



Eden Hermsen



THE DIVERSITY OF THE MAJOR HISTOCOMPATIBILITY COMPLEX CLASS II (MHCII) IN WOMBATS

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Old

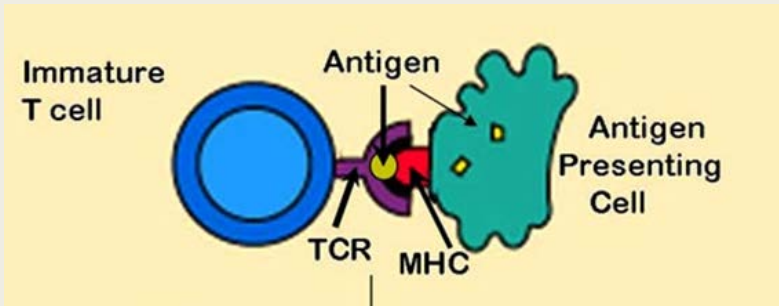
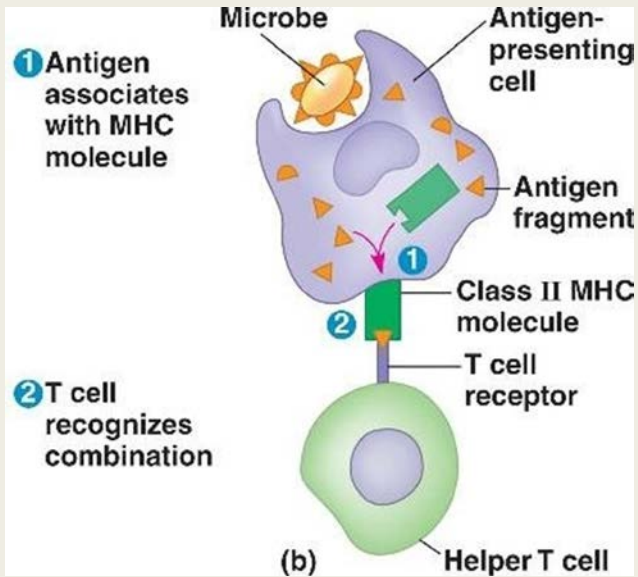


Wombats and Sarcoptic Mange

- Wombats are ‘ecosystem engineers’
 - *burrowing assists soil turnover, nutrient cycling and water infiltration*
- All species highly impacted by anthropogenic changes¹
- Sarcoptic mange: Caused by *sarcoptes scabiei*²
- Found in bare-nosed and southern-hairy nosed wombats
 - *not recorded in northern-hairy nosed wombats*
 - *bare-nosed wombats most impacted*



Photo credit: abc.net.au; Nick Bean



Major Histocompatibility Complex Class II

- Highly polymorphic group of genes
- Codes for proteins on cell surface that aid in recognition of foreign substances ^{3,4}
- Present antigenic peptides to T cells – invoking an immune response
- Three classes
 - *MHCI and MHC II are associated with antigen presentation*
 - *MHCIII is associated in inflammation*
- Focus on MHCII:
 - *Presents exogenously derived peptides to T helper cells*

Why MHC?

- MHC has defined links to population immunity^{5,6,7}
- Highly researched
- MHC diversity should be considered when assessing population health
- Lowered MHC diversity is a compromised capability to respond to environmental changes and emergent diseases
- Lowered MHC diversity compromises the long-term survival of a population



Photo credit: Australian Geographic

Methods

Sample Collection

- Scat samples
- Scat was also collected from captive southern hairy-nosed and bare-nosed wombats at Featherdale Wildlife Park
- DNA from northern hairy-nosed wombat was donated by Dr Andrea Taylor
- Opportunistic tissue samples

DNA Extraction and Sequencing

- DNA extracted by Australian Genomic Research Facility (AGRF)
- NGS done by AGRF for MHCII
- DAB Primers used are known to amplify MHCII in a range of marsupials ⁹

Bio-informatic analysis will involve

- Programs available through Genbank and NCBI databases including BLAST and ClustalW



Photo credit: Julie Old

Results

- Initial results from work by Eden Hermsen ¹⁰
- Total of 44 scats and 40 tissue samples were amplified with DAB primers and sequenced successfully
- Total of 21 variants and 19 potential peptide sequences were found
- DBBE successfully amplified in 6 scats and 2 tissue, 5 variations were found

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      10      20      30      40      50      60      70
.....|.....|.....|.....|.....|.....|.....|.....|.....|.....|.....|.....|.....|
ATGGAGCGCGTGC GG TATGCGGATAGACACATCCACA ACTGCGGGGAGTACGCGCGCTTCGACACCGACTTGC
.C.....A.....T..T.A.G...T..T..T....C.G.A.....G...G...
.....
.....TT.....
.C.....A.....T.AT...C...T..T..T....C.G.A.....A.....G...G...
.....G...G...
.....TT.....
.....G...G...
.C.....A.....C...T.C.C.....T....C.G.A.....G...G...
.....T..T.A.G...T..T..T....C.G.A.....G...G...
.....
.....
.....A.....
.....T..T.A.G...T..T..T....C.G.A.....G...G...
.C.....A.....T.AT...C...T..T..T....C.G.A.....A.....G...G...
VourDBBE04 ..AT.....G.T.....A.....
VourDBBE05 ..AT.....GA.....G.T.....G...C.....
      90      100     110     120     130     140     150     160
VourDBBE01 CAGGAATATGTC CGCTTCGACAGCGACGTGGGGGAGTTCGTTGGCTGTGACGGAGCTGGGGCGGCCACTGCTAAGTACTG
VourDBBE02 .....T.....G.T...G.....C.....A.A.TGG.....
VourDBBE03 .....T.....CA.....GA...G.....
VourDBBE04 .....G.....CA.....GA...G.....
VourDBBE05 G.....T.....GA.....G.....T
      170     180     190     200     210     220     230     240
VourDBBE01 GAACAGTCAGGAGGACTACCTGGAGGAACAACGAGCCTGCGTGGACACGCTGTGCAGACACA ACTACGGGATAGACAAGG
VourDBBE02 .....T.....G.....
VourDBBE03 .....AT.....AGTAC.....GAA.....G.....A.G..T....C
VourDBBE04 .....C.....CTT.CTGGA.AGAAG..G.GCC.AAGTG.ACACGGT.TGCA.ACACA..T..GGGATAGAGA.GC---
VourDBBE05 T.....AT.....AG.A.....GAA.....G.....A.T...GA...C

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Ongoing study



Photo credit: JJ Harrison; Wikipedia.com

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- Initial study used PCR and traditional cloning techniques
- Success using DNA from scats
- NGS used in this study on larger number of samples
 - DNA Extraction and Sequencing
- DNA extracted by Australian Genomic Research Facility (AGRF)
- NGS done by AGRF for MHCII
- DAB Primers used are known to amplify MHCII in a range of marsupials⁹
 - Bio-informatic analysis will involve
 - Programs available through Genbank and NCBI databases including BLAST and ClustalW

Future Directions

- Future results from next-gen sequencing hope to increase accuracy of diversity estimation
- Diversity of MHCII is an important indicator of population immune fitness
- Further correlation to sarcoptic mange instance and MHCII diversity is needed to determine if there is an impact on disease occurrence in wild populations
- Initial results focus on bare-nosed wombats
- Further research needed on hairy-nosed wombats to determine if there is a correlation to mange severity and MHCII diversity

Photo credit: dailymail.co.uk



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- Emirates One&Only Wolgan Valley Resort
- Wombat Protection Society
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- Donations to wombat giving

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QUESTIONS?