



Image on reverse: Australian Old Endemic rodent, *Notomys fuscus*.
Image modified from Strahan 2002.

Molecular evolution of the carboxy terminal,
the putative sperm-ZP binding site,
of the zona pellucida 3
glycoprotein in Old World murine rodents

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Abstract

In mammals, before fertilisation can occur, sperm have to bind to, and penetrate, the extracellular coat of the oocyte, the zona pellucida (ZP). In the laboratory mouse, which has been used as a model system for fertilization studies, sperm-ZP binding has been found to be mediated by a region near the carboxy terminal, encoded by exon 7 of the *Zp3* gene. This region shows considerable interspecific sequence diversity in North American cricetid rodents, with some evidence of adaptive evolution, suggesting that this may contribute to species specific sperm-ZP binding. However, by contrast, in a preliminary study of three species of Australian murine rodents an identical protein sequence of the region encoded by exon 7 of *Zp3* was found to be present.

The aim of this present study was to determine the pattern of sequence diversity of this region in the most speciose subfamily of mammals, the murine rodents, and to obtain insight into the selective pressures involved in its evolution. For this, DNA was extracted from murine rodents of Africa, Eurasia, South-east Asia, New Guinea and Australia. The nucleotide and predicted amino acid sequence of exons 6 and 7 of *Zp3* in 96 murine species from 14 divisions, as recently defined by Musser and Carleton (2005), was determined and compared.

Generally, it was found that closely related species shared a highly similar ZP3 sequence. Maximum likelihood analyses of codon substitution models using representatives from 14 murine divisions, suggested that positive selection had occurred within only a few lineages at several different codon sites adjacent to, or within, the putative combining-site for sperm of ZP3. Positive selection was not evident when the analysis was restricted to the Australian taxa which showed low levels of both intra- and inter-generic sequence divergence. There was no good evidence that this region contributes to species specificity of sperm-ZP binding in these species.

These findings thus suggest that the selective forces acting on the *Zp3* exon 7 region during the evolution of the murine rodents have varied possibly due to a range of selective pressures not necessarily restricted to the prevention of hybridization. It seems unlikely, therefore, that the amino acid sequence of the exon 7 coding region contributes to species specificity of sperm-ZP binding within most of the lineages from this most speciose subfamily of eutherian mammals.

Declaration

This work contains no material which has been accepted for the award of any other degree or diploma in any university or other tertiary institution and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference has been made in the text.

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Christine A. Swann

Dated

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Publications

Preliminary study published during course of PhD:

Swann, CA, Hope, RM & Breed, WG 2002, 'cDNA nucleotide sequence encoding the ZPC protein of Australian hydromyine rodents: a novel sequence of the putative sperm-combining site within the family Muridae', *Zygote*, vol. 10, pp. 291-299.

Publications resulting from this thesis:

Swann, CA, Cooper, SJB & Breed, WG 2007, 'Molecular evolution of the carboxy terminal region of the zona pellucida 3 glycoprotein in murine rodents', *Reproduction*, vol. 133, pp. 697-708.

Conference abstracts:

Swann, CA, Cooper, S, Hope, RM & Breed, WG 2003 Evolution of zona pellucida C glycoprotein in Australasian native rodents. *Proceedings from the 49th Australian Mammal Society Annual Conference*, University of Sydney, Sydney July 2003.

Swann, CA, Cooper, S, Hope, RM and Breed, WG 2004 Evolution of the putative sperm adhesion region of zona pellucida C glycoprotein in murid rodents: Is there evidence for species specificity? *Reproduction Abstract Series* No. 31.

Swann, CA, Cooper, S, Hope, RM and Breed, WG 2004 Glycoconjugates of the zona pellucida of murid rodents: Is there evidence for species specificity? *Reproduction Abstract Series* No. 31.