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Titre du stage : Mechanosensing in T cell activation : looking at T cell / APC interface

Scientific context

Cell/cell interactions are a paradigm in cell biology since they establish, through biochemical and mechanical contacts, the structures of the living, allowing the cell's function to take place and potentially synchronise their action. While the biochemical side has been intensively studied, the mechanics , and its articulation with biochemistry, together termed as mechanotransduction, is a current topic of enormous interest. , Its importance is more and more recognized to be instrumental to finely regulate cellular functions. The interface formed between a T cell and an antigen presenting cell during immune response, often called the immune synapse, is a prototypical interaction where mechanotransduction is a key regulating mechanism. . This particular moment is crucial for adaptive immunity as this is when the pathological antigen is recognized by the T cell and the decision is taken whether to mount a response or not. The formation of the immune synapse ultimately leads to the production of inflammatory signals and mutual exchange of signals between the cells, as a prelude to the elimination of the pathogenic condition [3].

Positioning and preliminary data

In order to observe the mechanical modulations during early T cell activation, at the interface with an APC, we recently used AFM Single Cell Force Spectroscopy. We immobilized 3A9 murine CD4+ T cell hybridomas on an aCD45 coated surface which density does not prevent efficient activation of the T cells by soluble aCD3 or peptide loaded APCs in suspension. To prevent the rise of signaling cascades, our measurements were performed in presence of the pan-Src kinase inhibitor PP2. In addition our surrogate APCs have been extensively documented to be devoid of co-stimulation and adhesion molecule that could interfere with TCR/pMHC interactions

We established that the contact mechanics was, as expected, not modified by the presence or absence of peptide on the APC. Then, by measuring the forces during the separation of APC and T cell, we observed that the maximal detachment force was significantly increased when a peptide was present, with an effect that shown to be peptide dependant. The forces for the small events, potentially single pMHC/TCR detachments, were on the same range as the one we reported earlier. All of these elements support the peptide specificity of our SCFS experiments. We observed a striking and unexpected difference in the force relaxation shortly after the cell/cell contact when APCs were loaded with a saturating concentration of antigenic peptides. We made a very simple, model independent, quantification of the relaxation and obtained that the decrease was, indeed, significantly affected by the presence of peptide, indicating that mechanics during the APC/T cell contact was modulated, even at very short time scales (below 10 sec) without any biochemical signalling (since the presence of PP2 prevents it). Essentially, the present data points toward a more viscous behaviour triggered when a peptide is recognized at the interface between a T cell and an APC. An article is in preparation. During this relaxation, we observed some weak force fluctuations that appeared to be also peptide dependant, with a periode of ~ 5-10 sec, and amplitude of ~ sev. 10 pN.

In the frame of this present internship, we would like to confirm and extend the observation and quantification of these mechanical events, which could be related to an active testing of the APC by the T cell and also to an active response to a given peptide, without the need of biochemical signalling.

References

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