Enspire labelfree technology strengthens the importance of galanin receptor 3 in mediating important functions on polymorphonuclear neutrophils

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Galanin is a bioactive peptide that participates in the recruitment and activation of polymorphonuclear neutrophils (PMNs). To date, three galanin receptors (GALR1, GALR2, GALR3), belonging to the G-protein coupled receptor (GPCR) family are known. However, the receptor(s) mediating the effects of galanin are unclear.

To identify the nature of the receptor involved in galanin mediated PMN activation we employed the Enspire cell-based labelfree technology that measures dynamic mass redistribution (DMR) within the cell.

We were able to show that GALR2/3 are found to be expressed in human resting PMNs and western blot analysis confirmed that mRNA detected by RT-PCR was translated into protein. Treatment of PMNs with galanin in combination with IL-8 increases the DMR in a dose-dependent manner suggesting receptor specific activation. Galanin alone, on the other hand, was not able to change DMR of PMNs, which indicates the requirement of certain co-stimulatory factors for activating signaling pathways.

To further investigate the receptor subtype mediating galanin PMN activation, we were using the specific GALR3 antagonist SNAP-37889. We found, that preincubation of PMNs with 10 µM SNAP-37889 for 30 minutes could almost completely block the galanin-IL-8 signal in labelfree assays. Furthermore, SNAP-37889 treatment significantly reduced the secretion of azurophilic granules and beta-2-integrin CD11b expression in galanin stimulated PMNs.

Taken together, we could demonstrate that GALR3 is mandatory for galanin-reliant functions of human PMNs which was further proofed by cell-based labelfree technology.