CLIC2 MODULATES JAK/STAT SIGNALING CONDITIONING MONOCYTES DIFFERENTIATION IN THE TUMOUR MICROENVIRONMENT

Francesco Albano[1,2,3], Pellegrino Mazzone[3,4], Viviana Longo[1], Pietro Zoppoli[2,5], Giuseppina Di Paola[3,4], Giuseppe Cesta[2], Margherita Luongo[2], Claudia Sabato[2], Giovanni Calice[2], Sabino Russi[2], Simona Laurino[2], Giuseppe Patitucci[2], Chiara Balzamo[1], Valentina Pagliara[6], Giuseppina Amodio[6] and Geppino Falco[1,2,3]

[1] *Department of Biology, University of Naples Federico II, Naples (NA), Italy*

[2] *Laboratory of Preclinical and Translational Research, IRCCS CROB Centro di Riferimento Oncologico della Basilicata, Rionero in Vulture (PZ), Italy*

[3] *Biogem S.c.a.r.l., Istituto di Ricerche Genetiche "Gaetano Salvatore," Ariano Irpino (AV), Italy*

[4] *CaWUR s.r.l., organoids factory, Ariano Irpino (AV), Italy*

[5] *Department of Molecular Medicine and Medical Biotechnologies, University of Naples Federico II, Naples (NA), Italy*

[6] *Department of Medicine, Surgery and Dentistry "Scuola Medica Salernitana", University of Salerno, Baronissi (SA), Italy*

Phone: +39081679083 e-mail: francesco.albano@unina.it

Chloride intracellular channels (CLICs) are a family of six evolutionarily conserved proteins with heterogeneous functions (ion channels, redox proteins, enzymes, scaffolding proteins) and previously, we reported that the family member *CLIC2* was upregulated in gastric cancer (GC). To investigate Clic2 function in GC, we first determined Clic2 distribution in normal and in GC human tissues detecting Clic2 signal in dendritic cells (DCs), endothelial (ECs) and macrophages (MCs), with increased intensity in tumour samples. Since both DCs and MCs are derived from the differentiation of monocytes, we used THP-1 cells, a monocytic cell line, to investigate whether Clic2 could have a role during differentiation or in the function of those cells. We started defining Clic2 intracellular localization in differentiated naïve cells, finding it expressed in the Golgi apparatus and in the plasma membrane. Next, we generated *CLIC2*-KO THP-1 cells to explore cell differentiation mechanisms and functions. Differentiated naïve KO cells, exhibited a different morphology, suggestive of an activated DC phenotype, as confirmed by increased expression of CD11c, CD80 and CD86 markers. In addition, when we characterized cytokines secretion and Jak/Stat signalling, we observed in KO differentiated naïve cells the increase of chemotactic cytokines CCL7 and CCL8, the reduction of IL-6 secretion, increased phosphorylation of Shp1/Shp2 phosphatases and the absence of Stat3 phosphorylation with the resulting impairment of its signalling. We thereby suggest that Clic2 plays a central role in regulating DCs differentiation and function, by the modulation of inhibitory signals of the Jak/Stat pathway contributing to support GC progression by tumour a tumour-permissive microenvironment.