenii*)

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The embryonic α -type globin chains in mammals have 50-60 sequence difference from the adult α -type chains, and are known as zeta (ζ) chains. There are known instances of multiple adult α -chains but no other mammal has been reported to have more than one ζ -chain. We had previously reported that the Tammar had 2 different ζ -chains in the embryonic and neonatal period, and have now obtained full sequences of both ζ type chains (141 amino acids). Initial sequencing was by PCR and DNA sequencing, but this gave the sequence of only one chain. However both ζ and ζ' were sequenced from the expressed protein from the blood. After red cells lysis the Hb types were separated by anion exchange chromatography, and, after dissociation of the hemes, the individual chains were separated by reverse phase chromatography. After tryptic digestion each peptide was analyzed by mass spectrometry (MS/MS). The sequence found for ζ in this way agreed with that from DNA. The main features of the sequences were: (i) Both were typical ζ -type chains showing, inter alia, acetylation of the N-terminal Ser (ii) There was 72-78% identity between ζ and other mammalian ζ -chains but only 58% identity between ζ and Tammar adult α . (iii) There were 12 differences between ζ and ζ' . It is concluded that there are 2 separate ζ -type genes expressing their globin products. (iv) Position 69 in ζ' was either Ala or Val, this difference being attributed to a polymorphism. We have not yet determined the functional significance of the presence of the two embryonic α -like chains.

Respiratory adaptations in terrestrial Hermit crabs: an anatomical study

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Most land crabs, including *Birgus latro* have developed branchiostegal lungs for aerial gas exchange. However, in the shell-carrying coenobitids, the branchiostegites have been restricted by need to retract into the shell and are thus narrow with a relatively small surface area and the internal space is occupied predominantly by the gills. In consequence, coenobitids have developed an alternative and supplementary respiratory site on the dorsal surface of the abdomen, which is protected by the shell. The dorsal cuticle of the abdomen is very thin and transparent and is formed into numerous tiny folds that greatly increase the surface area. Corrosion casts show this site to be highly vascular, with a complex network of lateral vessels supplied by paired longitudinal veins. Sections through this tissue also show that a very thin cuticle and epithelium line the exchange lacunae, providing a short blood/gas diffusion distance (300-500nm).

Expression of the natriuretic peptide receptors in the kidney of water deprived *Spinifex* hopping-mice

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Atrial natriuretic peptide (ANP) is a member of a family of structurally related peptides that mediate diuresis and natriuresis and cause vasodilation. ANP exerts its effect by binding to cell surface receptors of which three forms have been identified; natriuretic peptide receptor type A (NPR-A), NPR-B and NPR-C. The expression of ANP and the three-receptor forms have been detected in the mammalian kidney. For desert mammals, the regulation of body fluid is critical because of their dry environments. The *Spinifex* hopping-mouse, *Notomys alexis*, is capable of tolerating an absence of free water for prolonged periods by excreting a concentrated urine and utilising metabolic water. We have previously shown that cardiac ANP mRNA in *Notomys* was not significantly affected by periods of 7-, 14- or 28-days of total water deprivation (TWD), but in contrast, renal ANP mRNA was significantly reduced at 14 days, but not at 7 or 28 days. These data indicated that down regulation of the renal natriuretic peptide system might be an important adaptation to a xeric environment. In the present study we have partially cloned the three natriuretic peptide receptor forms in *Notomys* and subsequently measured the mRNA expression of NPR-A, B and C in kidney tissue obtained from the 7-, 14- and 28-day TWD study periods using a semi-quantitative PCR-based method. Compared with controls, the steady-state levels of mRNA for NPR-A were increased for 7- and 28-day TWD *Notomys* but were unchanged for the 14-day time period. NPR-B mRNA was decreased in all TWD animals for all time periods, while NPR-C mRNA expression remained unchanged. These results imply that the expression of NPR-A and NPR-B are responsive to periods of total water deprivation in *Notomys*.

Preparing to breathe: Ultrastructural changes in a lizard lung

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The bronchoalveolar lung of mammals enters at least three distinct stages during development. The epithelium changes from undifferentiated, high columnar cells, comprising large masses of glycogen with few organelles, to cuboidal epithelial cells, which begin degrading glycogen stores, before differentiating into primitive type II cells. The type II cells accumulate large stores of lamellar bodies, the intracellular storage site for surfactant, with advancing gestation. Glycogen provides the substrate for surfactant lipid synthesis and subsequent storage in lamellar bodies. The type II cells differentiate into type I cells, which contribute to the formation of the air-blood barrier. Shortly after differentiation of the epithelial cells, surfactant and tubular myelin are found in the developing airspaces. The unicameral lung of the lizard, *Pogona vitticeps* undergoes similar maturational changes to that of mammals. During the final stages of lung development, following 92% of incubation, the cuboidal type II cells differentiate into type I cells. Remaining type II cells show decreases in glycogen content with concomitant accumulations of lamellar bodies. After pipping, surfactant aggregates appear in the airspace whereas no change is observed after hatching. Such qualitative evidence suggests that, as with mammals, the lacertid lung enters similar distinct changes during late development and that glycogen provides a substrate for surfactant lipid synthesis in lizards. Furthermore, the structural changes appear relatively later in development and over a shorter time when compared with mammals, reflecting the highly precocial state of the young and the need for a fully functional respiratory system and surfactant system at hatching in order to flee the nest.

Tachykinin receptors and actions in the chicken intestine

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The tachykinin peptides neurokinin A (NKA) and [Arg³]-substance P ([Arg³]-SP) have been isolated from the chicken small intestine. In this study, we investigated the tachykinin receptors in chicken intestine using [¹²⁵I]Bolton-Hunter substance P ([¹²⁵I]BH-SP), and examined the actions of [Arg³]-SP and other tachykinins in isolated segments of the ileum. In vitro autoradiographic studies showed localisation of binding sites over myenteric ganglia and the inner circular muscle layer. Binding of [¹²⁵I]BH-SP to chicken ileal membranes was rapid, saturable, of high affinity and to a single population of binding sites with K_d 0.72 nM and B_{max} 0.48 fmol/ wet weight tissue. The rank order of competitors was [Sar⁹]-SP > [Arg³]-SP > SP > [Pro⁹]-SP ≥ NKA > eledoisin > [Sar⁹,Met(O₂)¹¹]-SP >> [Lys⁵,MeLeu⁹,Nle¹⁰]-NKA(4-10) >> senktide, suggesting similarities to the mammalian NK₁ receptor. Tachykinin receptors antagonists were weak or ineffective competitors. The radioligand [¹²⁵I]NKA showed no specific binding. Isolated ileal longitudinal segments responded to tachykinins with dose-dependent contractions (NK₁ > NK₂ agonists). Responses to [Arg³]-SP, NKA and senktide were greatly reduced by tetrodotoxin, suggesting that neurally-mediated responses were primarily involved. NK₁ receptor antagonists were ineffective. [Arg³]-SP and NKA acted mainly by increasing release of acetylcholine, prostaglandins and probably tachykinins. Senktide-induced contractions were inhibited by the NK₃ receptor antagonist, SR142801, but were unaffected by atropine or L-NAME. The study provides evidence for an [Arg³]-SP preferring tachykinin receptor with similarities to the NK₁ receptor in the chicken small intestine. In addition, senktide may act on a receptor similar to the mammalian NK₃ receptor.

Isolation and contractile activities of a substance P-related peptide from the gut of the Australian lungfish, *Neoceratodus forsteri*Lu Liu¹, J. Michael Conlon², Jean M.P. Joss³ and Elizabeth Burcher¹

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The tachykinins are phylogenetically very ancient neuropeptides and have been isolated from a diverse array of species from invertebrates and vertebrates. Using well established techniques (1), a peptide with mammalian substance P (SP)-like immunoreactivity was isolated from an extract of the spiral intestine of the Australian lungfish, *Neoceratodus forsteri*. The primary structure of this peptide was established as Lys-Pro-Arg-Pro-Asp-Glu-Phe-Tyr-Gly-Leu-Met-NH₂, showing 64% identity with mammalian SP. In isolated preparations of lungfish foregut circular muscle, lungfish SP produced a slow, long lasting tonic contraction, with an EC₅₀ value of 6.4 nM. Lungfish midgut circular muscle preparations responded to lungfish SP rapidly and in a more complex manner. There was an increase in the frequency of spontaneous activity (EC₅₀, 1.7 nM), associated with diminished amplitude of the spontaneous contractions (EC₅₀, 0.6 nM), also coupled with a tonic contraction (EC₅₀, 3.7 nM), in some preparations. The response patterns of foregut and midgut circular muscle to acetylcholine (ACh) were very similar to those seen to lungfish SP. Lungfish SP and ACh, however, had very weak effects on both foregut and midgut longitudinal muscle. These data demonstrate that lungfish SP may be a physiologically important regulator of gastrointestinal motility in *Neoceratodus*. This study further confirmed that the structures of SP-related peptides have been strongly conserved under the pressure of vertebrate evolution. The sequence of lungfish SP is identical to that of bufokinin, a SP-related peptide previously isolated from the intestine of the cane toad, *Bufo marinus* (1), supporting the hypothesis that lungfishes and amphibians share a common ancestor.

Postnatal development and control of the pulmonary surfactant system in the Tammar wallaby, *Macropus eugenii*

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Marsupials are born at an early stage of development and are adapted for future development inside the pouch. Whether the pulmonary surfactant system is fully established at this altricial stage is unknown. This study correlates the presence of the surfactant proteins (SP-A, SP-B and SP-D), using immunohistochemistry, with the *ex-utero* development of the lung in the Tammar wallaby, *Macropus eugenii*, and also determines the control of phosphatidylcholine (PC) secretion from developing alveolar type II cells. All three surfactant proteins were found at the site of gas exchange in the lungs of joeys at all ages, even at birth when the lungs are in the early stages of the terminal air-sac phase. Co-cultures of alveolar type II cells and fibroblasts were isolated from lungs of 30 and 70 day old joeys and incubated with the hormones, dexamethasone (10 μ M), prolactin (1 μ M) or triiodothyronine (100 μ M), or the autonomic secretagogues, isoproterenol (100 μ M) or carbamylcholine chloride (100 μ M). Basal secretion of PC was greater at 30 days of age than at 70 days. Co-cultures responded to all five agonists at 30 days of age, but only the autonomic secretagogues caused a significant increase in PC secretion at 70 days of age. This demonstrates that as the cells mature, their activity and responsiveness is reduced. The presence of the surfactant proteins at the site of gas exchange at birth suggests that the system is fully functional. It appears that surfactant development is coupled with the terminal air-sac phase of lung development rather than birth, the length of gestation or the onset of airbreathing.

Expressional analysis of the natriuretic peptide system in the cane toad, *Bufo marinus*, in response to volume loading

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The natriuretic peptide (NP) system in mammals functions to reduce blood pressure and blood volume, by inducing vasodilation in the blood vessels and natriuresis and diuresis in the kidneys, respectively. Atrial natriuretic peptide (ANP) is the principal NP in the control of blood volume regulation, and predominately binds to two types of receptors, NPR-GC, which are guanylyl cyclase linked receptors and NPR-C, a clearance receptor. The biological actions of ANP are mediated through the NPR-GCs, activation of the guanylyl cyclase, and the resulting increase of the second messenger, cGMP. In lower vertebrates however, this system is poorly understood, with the need to regulate body fluid due to their amphibious lifestyle, anurans present themselves as model organisms to study body fluid regulation. This study was therefore designed to increase our understanding of the NP system and its role in body fluid regulation in lower vertebrates. To do this, the study was divided into two parts, firstly isolate and clone toad ANP cDNA, and secondly, monitor mRNA expression of ANP, NPR-GC and NPR-C in response to volume loading conditions. A partial mRNA sequence of toad ANP was cloned using degenerate PCR primers based on existing amphibian ANP sequences. Once this sequence was obtained a semi-quantitative PCR assay was developed to measure mRNA expression for ANP, NPR-GC and NPR-C. Isotonic volume loading caused a significant decrease in both NPR-GC and NPR-C transcripts; data for ANP expression is still being collated. These results suggest that ANP is secreted in response to an increase in blood volume, and that the receptors, NPR-GC and NPR-C are down-regulated, in what we believe is a ligand dependant regulation event. This data suggests that the amphibian NP system responds in a similar manner to the mammalian NP system.

Possible maternal-fetal relations in antifreeze production in the eelpout *Zoarces viviparus*

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Recently Sørensen & Ramløv (2001) reported the presence of antifreeze proteins in the European eelpout *Zoarces viviparus*. This viviparous teleost inhabits the coastal areas of the North Atlantic and Baltic region. The female has a single ovary in which the oocytes are fertilised internally shortly after ovulation in August-September. The embryos remain incubated in ovarian fluid for about 4 months until parturition in shallow waters during winter. Absorption through the alimentary canal is considered the principal pathway by which the embryos secure nutrition from the ovarian fluid (Kristoffersen et al. 1973). Antifreeze activity was measured in serum from pregnant females and embryos during their embryological development inside the ovary and found to be up to 0.9°C and 0.8°C respectively. Lower levels of antifreeze activity (up to 0.3°C) were discovered in ovarian fluid. SDS-PAGE of ovarian fluid shows few proteinbands during embryo development in contrast to adult and embryo serum. These findings indicate that the embryos synthesise their individual antifreeze defence, possibly utilising low molecular weight substances such as amino acids and peptides present in the ovarian fluid. Embryo serum and ovarian fluid in physiological concentrations showed distinct bipyramidal ice crystal morphology. The bipyramidal ice crystal morphology was less pronounced in adult serum as compared to ovarian fluid. This could be due to the higher concentration of antifreeze protein in serum compared to ovarian fluid or more interestingly that a more complex mixture of antifreeze proteins in serum co-operate in binding several planes of the ice crystal.

Trawl stress in Degens leatherjacket (*Thamnaconus degeni*)

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A part of the catch in a trawl fishery is discarded as by-catch. The species that are discarded either have no commercial value, or are prohibited under fishery regulations. A prominent part of the by-catch of the South Australian prawn fishery is Degens leatherjacket *Thamnaconus degeni*. Although little is known about the physiological state of these fish following their capture, they appear alive when discarded. Stress is known to cause major physiological changes in fish, which may result in death. The current research examined the stress level of discarded Degens leatherjackets in order to establish if there is a relationship between trawl time and fish stress. Fish were collected by trawling in the Spencer Gulf of South Australia in March 2001. Three different trawl times (0, 15, and 45 minutes) were carried out. Muscle and blood samples were immediately collected from trawl captured fish and analysed for adenylates, glycogen and lactate, and plasma lactate and glucose. Our data shows that muscle lactate concentrations increased and glycogen decreased as a function of trawling time. AEC values decreased and IMP load increased with increased trawl time. Blood glucose levels fluctuated with trawl time while blood lactate increased. These results indicate that discarded Degens leatherjackets are subjected to considerable stress during trawling, which clearly increases with trawl time, and is likely to contribute to the incidence of death in discarded fish.

**Branchial reprocessing of the urine in the terrestrial Christmas Island red crab,
*Gecarcoidea natalis***

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Crabs invariably produce urine isosmotic to their haemolymph but terrestrial crabs can conserve salts by diverting the urine over the gills to form a modified final excretory fluid, termed P. Homeostatic aspects of P production in *G. natalis* were investigated by switching their drinking water from fresh water (FW crabs) to 70% sea water (SW crabs) and by measuring chloride fluxes following short- and long-term salt loading in crabs with branchial chambers perfused with iso-ionic saline. Crab drinking fresh water released dilute P ([Cl] 10 mmol L⁻¹). Supplying sea water caused the [Cl] of the P to rise (mean 376 mmol L⁻¹ after 5 d), approaching haemolymph [Cl]. FW crabs with perfused branchial chambers initially reabsorbed chloride at a high rate (10 mmol kg⁻¹ h⁻¹) causing haemolymph [Cl] to increase at 20 mmol L⁻¹ h⁻¹ but SW crabs, (with elevated haemolymph osmolality and ion concentrations) showed zero branchial chloride uptake. Self-loading of FW crabs with salt during prolonged perfusion caused down-regulation of chloride uptake, and cessation after several hours. Rates of down-regulation, but not initial chloride fluxes, were dependent on initial haemolymph [Cl]. Intravascular injection of NaCl (raising haemolymph [Cl] by about 80 mmol L⁻¹ in a few minutes) caused an immediate 80% reduction of branchial chloride uptake. Dopamine up-regulated branchial chloride transport in *G. natalis*. This is consistent with the osmoregulatory role of this neurohormone in euryhaline marine brachyurans but contrasts with a reported inhibitory effect in the anomuran *Birgus latro*. Dopamine also stimulated increased urine formation in *G. natalis*.

Circulatory effects of clamping to the substratum in the paua, *Haliotis iris*

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The usual avoidance strategy of abalones is to clamp down to the substratum. Contraction of the massive shell adductor muscle and the foot generate suction, while helping to retract the body into the shell cavity. The adductor muscle has a high anaerobic capacity and is believed to be poorly perfused, or isolated, from the main circulation during clamping. We observed that, even in the resting (unclamped) abalone, the adductor muscle circulation was remarkably isolated. The half time for complete mixing of ¹⁴C-inulin between the central vasculature and the adductor muscle haemocoel was 3.7 ± 0.9 h and was not significantly elevated in clamped animals (5.5 ± 2.4 h; paired *t* test). However, injection of fluorescent microspheres (15 µm) into the aortic stream before and during sustained clamping indicated that the relative blood flow to the foot and adductor muscles was decreased. Prolonged or repeated clamping was associated with decreased haemolymph volume. The ¹⁴C-inulin space decreased from 52.2 ± 3.5 ml (100 g tissue)⁻¹ at rest to 41.1 ± 1.2 ml (100 g tissue)⁻¹ during 2 h sustained clamp. This was associated with an increase in the inulin clearance (from 10.6 ± 1.5 to 15.8 ± 1.5 ml (100 g tissue)⁻¹ d⁻¹), with haemoconcentration (7% increase in [Haemocyanin]), and with a decrease in the total tissue mass (to 89.6 ± 2.0 % of the undisturbed value). A decrease in body volume probably facilitates retraction of the whole animal into the shell cavity, which, in the undisturbed animal, is too small to accommodate it.

The potential uses of an antimicrobial, silver coated mesh (X-static™) in aquaculture

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The intensification of finfish aquaculture systems worldwide has led to an increase in the prevalence of disease problems caused by microbial agents, and stimulated the requirement for the direct application of antimicrobial chemicals to treat the livestock at various life-stages. This process however, can in some cases be detrimental to the health of the livestock and may ultimately reduce the quality of the final product. These inherent problems have stimulated the search for alternative methods of disease prevention and treatment. The antibacterial properties of silver and silver coated materials have been well characterised, however the antibacterial use of silver in the aquaculture industry has not been investigated. This study examined the potential uses of a silver coated nylon mesh (X-static™) for various applications in intensive finfish aquaculture. Exposure of goldfish, rainbow trout and brown trout eggs and / or embryos to X-static mesh resulted in no significant change in egg viability and subsequent embryo development to hatch. However, *in situ* exposure of zebrafish embryos to X-static and exposure of zebrafish embryos to X-static “conditioned” culture water resulted in a significant reduction in embryo survival and percentage hatch respectively (72 hour LD₅₀ < 0.4 mg total silver L⁻¹). X-static was also found to have significant antimicrobial activity against plate and liquid cultures of *Pseudomonas putida* and natural bacterial, yeast and fungal populations isolated from aquarium systems. In summary, the silver-coated X-static mesh was shown to have antibacterial activity against freshwater and saltwater bacteria, yeast and fungi however, the study also revealed evidence regarding toxicity to the non-target livestock. For this reason X-static, when used in the capacity detailed in this study, was not considered an effective alternative to existing chemical antimicrobial treatments.

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