VIEWPOINT: The Emerging Fibroblast-like Synoviocyte Channelome.

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Synovial joint arthritis is a leading cause of disability with treatment options being remarkably limited. The synovial membrane lines the joint space, producing synovial fluid to facilitate joint lubrication, and is a tissue richly innervated with nociceptors. It also acts as a key gateway between the avascular joint space and the circulatory system. Therefore, as we work towards new treatments that target joint disease, building an understanding of how the cells within the synovial membrane are regulated is of critical importance. Sadly, whilst we do know a fair amount about the composition and function of the synovium, we still have a lot to learn about how to control the important fibroblast-like synoviocytes within. The latest study by Kondo *et al* 2018 goes some way to address this by developing a fundamentally important working hypothesis for control of synoviocytes that links, purinergic and histamine receptor activation to intracellular calcium mobilisation and cell membrane hyperpolarisation.

Kondo *et al* 2018 use an unusually well laden tool bag of cell physiological techniques to put this schema together; with a panel of 19 qPCR primer pairs, intracellular Ca^{2+} imaging, flash photolysis of caged ip3 and of course patch-clamp electrophysiology. Two separate inflammatory mediators, ATP and histamine both act in parallel to trigger PLC activation, IP3 elevation, thence Ca^{2+} elevation and activation of a potassium conductance. A particular strength is that the entire model is built in human fibroblast-like synoviocytes bringing two strengths over previous models, which have tended to be based on either healthy rodent joints or rheumatic human joints. Firstly, in the absence of specific markers for synovial fibroblasts, the use of tissue from a larger organism ensures that there can be greater confidence that the fibroblastic population being cultured is chiefly of synovial origin. Secondly, it is of course vital to work-out what happens in a rheumatic joint, but surely the ideal starting models would be the normal "physiological" system in a healthy synovium.

How does Kondo *et al* 2018's new working hypothesis fit with the existing literature? Rather well it turns out. It is known that during inflammation, histamine is released and ATP levels also increase as the nucleotide is released, typically, through activated pannexin channels. This paper shows, moderate levels of pannexin expression (PANX1 by qPCR), together with that of a connexin (CX43), which could hypothetically form an alternative ATP release conduit. There was also high expression of both intermediate ($K_{Ca}3.1$) and large (BK/$K_{Ca}1.1$) Ca^{2+}-activated potassium channels. Encouragingly for those working with other model systems these ion channels were identified in both rheumatoid arthritis-derived and rodent model FLS studies (Friebel et al., 2014; Hu et al., 2012; Tanner et al., 2015). The other most highly expressed ion channels (again identified by qPCR) were KCNK2, ANO6, ANO10 and KCNK6. KCNK2/6 are members of the twin-pore potassium channel family and are particularly thought of as molecular sensors, whereas the ANO (anoctamin) channels are members of the large chloride channel family. The family is relatively understudied compared to potassium channels, but ANO6 (TMEM16F) is, interestingly, thought to be a Ca^{2+}-activated chloride channel likely, therefore, to be activated in parallel to Ca^{2+}-activated potassium channels.

Where next? Well this paper documents the all-important control of synoviocytes rather than how that system will practically influence joint function or even joint pain, but it does not take much extrapolation to contextualise this. Activation of Ca^{2+}-activated potassium channels somehow drives invasiveness of synoviocytes and progression of arthritis in both human and rodent RA models by increasing invasiveness, proliferation and production of both inflammatory mediators and catabolic enzymes (Friebel et al., 2014; Hu et al., 2012; Tanner et al., 2015). But that does leave further questions; how does activation of K_{Ca} and hyperpolarisation of a cell drive its *activation*? Is it that hyperpolarisation puts the cell into an active state *per se* …or is it an analogous mechanism to that we modelled mathematically in neurones where hyperpolarisation serves to draw extracellular Ca^{2+} into the cell (by increasing the driving force for Ca^{2+} entry through a non-voltage gated Ca^{2+} entry pathway (Feetham et al., 2015)). …and why, physiologically speaking, would activation of FLS even cause progression of disease and pain? Or is it the other way around; does activation of FLS result from disease in a compensatory physiological "attempt" to restore joint homeostasis. Investigation of all these questions will now be a far more realistic possibility in the light of the new Kondo *et al* 2018 "working hypothesis" model.

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