ABSTRACT

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Grant Title: Unravelling ClC-5 role on glycosylation and its link with proximal tubule functioning

The Cl^-/H^+ antiporter ClC-5, encoded by *CLCN5* gene, is a key element for the acidification control of the endolysosomal system and renal proximal tubule function. The proximal tubule (PT) is the region of the nephron (kidney functional unit) that reabsorbs low molecular weight proteins and other molecules that scape the glomerular filtration barrier. Notably, ClC-5 is localised at the apical plasma membrane and endosomes of PT epithelial cells, where it has a crucial role in protein endocytosis. Loss-of-function mutations on *CLCN5* are the genetic cause of **Dent's disease type 1** (DD1), a rare renal disease characterized by hypercalciuria, low molecular weight proteinuria and progression to renal fibrosis and kidney failure.

The main objective of this project was to understand **ClC-5 role on glycosylation** and how loss-of-function mutations cause loss of epithelial markers (such as MUC1), cell polarization and differentiation, leading to PT dysfunction and kidney failure. Our group has used genetically modified cell lines as well as urine samples from DD1 patients, to achieve these aims. During these two years, we have demonstrated that ClC-5 loss-of-function mutations affect the secretory pathway, causing fragmentation of the Golgi apparatus and altering mucin-1 (MUC1) and other epithelial markers trafficking to the plasma membrane, promoting epithelial-mesenchymal transition (**Duran et al, 2024**). We have further studied the role of ClC-5 on glycosylation, our data suggests that some proteins (possibly MUC1) are differently glycosylated in cells expressing ClC-5 loss-of-function mutants. Besides, we have found that **FUT8** (Fucosyltransferase 8), the unique enzyme responsible of core-fucosylation which modulates mucins (reviewed in **Cantero-Recasens, 2024**), is increased in ClC-5 depleted or mutated cells compared to control or ClC-5 WT cells. Notably, we have found that levels of specific glycoproteins are reduced in samples from DD1 patients, which also present lower molecular weight in patients compared to controls, suggesting a defect on glycosylation.

In conclusion, our findings have provided a **new mechanism linking ClC-5 loss-of-function with proximal tubule dedifferentiation**, and we have suggested a potential impact of glycosylation (specially highly glycosylated mucins) in this process.

References

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