

# Professional secrets

Craig R. Roy

Looking inside the compartments of certain immune cells — professional antigen-presenting cells — has revealed how the immune system can trigger a cell-killing response to extracellular pathogens.

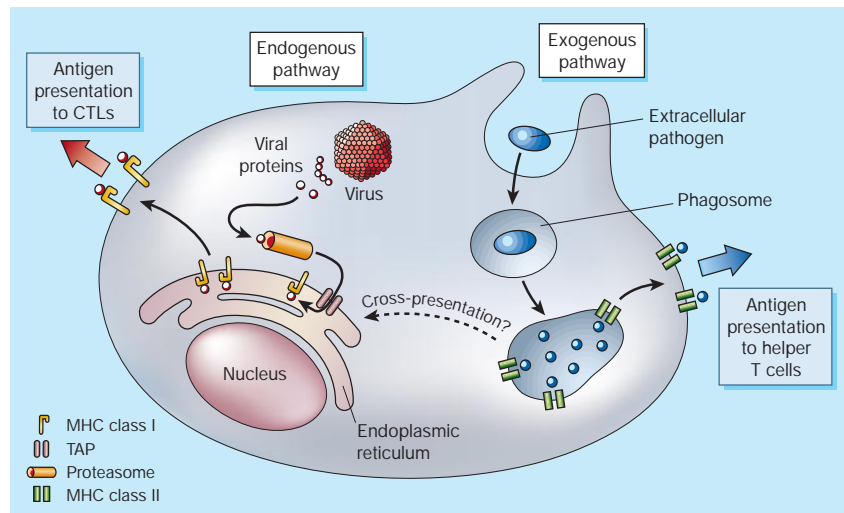
Our immune system provides effective protection against most pathogens. It can mount a highly specific 'adaptive' response that distinguishes between different sorts of pathogens and activates the appropriate mechanisms to eliminate them. Writing in this issue, Amigorena and colleagues<sup>1</sup> and Desjardins and colleagues<sup>2</sup> suggest how an adaptive immune response typically reserved for pathogens that replicate inside immune cells can also be generated against pathogens that exist outside these cells.

Macrophages and dendritic cells are specialized cells that initiate adaptive immune responses. They are sometimes called professional antigen-presenting cells (APCs) because one of their primary duties is to degrade microbial proteins and display the resulting peptide fragments, or antigens, on their surface. The kind of immune response that is then produced depends on how the antigens are displayed by the APCs.

In general, peptide antigens from pathogens replicating inside APCs (endogenous antigens) are transported into a cellular compartment, the endoplasmic reticulum (ER), and bind to a protein complex called MHC class I. From the ER, the MHC class I–antigen complex is shuttled to the cell surface, where it stimulates antigen-specific cytotoxic T lymphocytes (CTLs) — killer cells that seek out and eliminate infected cells (Fig. 1).

Antigens from extracellular pathogens are usually processed differently from endogenous antigens, and with different results. Extracellular pathogens are engulfed by APCs and degraded in phagosomes — membrane-bound cellular compartments. Peptide fragments produced in phagosomes are not transported to the ER. Instead, they bind directly to a different set of MHC molecules, MHC class II, inside the phagosome and then return to the cell surface, where they stimulate a subset of helper T cells (Fig. 1). Unlike the CTLs, helper T cells do not kill other cells, but instead produce chemical signals. These trigger the production of antigen-specific antibodies and elicit an inflammatory response that eliminates the microbial pathogens.

So the rules governing antigen presentation seem to be simple — endogenous antigens are presented by MHC class I molecules to stimulate a cell-killing response, and exogenous antigens are presented by MHC class II molecules to stimulate a helper



**Figure 1 Professional antigen-presenting cells process intracellular and extracellular pathogens differently. In the endogenous pathway, proteins from intracellular pathogens, such as viruses, are degraded by the proteasome and the resulting peptides are shuttled into the endoplasmic reticulum (ER) by TAP proteins. These peptides are loaded onto MHC class I molecules and the complex is delivered to the cell surface, where it stimulates cytotoxic T lymphocytes (CTLs) that kill the infected cells. In contrast, extracellular pathogens are engulfed by phagosomes (exogenous pathway). Inside the phagosome, the pathogen-derived peptides are loaded directly onto MHC class II molecules, which activate helper T cells that stimulate the production of antibodies. But some peptides from extracellular antigens can also be 'presented' on MHC class I molecules. How this cross-presentation occurs has now been explained<sup>1,2</sup>. It seems that by fusing with the ER, the phagosome gains the machinery necessary to load peptides onto MHC class I molecules (see Fig. 4g on page 405).**

response. But in reality, the situation is not so straightforward. Professional APCs can break these rules and present exogenous antigens on MHC class I molecules<sup>3</sup>, in a process known as cross-presentation. The cross-presentation pathway seems to be required for CTL responses to certain exogenous antigens<sup>4,5</sup>. For example, some viruses do not infect professional APCs, but in order to generate a CTL response that will eliminate virus-infected cells, the APC must present exogenous viral proteins on MHC class I molecules. Cross-presentation might also be important for triggering CTL responses to some cancer cells.

So how do exogenous antigens cross over to the MHC class I pathway? Amigorena and colleagues had previously shown<sup>6</sup> that exogenous proteins could escape from phagosomes and enter the cytosol (the intracellular space). So it was thought that cross-presentation might simply involve the transport of exogenous proteins from the phagosome into the cytosol, where they

could join the pathway used to process endogenous antigens. This view was supported by data<sup>7,8</sup> showing that cross-presentation depends on two components of the MHC class I pathway — the proteasome complex and the 'transporter associated with antigen presentation' (TAP). The proteasome complex degrades proteins in the cytosol and TAP shuttles the resulting peptide antigens into the ER for loading onto MHC class I molecules. Furthermore, cross-presentation is sensitive to the drug brefeldin A (refs 7, 8), which disrupts the transport of the MHC class I–antigen complex from the ER to the cell surface<sup>9</sup>.

Desjardins and colleagues, meanwhile, had provided a clue to how exogenous antigens might escape from the phagosome<sup>10,11</sup>. In an effort to understand how phagosomes form and mature, these authors had catalogued the proteins contained on phagosomes that had been purified from macrophages. Surprisingly, they detected proteins that were thought to reside only in the ER<sup>10</sup>. Further analysis

revealed that phagosomes fused with the ER during, or shortly after, pathogen engulfment at the cell surface<sup>11</sup>. This was an important finding because a protein-translocation channel called the Sec61 complex is embedded in the ER membrane. Sec61 both imports newly synthesized proteins into the ER and exports proteins from it, targeting the proteins for degradation by the proteasome<sup>12,13</sup>. So it was predicted that the Sec61 complex might be delivered to the phagosome after fusion with the ER. If true, it could be the missing link in the cross-presentation pathway — the transporter responsible for exporting exogenous proteins from the phagosome into the cytosol.

The two groups<sup>1,2</sup> now provide evidence to support this hypothesis. Looking at phagosomes from dendritic cells<sup>1</sup> and macrophages<sup>2</sup>, respectively, they showed that Sec61 is indeed present on the phagosome membrane. And both groups found that a fluorescently tagged version of the protein ovalbumin could be exported from the phagosome into the cytosol. But did protein export involve Sec61? To find out, Desjardins and colleagues looked at the export of the cholera toxin A1 subunit (CTA1). This protein moves from outside the cells to the ER and is then exported from the ER into the cytoplasm through the Sec61 channel<sup>14,15</sup>. The authors observed that CTA1 could also be exported from the phagosome into the cytoplasm, which strongly suggests that, after Sec61 is delivered to the phagosome, it can still export proteins.

What happens to proteins once they have been exported from the phagosome? First, they must be turned into peptide antigens by the proteasome. Desjardins and colleagues showed that proteasomes are associated with the cytosolic side of the phagosome membrane. And both groups showed that TAP and the MHC class I molecules are delivered to the phagosomes and remain active. Their further analyses revealed that the peptide antigens are loaded onto MHC class I molecules inside the phagosome.

So it seems that after the exogenous proteins are exported from the phagosome through the Sec61 channel, they are degraded by the proteasome and the resulting peptide antigens are shuttled back into the phagosome by TAP. There, they are loaded onto MHC class I molecules on the inside of the phagosome membrane. The results confirm observations<sup>16</sup> that the phagosome is a fully competent antigen-processing compartment for the MHC class I pathway.

The new studies provide strong support for a model of cross-presentation in which ER-phagosome fusion occurs. But they do not show that such fusion is necessary. Arguments have also been made that cross-presentation occurs only in cultured cells and might not be relevant in animals<sup>17</sup>. Now that a molecular mechanism governing cross-presentation has been proposed,

experiments to test the importance of this immunological process are certain to follow. But the debate on cross-presentation continues — to resolve it, the professionals will have to reveal all of their secrets. ■

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1. Guernonprez, P. *et al. Nature* **425**, 397–402 (2003).
2. Houde, M. *et al. Nature* **425**, 402–406 (2003).
3. Bevan, M. J. *J. Exp. Med.* **143**, 1283–1288 (1976).
4. den Haan, J. M. & Bevan, M. J. *Curr. Opin. Immunol.* **13**, 437–441 (2001).
5. Sigal, L. J., Crotty, S., Andino, R. & Rock, K. L.

- Nature* **398**, 77–80 (1999).
6. Rodriguez, A., Regnault, A., Kleijmeer, M., Ricciardi-Castagnoli, P. & Amigorena, S. *Nature Cell Biol.* **1**, 362–368 (1999).
7. Brossart, P. & Bevan, M. J. *Blood* **90**, 1594–1599 (1997).
8. Kovacsics-Bankowski, M. & Rock, K. L. *Science* **267**, 243–246 (1995).
9. Nüchtern, J. G. *et al. Nature* **339**, 223–226 (1989).
10. Garin, J. *et al. J. Cell Biol.* **152**, 165–180 (2001).
11. Gagnon, E. *et al. Cell* **110**, 119–131 (2002).
12. Koopmann, J. O. *et al. Immunity* **13**, 117–127 (2000).
13. Romisch, K. J. *Cell Sci.* **112**, 4185–4191 (1999).
14. Roy, C. R. *Trends Microbiol.* **10**, 418–424 (2002).
15. Schmitz, A., Herrgen, H., Winkler, A. & Herzog, V. J. *Cell Biol.* **148**, 1203–1212 (2000).
16. Ramachandra, L., Song, R. & Harding, C. V. *J. Immunol.* **162**, 3263–3272 (1999).
17. Zinkernagel, R. M. *Eur. J. Immunol.* **32**, 2385–2392 (2002).

## Plasma physics

# Cosmic waves in the lab

Rod Boswell

An Alfvén-wave maser, a feature of atmospheric and astrophysical science, has been created in a laboratory, and opens the way for further Earth-bound investigations of cosmic phenomena.

In general, to be an observational astronomer is to be a spectroscopist, unravelling the workings of the cosmos through the varying wavelengths of radiation detected. Observations that began at optical wavelengths now extend to wavelengths ranging from hundreds of kilometres down to tens of nanometres, and satellites have also proved immensely useful in refining our image of the Universe. But, despite the assiduous measurements, the radiation is often produced, particularly at radio and X-ray wavelengths, by very complex processes that we struggle to understand. Nevertheless, a positive step has now been taken: in *Physical Review Letters*, J. E. Maggs and G. J. Morales report the resonant amplification of a typical astrophysical wave in their laboratory — an Alfvén-wave maser

(*Phys. Rev. Lett.* **91**, 035004; 2003).

For some years, the aim of these authors has been a laboratory study of magneto-hydrodynamic turbulence — that is, turbulence in the interactions between a plasma and a magnetic field. The first step on the way is to create the basic element of the phenomenon, an Alfvén wave. Postulated in 1942 by Hannes Alfvén (*Nature* **150**, 405–406; 1942), an Alfvén wave is a low-frequency electromagnetic wave that can be generated in magnetized plasmas throughout the Universe. These waves are thought to be intimately involved in diverse phenomena, such as the precipitation of electrons through the atmosphere in the auroral regions that are connected with the dazzling northern and southern lights — and that can take out power distribution systems. Alfvén waves are also implicated in the heating

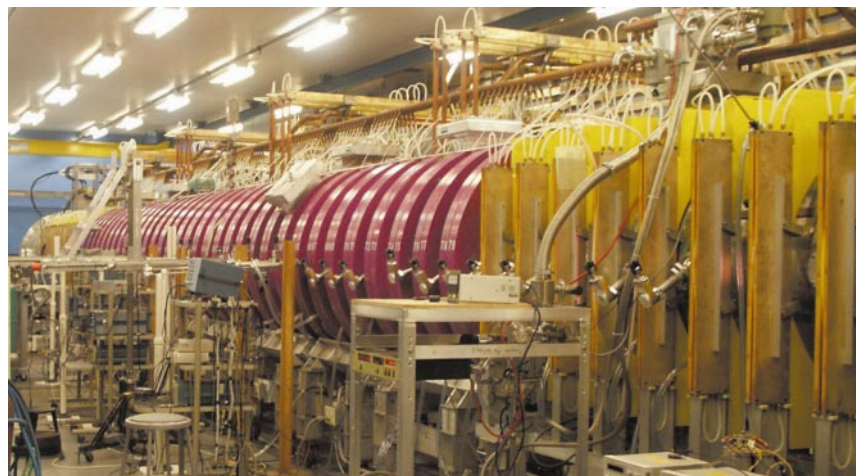


Figure 1 Unique facility — the Large Plasma Device at the University of California, Los Angeles. In a helium plasma inside this 19-metre-long column of magnets, Maggs and Morales have created an Alfvén-wave maser.