

Internet Appendix A27: Immunology

Figure A27.1 Illustrative Pitch Template Example on Dendritic Cells (reverse-engineered)

This example is reversed engineered based on: Tan J. K. H, Quah B. J. C, Griffiths K. L, Periasamy P, Hey, Y-Y, O'Neill HC. (2011). Identification of a novel antigen cross-presenting cell in spleen: a counterpart to cells produced in long-term culture. *J Cell Mol Med*, 15(5):1189-99.

Pitcher's name	Marita Smith	For category	Immunology	Date completed	9/2/15
(A) Working Title	Identification of novel immune cells				
(B) Basic Research Question	What are the uncharacterized dendritic cells that play a role in human immunity and how do they function?				
(C) Key paper(s)	<p>O'Neill H. C, Wilson H. L, Quah B, Abbey J.L, Despars G, Ni K. (2004). Dendritic cell development in long-term spleen stromal cultures. <i>Stem Cells</i>, 22:475-86.</p> <p>Ni K, O'Neill H.C. (2001). Development of dendritic cells from GM-CSF^{-/-} mice in vitro: GM-CSF enhances production and survival of cells. <i>Dev Immunol</i>, 8: 133-46.</p> <p>O'Neill H.C, Jonas N, Wilson H, Ni K. (1999). Immunotherapeutic potential of dendritic cells generated in long-term stroma-dependent cultures. <i>Cancer Biother & Radiopharmac</i>, 14: 263-76.</p>				
(D) Motivation/Puzzle	<p>In humans and mice, the spleen produces a range of cells active in the immune system. A subset of these cells called dendritic cells has previously been characterized into several main categories. Recently, evidence for an unusual type of dendritic cell has been found that plays an important role in inflammatory conditions. How this particular cell lineage develops and functions is not well understood. Exploring and characterizing this new cell could be invaluable in treating disease.</p>				
THREE	Three core aspects of any empirical research project i.e. the "iDioTs" guide				
(E) Idea?	<p>A broad understanding of the immune system is crucial to treating modern disease, including cancer. The immune cells involved in identifying and process foreign antigens are complex. Evidence for an uncharacterized type of dendritic cell requires investigation for a more complete understanding of the human immune response. Cultures containing these novel dendritic cells have begun to be developed, and should be analysed in order to investigate their functionality. Do they display a characteristic phenotype? Do they interact with antigen or other immune cells in a unique way? Can they be harnessed in new health treatments?</p>				
(F) Data?	<ul style="list-style-type: none"> - Long term cultures will be developed from spleens of lab mice in controlled conditions - Data will be sourced from these cultures using a method called flow cytometry. This involves assessing the cell surface marker expression using known antibodies. This will provide a full work-up of the unique capabilities of the novel dendritic cell. - Once the novel cells have been fully characterized <i>in vitro</i>, its role in the immune system can be analysed <i>in vivo</i> using a range of 				

	mice studies. Cells will be taken directly from live splenic samples and analysed using flow cytometry.
(G) Tools?	Physical apparatus will be necessary, including animal facility and analysis equipment (Flow Cytometer). Records of specific immune cells will be generated using laboratory work and computer software to provide raw data for identification of cell subsets. Statistical analysis will also be required.
TWO	Two key questions
(H) What's New?	This novel dendritic cell has not been classified, nor its role in the immune system documented. This project will facilitate its identification using state-of-the-art cytometric technique to separate this cell from known subsets. This will potentially provide data on a new immune cell useful in designing disease treatments or vaccinations.
(I) So What?	Cancer and other diseases are on the rise. Innovation in health is only possible with a firm understanding of the immune system. Targeted immune treatments are effective and necessary. The data derived from this project is likely to inform future health strategies and disease treatments.
ONE	One bottom line
(J) Contribution	The primary source of the contribution will be processed phenotypic data of the new dendritic cell both in culture studies (<i>in vitro</i>) and in mice studies to mimic its role <i>in vivo</i> . It will provide a complete profile of the new cell and its role in the immune system.
(K) Other considerations	<p>Is Collaboration needed/desirable?</p> <ul style="list-style-type: none"> -Idea: no; -Data; yes –multi-institutional preferred -Tools; yes –representatives and funding from various institutions <p>Target journals – <i>Nature</i>, <i>Nature Reviews Immunology</i>, <i>Journal of Immunology</i></p> <p>“Risk” assessment:</p> <ul style="list-style-type: none"> -“no result” risk: moderate. If the new cell cannot be isolated from the splenic cultures, further culturing will be required before analysis is possible. This may require repeat mice studies. -“competitor risk”(i.e. being beaten by a competitor): moderate. The identification of novel immune cells is a focus internationally to aid health and disease treatment. However, it is difficult to establish and maintain <i>in vitro</i> cultures over the long term, and many research groups are primarily interested in other subsets of immune cells such as B and T lymphocytes. Dendritic cells are not as well characterized or understood and do not draw the same level of interest. -risk of “obsolescence”: Low. The immune response is an ever-important and constantly evolving topic worldwide.