Prostamide $F_{2\alpha}$ and Bimatoprost induce preadipocyte proliferation and inhibit PPARG activity in a MAPKK-dependent manner

Besma Boubertakh [1,2], David Marsolais [1], Vincenzo Di Marzo [1,2], Cristoforo Silvestri [1,2]

[1] Centre de recherche de l'Institut universitaire de cardiologie et de pneumologie de Québec (CRIUCPQ), Faculté de médecine, Université Laval, Québec, QC, Canada

[2] Canada Excellence Research Chair on the Microbiome-Endocannabinoidome Axis in Metabolic Health, Québec, QC, Canada

Introduction: The World Health Organization defines obesity as excessive fat accumulation that presents a risk to health, however intensive research still fails to define its pathways and treatments. Interestingly, the ophthalmic drug Bimatoprost (Bim.) and the endocannabinoidome lipid mediator prostamide $F_{2\alpha}$ ($PMF_{2\alpha}$), which are chemically and pharmacologically similar, are both anti-adipogenic. It is noteworthy that patients on Bim. initially lose eye fat pads which return upon cessation. Treating 3T3-L1 murine preadipocytes with these drugs to study their anti-adipogenic mechanisms led us to the unprecedented observation of bigger cell pellets, which led us to develop the hypothesis that they might induce cellular proliferation.

Objective: To determine whether Bim./$PMF_{2\alpha}$ cause preadipocyte proliferation, and to identify the potential proliferative and anti-adipogenic mechanisms.

Methods: Tetrazolium dye MTT, crystal violet assays, and cell counting were used to assess cell viability/number, whilst we employed flow cytometry and qPCR techniques to check Bim./$PMF_{2\alpha}$ effects on the cell cycle. We conducted reporter assays to measure the effect on PPARG activity, the master regulator of adipogenesis. The role of mitogen-activated protein kinase (MAPK) kinase was investigated using its selective pharmacological inhibitor PD 98059.

Results: Bim./$PMF_{2\alpha}$ specifically triggered preadipocyte proliferation through activating MAPK signaling. They advanced the preadipocytes in the cell cycle towards S and G2 phases, and decreased *p21* and *p27* cyclin dependent kinase inhibitor gene expression. Furthermore, Bim./$PMF_{2\alpha}$ inhibited PPARG activity dependently on MAPK signaling.

Conclusion and Discussion: These results suggest that $PMF_{2\alpha}$ induction of preadipocyte proliferation and inhibition of adipogenesis together maintain adipose tissue homeostasis, by regulating the cellular pool of preadipocytes ready to differentiate and store lipids healthily through hyperplasia. Dysregulation of this system may result in hypertrophy, which is behind obesity complications. The discovered Bim. proliferative effect might as well explain the quick recovery of patients who utilize Bim.