Istaroxime Effects on Pulmonary Artery Smooth Muscle Cells Ca^{2+} Dynamics

Alessia Metallo[1], Virginia D'Angeli[1], Martina Arici[1], Mara Ferrandi[2], Paolo Barassi[2], Marcella Rocchetti[1]

E-mail: a.metallo1@campus.unimib.it

[1]*Department of Biotechnology and Biosciences, Università degli Studi di Milano-Bicocca, Milan, Italy*

[2]*Windtree Therapeutics Inc., Warrington, Pennsylvania, USA.*[3] *Institution, country*

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Abstract: Smooth muscle cells show a considerable plasticity and can undergo profound and reversible phenotypic changes (from contractile to proliferative) in response to vascular damage or disease, causing alterations in the expression of Ca^{2+} handling proteins, such as SERCA2a, more expressed under smooth muscle cell contractile phenotype than proliferative one. Istaroxime is a luso-inotropic agent endowed of a double mechanism of action consisting in the ability to inhibit Na^+/K^+ ATPase and enhance sarcoplasmic reticulum (SR) Ca^{2+} ATPase (SERCA2a) activity in cardiac preparations. Moreover, istaroxime is reported to reduce store-operated Ca^{2+} entry (SOCE) in prostate cancer cells.

The aim of the project is to investigate the effect of istaroxime in pulmonary artery smooth muscle cells (PASMCs), in order to verify the effect of SERCA2a stimulation in inducing vascular smooth muscle cells contractile phenotype.

First of all, we evaluated the effects of the compound in cell proliferation; then, we focused the attention on intracellular Ca^{2+} dynamics, measuring drug effects on SR function, by analyzing the ATP-induced SR Ca^{2+} release through IP_3 receptors (in the absence of extracellular Ca^{2+}).

Istaroxime affected PASMCs proliferation in a dose-dependent manner. However, the amplitude and kinetic of ATP-induced Ca^{2+} transient was not significantly modified by the drug, both under proliferative (10% FBS) and starving (0.1% FBS) culture conditions. Moreover, SERCA2a and SERCA2b protein levels were evaluated by western blotting, showing that SERCA2b is the prevailing protein isoform at least under proliferative culture condition. Preliminary results aimed to evaluate istaroxime effects on SOCE showed a tendency of drug-induced SOCE inhibition also in PASMCs.

Overall, these preliminary results suggest that istaroxime might affect smooth muscle cells Ca^{2+} dynamics and proliferation through mechanisms apparently not related to SERCA2a stimulation. Further studies are necessary to better investigate drug effects on PASMCs.