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From protein uptake to Dent disease: An overview of the *CLCN5* gene

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Abstract

Proteinuria is a well-known risk factor, not only for renal disorders, but also for several other problems such as cardiovascular diseases and overall mortality. In the kidney, the chloride channel Cl^-/H^+ exchanger C1C-5 encoded by the *CLCN5* gene is actively involved in preventing protein loss. This action becomes evident in patients suffering from the rare proximal tubulopathy Dent disease because they carry a defective C1C-5 due to *CLCN5* mutations. In fact, proteinuria is the distinctive clinical sign of Dent disease, and mainly involves the loss of low-molecular-weight proteins. The identification of *CLCN5* disease-causing mutations has greatly improved our understanding of C1C-5 function and of the ClC-5-related physiological processes in the kidney.

This review outlines current knowledge regarding the *CLCN5* gene and its protein product, providing an update on C1C-5 function in tubular and glomerular cells, and focusing on its relationship with proteinuria and Dent disease.

Keywords

ClC-5; ClC-5 mutant proteins; Kidney; Proximal tubular cells; Podocytes; Endocytosis

1. Introduction

Excess protein in the urine (proteinuria) is a well-known risk factor not only for renal disorders (Tsai et al., 2016), but also for several other problems, such as cardiovascular diseases (Schmieder et al., 2011; Lim et al., 2017; Wang, 2018), bone disorders (Tung et al., 2018); diabetes (Klein et al.,1993); and overall mortality (Grimm et al., 1997).

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CRediT authorship contribution statement

Lisa Gianesello: Conceptualization, Writing - original draft, Writing - review & editing. **Dorella Del Prete:** Writing - original draft. **Monica Ceol:** Writing - original draft. **Giovanna Priante:** Visualization, Writing - review & editing. **Lorenzo Arcangelo Calò:** Writing - review & editing. **Franca Anglani:** Writing - review & editing, Supervision.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

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Nowadays, we know that maintaining the right level of protein in the urine demands a coordinated effort in the kidney on the part of two main actors the glomerular filtration barrier (GFB) —a zip-structure built by podocytes, fenestrated endothelial cells, and the glomerular basement membrane (GBM), which prevent protein loss by acting as a size- and charge-exclusion filter (Kriz and Lemley, 2017) — and the proximal tubular cells (PTCs), which are implicated in protein re-uptake (Maunsbach, 1966).

In the past, proteinuria was considered a consequence of glomerular rather than tubular damage because it was believed that the GFB was impossible for proteins to cross. Micropuncture experiments in mice (Oken and Flamenbaum, 1971; Stolte et al., 1979; Tojo and Endou, 1992; Remuzzi et al., 2007) have demonstrated, however, that proteins can easily pass through the GFB, and that PTCs, which reabsorb more than 99% of them, have a more important role (Castrop and Schießl, 2017).

The main player in albumin and low-molecular-weight (LMW) protein reuptake by PTCs is the endocytic complex. It comprises two multiligand receptors, megalin (encoded by the *LRP2* gene) and the cubilin-amnionless complex (protein products of the *CUBN* and *AMN* genes, respectively), which are well-known partners of the chloride channel Cl^-/H^+ exchanger ClC-5 encoded by the *CLCN5* gene (Gianesello et al., 2016). Disruption of this macromolecular complex clearly plays a part in the onset of the proteinuria that points to the clinical diagnosis of a rare renal proximal tubulopathy known as Dent disease (Wrong et al., 1994).

In this review we discuss the *CLCN5* gene and its gene product C1C-5, whose mutations have been found responsible for Dent disease type 1. We also report on recent findings concerning the role of ClC-5 in renal glomerular cells.

2. The *CLCN5* gene

The human *CLCN5* gene (MIM#300008, reference sequence NG_007159.2) was identified 25 years ago (Scheinman et al., 1993; Fisher, 1994). It is located in the pericentromeric region on chromosome Xp11.23. Spanning about 170 Kb of genomic DNA, it has a coding region of 2,238 bp, and consists of 17 exons, including 11 coding exons (from 2 to 12) (Scheinman et al., 1993; Fisher, 1994; Fisher et al., 1995; Scheel et al., 2005). The *CLCN5* gene has 201 orthologues among jawed vertebrates (Gnathostomata), and 8 paralogues (*CLCN1, CLCN2, CLCN3, CLCN4, CLCN6, CLCN7, CLCNKA, CLCNKB*).

CLCN5 has five different transcripts, one of which does not encode for any protein (transcript variant 5, NM_001272102.2), while two (transcript variants 3 [NM_000084.5] and 4 [NM_001282163.1]) encode for the canonical 746 amino acid protein (Table 1), and the other two (transcript variants 1 [NM_001127899.3] and 2 [NM_001127898.3]) encode for the NH_2-terminal extended 816 amino acid protein (Ludwig et al., 2003).

The 5′ untranslated region (5′UTR) of *CLCN5* is complex and has yet to be fully clarified. It was predicted that the *CLCN5* gene would include two strong, and one weak functional promoters (Tosetto et al., 2014; Hayama et al., 2000). Several different 5′ alternately-used exons, some remaining untranslated, have been identified (Ludwig et al., 2003; Tosetto et

al., 2014; Hayama et al., 2000; Lloyd, 1996; Forino et al., 2004). In the human kidney, three promoters give rise, with a varying degree of efficiency, to 11 different mRNAs, with transcription initiating from at least three different start sites (Tosetto et al., 2014). When the significance of these different 5′UTR isoforms was recently investigated in control and proteinuric kidney biopsies, a different expression pattern emerged, suggesting a possible involvement for proteinuria in the ClC-5 transcriptional regulation (Ceol et al., 2020).

3. The ClC-5 protein

ClC-5 is the protein product of the *CLCN5* gene, and part of the family of ClC channels and ion exchangers (Jentsch et al., 2002). Studies on rat tissues demonstrated that ClC-5 is less widely distributed than ClC-3 or ClC-4, indicating a prominent expression in the epithelia of kidney and intestine in addition to other tissues such as brain and liver (Steinmeyer et al., 1995; Vandewalle, 2001).

3.1. Protein structure

The ClC-5 protein's structure is complex. The canonical isoform consists of 746 amino acids (Table 1), but there is also a second isoform comprising 816 amino acids of unknown biological significance (Ludwig et al., 2003). In 2003, the sequence alignment of the canonical human ClC-5 with its paralogues from *Escherichia coli* and *Salmonella enterica serovar typhimurium* led to the identification of the 18 α-helices (named A to R) that form its secondary structure (Wu, 2003) (Fig. 1). Helices B, H, I, O, P, and Q are the six main helices involved in the dimer interface's formation, and are necessary for proper pore configuration (Wu, 2003; Mansour-Hendili, 2015). In particular, helices B and I are on the side of the subunit, while helices O and P are located more centrally at the dimer's interface (Wu, 2003). The Cl^- selectivity filter is provided mainly by helices D, F, N, and R, which are brought together near the center of the channel (Wu, 2003; Mansour-Hendili, 2015; Grand et al., 2009; Pusch et al., 1997). The role of the E helix in ClC-5 function is still unclear, but mutations nearby reportedly produce a loss of Cl^- conductance, or current failures due to endoplasmic reticulum retention (Lloyd; 1996; Wu, 2003; Grand et al., 2009). Molecular modelling studies indicated that many *CLCN5* missense mutations cluster in the helices at the dimer interface of ClC-5, potentially destabilizing its formation and disrupting the pore configuration, with a loss or reduction of channel activity (Wu, 2003; Günther et al., 1998; Lourdel et al., 2012; Pusch and Zifarelli, 2015).

The most important amino acids for proper ClC-5 function are the "proton glutamate" Glu 268 (Zdebik, 2008; Grieschat and Alekov, Mar. 2012) and the "gating glutamate" Glu 211 (Dutzler et al., 2003; Yin et al., 2004). The proton glutamate is crucial to the H^+ transport acting as an H^+ transfer site (Zdebik, 2008; Accardi et al., 2005; Neagoe et al., 2010). Its importance was further highlighted by experiments using *Xenopus* oocytes, in which mutations mapping in the loop between nearby helices H and I were reportedly associated with chloride current abolition (Igarashi, Dec. 1998). The gating glutamate Glu 211 is necessary for both H^+ transport and ClC-5 voltage dependence (Scheel et al., 2005; Friedrich et al., 1999; Picollo and Pusch, 2005). Mutations that affect this amino acid lead to

a mutant protein that conducts both outward and inward Cl^- currents and acts as a Cl^- channel (Smith and Lippiat, 2010).

Like all eukaryotic ClCs, ClC-5 has a large cytoplasmic C-terminus containing two cystathionine beta-synthase (CBS) domains. Several authors have shown that CBS domains are involved in regulating the activity of ClCs, including ClC-5 (Meyer et al., 2007; Wellhauser et al., 2011; Zifarelli and Pusch, 2009) (Fig. 1). Missense mutations affecting these domains are pathogenic (Grand et al., 2009), leading to mutant proteins that target the plasma membrane and early endosomes correctly, but with an altered ClC-5 electrical activity (Lourdel et al., 2012). Studying nonsense mutations at the C-terminus, the Authors found that the resulting truncated proteins targeted the cell surface, but only one of the mutations studied had some residual activity (Grand et al., 2009), and another reportedly resulted in endoplasmic reticulum retention, underscoring the importance of the C-terminus for passing the protein quality control in this organelle (Grand et al., 2009). These findings suggest that truncated ClC-5 proteins at the C-terminus could induce loss of function, also due to a defective protein processing.

Finally, N-glycosylation is known to play a part in protein folding and oligomerization, and proteins that fail to fold or assemble properly undergo endoplasmic reticulum-associated degradation. It has been demonstrated that ClC-5 N-glycosylation site mutants may be poly-ubiquitinated, and conveyed to proteosomal degradation (Schmieder et al., 2007; D'Antonio et al., 2013).

3.2. Protein function

ClC-5 has a dual action, as it is involved in the acidification of early endosomes, and it participates in the protein uptake process at the brush border of PTCs (Günther et al.,1998; Devuyst et al., 1999).

ClC-5 was classified as one of the channels working mainly in intracellular membranes (with ClC-3, ClC-4, ClC-6 and ClC-7), rather than among those located largely on the plasma membrane (which include ClC-1, ClC-2, ClC-Ka, and ClC-Kb) (Jentsch et al., 2002).

All ClC channels need to dimerize to create the pore through which the ions pass (Wu, 2003; Mansour-Hendili, 2015; Lourdel et al., 2012), and *CLCN5* mutation studies have confirmed that a proper interaction between the two subunits is crucial to the proper functioning of ClC-5 as a dimer (Wu, 2003). It has also been demonstrated that ClC-5 can form both homo- and hetero-dimers due to its marked sequence homology with ClC-3 and ClC-4 (Jentsch et al., 1995; Okkenhaug et al., 2006; Mohammad-Panah, 2003).

Whether ClC-5 works as a Cl^- channel or as a Cl^-/H^+ exchanger was a debated question. ClC-0, the founding member of this protein family discovered in the electric organ of *Torpedo marmorata*, is a chloride channel, so all the CLC genes were initially expected to encode for anion channels (Zdebik, 2008). Three independent groups demonstrated, however, that - instead of being a simple Cl^- channel - ClC-5 functions as a Cl^-/H^+ antiporter when activated by positive voltages (Scheel et al., 2005; Zdebik, 2008; Picollo and

Pusch, 2005). The Cl^-/H^+ exchange process wastes more energy than recycling H^+ between endosomes and cytosol but leads to a more efficient lysosomal acidfication (Satoh, 2016).

A small portion of the ClC-5 channel is also located on the cell surface, where it is believed to mediate plasma membrane chloride currents (Günther et al., 1998; Friedrich et al., 1999; Devuyst et al., 1999; Luyckx, 1998), or participate in the macromolecular complexes at plasma membrane level that are responsible for LMW protein and albumin endocytosis (Hryciw et al., 2006).

3.3. ClC-5 protein partners

Receptor-mediated, clathrin-dependent endocytosis, and the subsequent metabolization of proteins and nutrients are considered as some of the most important functions of renal PTCs. Receptor-mediated endocytosis requires the coordinated functioning of numerous proteins and signal transduction molecules. In particular, megalin and cubilin have a core role in this process (Kerjaschki and Farquhar, 1982; Sahali et al., 1988). These receptors bind a variety of filtered ligands with varying affinities, fulfilling their task via receptor-mediated endocytosis, and mediating the delivery of their ligands to the lysosomes of the proximal tubule (Christensen, 1982; Christensen et al., 2009). The apical expression of both megalin and cubilin in PTCs is finely tuned by a recycling mechanism that involves the endosomal pathway (reviewed in (Christensen and Birn, 2002). Cubilin also depends on the single transmembrane protein amnionless for its proper anchorage to the plasma membrane (Fyfe, 2004). A downregulation of both megalin and cubilin was demonstrated in both murine (Christensen, 2003; Gabriel, 2017) and human PTCs (Santo, 2004; Tanuma et al., 2007; Gianesello, 2020) carrying a defective ClC-5, supporting the importance of ClC-5 in the proper localization and function of these two receptors, and in the LMW protein reuptake process. The Devuyst group reported a rescue in megalin and ClC-5 expression in PTCs of *Clcn5* knock-out mice after bone marrow transplantation with wild-type cells, supporting the role of ClC-5 in megalin localization and expression (Gabriel, 2017). The Na^+-H^+ exchanger regulatory factor 2 (NHERF2) and the Na^+-H^+ exchanger isoform 3 (NHE3) were also shown to take part in albumin endocytosis (Hryciw, 2006; Slattery, 2011). In rat PTCs, ClC-5 was found to interact with megalin via NHERF2 (Hryciw et al., 2012), but whether a direct protein-protein interaction exists between ClC-5 and NHE3 is still not clear, even if NHE3 was found dysregulated in the two ClC-5 knock-out mouse models (Wang, 2000; Piwon et al., 2000; Silva et al., 2003; Günther et al., 2003) (Fig. 2A).

Albumin endocytosis is known to proceed via clathrin-coated vesicles. This process needs a proper cytoskeletal structure, with some remodeling of the actin web to facilitate plasma membrane budding, and this gives rise to empty areas around the clathrin-coated pits (Fujimoto et al., 2000). In eukaryotic cells, the actin-depolymerizing factor/cofilin proteins regulate actin filament turnover and tertiary net formation (as reviewed in (Bamburg, 1999). Two-hybrid system experiments showed that the C-terminal end of ClC-5 can interact with cofilin. This interaction is needed for actin depolymerization and proper albumin endocytosis in two different PTC lines (Opossum -OK- and porcine -LLC-PK1), and it persists throughout the endocytotic cycle (Hryciw et al., 2003) (Fig. 2A).

KIF1B is also thought to facilitate the microtubular transport of endocytic vesicles away from the plasma membrane. It has already been demonstrated that an interaction between KIF1B and ClC-5 is needed for proper ClC-5 expression on the plasma membrane (see below) (Reed et al., 2010). All these data highlight the importance of an intact cytoskeletal structure for proper endocytosis.

An increasing acidification must also take place along the endosomal pathway for endocytosis to proceed (Clague et al., 1994). In PTCs, endosomal acidification is driven by the electrogenic vacuolar H^+-ATPase (V-ATPase) (Herak-Kramberger et al., 1998). The V-ATPase lacks any coupling to counter-ion transport, and this gives rise to its electrogenic action. In other words, acidification is accompanied by the generation of a positive potential of the inner endosomal membrane (Mellman et al., 1986). This positive potential needs to be counterbalanced by the influx of anions (negative charges) from the extracellular space, or by the efflux of cations (positive charges) from inside the vesicle in order to maintain H^+ translocation into the vesicle lumen (Mellman et al., 1986). Whether the endosomal acidification needs the collaboration between ClC-5 and the V-ATPase is still debated. Two independent groups demonstrated the ability of ClC-5 to drive endosomal acidification independently of the V-ATPase (Scheel et al., 2005; Smith and Lippiat, 2010). On the other hand, several Authors endorse the hypothesis that ClC-5 provides a shunt conductance in early endosomes, enabling an efficient intraluminal acidification by the V-ATPase (Günther et al., 1998; Devuyst et al., 1999; Luyckx, 1998; Hara- Chikuma et al., 2005). Co-localization experiments demonstrated that ClC-5 expression largely overlaps with that of V-ATPase, supporting a close collaboration between the two proteins in this ion balancing act, thanks to the influx of Cl^- induced by ClC-5 (Günther et al., 1998) (Fig. 2B). Such a cooperation was further supported in a recent paper by Satoh *et al.*, who found a reduced action of V-ATPase in a patient with Dent disease (Satoh, 2016).

3.4. Renal expression of ClC-5

In the human kidney, ClC-5 is consistently expressed not only in PTCs but also in collecting duct α-intercalated cells, and in epithelial cells of the thick ascending limb (TAL) of Henle's loop (Devuyst et al., 1999). This is a wider expression than in rat and mouse kidneys, where ClC-5 is expressed in PTCs, and in collecting duct α- and β-intercalated cells (Günther et al., 1998; Obermüller et al., 1998; Sakamoto, 1999). In PTCs of all these species, ClC-5 is predominantly located in intracellular subapical endosomes, and in the brush border membrane (Günther et al., 1998; Devuyst et al.,1999; Sakamoto, 1999) (Fig. 3).

The prominent effects of ClC-5 in PTCs have overshadowed those on collecting duct and TAL even if ClC-5 expression in these segments is well-described (Günther et al., 1998; Devuyst et al., 1999; Luyckx, 1998; Sakamoto, 1999). In the collecting duct, ClC-5 is localized to the α-intercalated cells that are crucial for acid-base homeostasis, and it colocalizes with the V-ATPase in the apical and subapical vesicles. Accordingly, ClC-5 might be important for insertion and recycling of these vesicles, and when ClC-5 function is lost, defective expression of V-ATPase may result causing impaired urinary acidification (Günther et al., 1998). The role of ClC-5 in TAL remains circumstantial and speculative (Pham, 2004). Studies suggest that endocytosis takes place in murine TAL where V-ATPase

is also present albeit at a lesser extent than in PTCs, thus suggesting a possible role for ClC-5 during endocytosis and exocytosis processes in this nephron segment (Bastani, 1997).

ClC-5 proximal tubular apical expression is fine-tuned by several proteins. In 2001, the Thakker group identified the interaction between the WW domain-containing protein 2 (WWP2) and a proline-rich stretch of amino acids in the ClC-5C-terminus. They also showed that WWP2 is needed to modulate ClC-5 retention in the plasma membrane (Schwake et al., 2001). In 2004, the Poronnik group added another piece to the puzzle, demonstrating the functional interaction of Nedd4–2 with ClC-5 (Hryciw, 2004) in Opossum kidney (OK) cells. WWP2 and the Nedd4-like proteins belong to the same ubiquitin-protein ligase family (Harvey and Kumar, 1999), which regulates membrane expression levels of target proteins. They remove specific proteins from the plasma membrane via the ubiquitination pathway, leading to the target protein's degradation into proteasomes (Schwake et al., 2001). Unfortunately, these findings were not confirmed in PTCs from *Clcn5*$^{-/-}$ mice (Rickheit, 2010). To complicate things further, the Thakker group demonstrated *in vivo* and *in vitro*, in both polarized and non-polarized OK cells, that the interaction between ClC-5 and KIF1B via the ClC-5 C-terminus is important for the plasma expression of ClC-5 as well (Reed et al., 2010). Wojciechowski *et al.* also recently reported that barttin plays a part in the trafficking and processing of ClC-5 too (Wojciechowski, 2018). These Authors made the point, however, that the interaction between barttin and ClC-5 probably occurs in the tubular segments where barttin is expressed, such as the thick ascending limb of Henle's loop and the intercalated cells of the collecting duct (Wojciechowski, 2018).

ClC-5 is not only expressed by tubular cells in the kidney. We recently found it expressed in human glomeruli as well, identifying these ClC-5-positive cells as podocytes. In particular, ClC-5 was found overexpressed in the glomeruli of patients with proteinuric nephropathies, at both mRNA and protein level, suggesting a role for this protein in albumin endocytosis by podocytes (Ceol, 2012). Prabakaran *et al.* demonstrated human podocyte positivity for both megalin and cubilin too, supporting the likelihood of a functional role for ClC-5 at glomerular level (Prabakaran, 2011; Prabakaran et al., 2012) (Fig. 3). Further *in vitro* studies on human podocytes confirmed the presence of ClC-5 (Gianesello, 2017; Solanki, 2018), and showed that protein overload can modulate the protein uptake macromolecular complex, inducing an increase in the gene expression of *CLCN5*, *CUBN*, and *AMN*, and in the protein expression of ClC-5 (Gianesello, 2017).

Podocytes are not the only type of glomerular cell expressing the protein uptake complex. We recently showed that human parietal epithelial cells (PECs) also express ClC-5, megalin and cubilin (Gianesello, et al., 2018) (Fig. 3). This evidence confirmed previous findings in kidneys of diabetic rats (Zhao et al., 2019). Zhao et al. also demonstrated that album intake by rat PECs via megalin led to an increase in CD44 (Zhao et al., 2019). The cluster of differentiation 44 (CD44) is a glycoprotein localized on the cell surface that plays a role in several cellular processes, such as cell differentiation, cell migration, cell-matrix binding, leukocyte trafficking, and scar formation (Nakamura et al., 2005). CD44 is now used to identify PECs directed towards a profibrotic phenotype and ultimately leading to glomerulosclerosis and kidney failure (Lim et al., 2016). Dent disease patients are now

known to have focal segmental glomerulosclerosis (FSGS) and/or focal global glomerulosclerosis (Wang, 2016), so the expression of ClC-5 and its protein partners in PECs points to an active involvement of these cells in the onset of the glomerular damage.

4. *CLCN5* and Dent disease 1

The term Dent's disease was first introduced in the 1990s (Wrong et al., Aug. 1994). It identifies a group of X-linked recessive renal disorders characterized by LMW proteinuria and variable degrees of hypercalciuria, nephrocalcinosis and/or nephrolithiasis (Lloyd, 1996; Thakker, 2000; Dent and Friedman, 1964). Mutations in the *CLCN5* gene are the most common cause of the disease (responsible for about 65% of cases), which is now called Dent disease type 1 (DD1; MIM#300009) (Jentsch et al., 1995; Thakker, 1997; Waldegger and Jentsch, 2000). Dent disease type 2 (DD2; MIM#300555) is due to mutations in the *OCRL* gene (Hoopes et al., 2005), identified in about 10–15% of Dent disease patients (Lieske et al., 1993). The remaining 25–35% of patients have neither of these mutations, and were classified as cases of Dent disease type 3 (DD3) (Anglani, 2015).

Several studies tried to identify other genes responsible for the phenotype observed in DD3 patients. In 2009, the Thakker and Anglani groups screened Dent disease patients for mutations in the genes encoding for cofilin and collectrin, but none came to light (Wu, et al., 2009; Tosetto, Oct. 2009). After technological advances in next-generation sequencing, a couple of studies tried to fill the gap between the DD3 phenotype and its genetic origin, but to no avail (Gianesello, 2020; Zhang et al., 2017; Anglani, 2018). Collectively, these studies give the impression that DD3 patients' phenotypes might be explained by a number of hypotheses: (i) they could be the result of defects in more than one gene; (ii) they may be atypical phenotypes of known hereditary nephropathies; or (iii) they may be blended phenotypes.

4.1. Allelic heterogeneity

Allelic heterogeneity is a characteristic of DD1. To date, 266 different *CLCN5* pathogenic variants have been described (Table S1) (Mansour-Hendili, 2015; Gianesello, 2020; Bitsori et al., 2019; Tang, 2016; Li, 2016; Wong et al., 2017; Guven, 2017; Günthner et al., 2018; Sancakli et al., 2018; Bignon, 2018; Wen et al., 2018; Matsumoto, 2018; Ye et al., 2019; Hoopes et al., 2004; Bhardwaj et al., 2016; Armanet et al., 2015; Danyel et al., 2019; Zaniew, 2017; Tosetto, 2009; Güngör, 2020). A few pathogenic variants were found in more than one family (Lieske et al., 1993): 48% were truncating (nonsense, frameshift or complex); 37% were non-truncating (missense or in-frame insertions/deletions); 10% were splice site mutations; and 5% were other types (large deletions, Alu insertions or 5′UTR mutations). In particular, the majority of the reported mutations were missense (with 93 different mutations) or frameshift (83), followed by nonsense mutations (44), splicing mutations (26), and large deletions (10) (Fig. 4).

The spectrum of pathogenic *CLCN5* variants shows great variety, and de novo pathogenic variants are common. They are scattered throughout the gene's coding sequence, generating truncated or absent ClC-5 protein in approximately 60% of cases (Lieske et al., 1993). Most pathogenic *CLCN5* variants have not been fully investigate in functional terms. DD1

phenotypic variability may be due to different degrees of the altered ClC-5 protein's function loss. Functional investigations on wild-type and mutant ClC-5 in *Xenopus* oocytes (Lloyd, 1996; Lloyd, 1997), in human embryonic kidney cells (HEK) (Smith and Lippiat, 2010; Grand et al., 2011), in conditionally immortalized proximal-tubular epithelial cell lines (ciPTEC) from patients with DD1 (Gorvin et al., 2013), and *in vivo* in mice harboring induced ClC-5 mutations (Novarino et al., 2010) have enabled *CLCN5* variants to be classified according to their functional consequences as follows (Lourdel et al., 2012; D'Antonio et al., 2013; Lieske et al., 1993; Smith et al., 2009; Ludwig et al., 2005).

Class 1: pathogenic variants that impair processing and folding, leading to the mutated protein's retention in the endoplasmic reticulum, and its degradation by proteosomes. This results in the mutant proteins being improperly N-glycosylated, and non-functioning due to a defective trafficking to the cell surface and to the early endosomes;

Class 2: pathogenic variants that generate a functionally defective protein, devoid of electric currents, and resulting in the failure of endosomal acidification;

Class 3: pathogenic variants that cause an abnormal subcellular localization of the mature protein;

Class 4: pathogenic variants generating a protein that normally targets the plasma membrane correctly, but exhibits reduced membrane currents.

The above evidences indicate that the mechanisms by which ClC-5 mutations lead to DD1 vary and are not necessarily linked to a primary defect in the endosomal acidification.

4.2. Clinical description

Since DD1 is an X-linked recessive disorder, males are more frequently affected than females. Females carriers are usually asymptomatic (Lieske et al., 1993). The clinical signs in patients with DD1 are the consequence of an involvement of both the tubular and the glomerular compartments. These signs generally appear in childhood or early adulthood. The signs of DD1 can vary, but always include proteinuria (Anglani, 2015). DD1 patients may also reveal a significantly reduced cubilin and megalin expression in PTCs, probably due to a disturbed intracellular trafficking of the receptors (Gianesello, 2020; Christensen et al., 2013); this is probably one of the reasons behind the onset of proteinuria.

Proximal renal tubular dysfunction is characterized by LMW proteinuria (LMWP), hypercalciuria, nephrocalcinosis and/or nephrolithiasis. Additional tubular defects, such as hyperphosphaturia, glycosuria and aminoaciduria, may sometimes appear too, but are less prominent features (Devuyst and Thakker, 2010; Hodgin et al., 2008). Males under ten years old may manifest only LMWP and/or hypercalciuria. Hypophosphatemia and bone disorders (rickets or osteomalacia) have also been reported, as well as growth restriction and short stature (Bökenkamp, 2009), usually without a complete Fanconi syndrome or metabolic acidosis (Reinhart et al., 1995) (Fig. 5).

This huge variability in the clinical phenotype of DD1 was further confirmed in two murine *Clcn5* knock-out models (Wang, 2000; Piwon et al., 2000; Silva et al., 2003; Günther et al.,

Jan. 2003): both models showed LMWP, polyuria, phosphaturia, and normal serum PTH; the Guggino model also revealed hypercalciuria, nephrocalcinosis, and an increase in serum 1,25(OH)2D (Anglani et al., Nov. 2019).

Dent disease progresses to chronic kidney disease (CKD) between the 3rd and 5th decades of life in 30–80% of male patients. In adult cases, the disease is usually suspected in the presence of nephrocalcinosis/nephrolithiasis or unexplained CKD in individuals with hypercalciuria, or in the context of familial cases of CKD and/or nephrolithiasis (Edvardsson et al., 2013; Ferraro et al., 2013).

In several case reports, DD1 patients had not only LMWP, but also severe proteinuria (even in the nephrotic range), suggesting the possibility of glomerular as well as tubular damage. This hypothesis was confirmed by histological findings of focal global and/or focal segmental glomerulosclerosis or mesangial proliferative glomerulonephritis, in addition to episodic evidences of isolated proximal tubular dysfunction (Wang, 2016; Zaniew, 2017; Copelovitch et al., 2007; Frishberg et al., 2009; Kaneko et al., 2010; Cramer et al., 2014; Bao et al., 2019; Fervenza, 2013). These histological signs suggest that there could be misdiagnosed cases of DD1 among patients diagnosed with FSGS. The recent study by Beara-Lasic *et al.* on pediatric FSGS and CKD cohorts found no *CLCN5* mutations in these children, however (Beara-Lasic eet al., 2019).

4.3. Atypical phenotypes

DD1 is rarely associated with extrarenal symptoms, which are more common in DD2 due its overlapping features with Lowe syndrome that is also caused by mutations in the *OCRL* gene (De Matteis et al., 2017). In fact, a case of DD1 associated with ocular anomalies, bone disease, and mild mental impairment was described as being caused by the coinheritance of mutations in both the *CLCN5* and the *OCRL* genes (Addis et al., 2013). DD1 is considered an exclusively renal disease, given that ClC-5 is expressed mainly in the kidney. Some reported cases of DD1 were associated with unusual symptoms, such as: night blindness (Sethi et al., 2009; Becker-Cohen et al., 2012); growth hormone deficiency (Samardzic et al., 2011; Sheffer-Babila et al., 2008); and, in one instance, growth hormone deficiency complicated by Budd-Chiari syndrome (Platt et al., 2014).

A blended phenotype should be suspected for DD1 associated with a Bartter-like phenotype. Bartter's syndrome (BS; MIM #s: type I, 601678; type II, 241200; type III, 607364; type IV, 602522; type IVb, 613090) and Gitelman's syndrome (GS; MIM # 263800) are autosomal recessive tubular disorders with characteristic sets of metabolic abnormalities, including: hypokalemia; metabolic alkalosis; hyperreninemia; hyperplasia of the juxtaglomerular apparatus; and hyperaldosteronism. They are caused by mutations in genes encoding proteins involved in renal electrolyte homeostasis in the thick ascending limbs of Henle's loop, and in the distal convolute tubule (Blanchard et al., 2017). DD1 and BS may be clinically diagnosed in the same patient, albeit rarely (Besbas et al., 2005; Bogdanović et al., 2010; Okamoto et al., 2012). It is currently hard to say whether this blended phenotype is the outcome of mutations in both the *CLCN5* and the BS or GS genes, or rather an expansion of the DD1 phenotype because of the role of ClC-5 in the other tubular segments of the nephron, which however has yet to be clarified. In fact, if on one hand not all of the BS and

GS genes were screened in these patients, on the other it was very recently reported that barttin, the protein impaired in Bartter's syndrome type 4 (which is an essential subunit of the ClC-Ka and ClC-Kb chloride channels), appears to regulate the subcellular localization and posttranslational modification of ClC-5 (Wojciechowski, 2018). The Authors hypothesized that abnormal barttin-ClC-5 interactions in the thick ascending limbs of Henle's loop and the collecting duct might contribute to the Bartter's phenotype seen in some DD1 patients.

4.4. Clinical management

There is no specific therapy for patients with DD1, so it is usual clinical practice to treat patients symptomatically. Pharmacological intervention generally aims to reduce proteinuria, hypercalciuria or rickets, or may be to prevent nephrolithiasis or nephrocalcinosis. Many of the therapeutic options available have not been tested in randomized clinical trials.

Thiazide diuretics have proved effective against hypercalciuria in DD1 patients (Raja et al., 2002), but the Authors recommended using hydrochlorothiazide only in those who were recurrent stone formers. This was the starting point for the first clinical trial on DD1 patients, conducted in France from 2003 to 2008 (ClinicalTrials.gov Identifier: NCT00638482), to examine whether high doses of thiazides could prevent nephrocalcinosis in children with Dent disease. The Authors found the treatment associated with a lower calcium excretion, but also with significant adverse events (which prompted the trial to be stopped early), so they recommended caution in the use of diuretics in pediatric patients with Dent disease (Lieske et al., 1993; Blanchard et al., 2008).

The presence of proteinuria usually induces physicians to treat DD1 patients with ACE and/or angiotensin II receptor blockers. Theoretically, drugs inhibiting the renin-angiotensin-aldosterone system (RAAS) should not be effective for tubular proteinuria, but the glomerular damage present in some DD1 patients could explain the positive effects of this treatment (Zaniew, 2017; Blanchard et al., 2016). Only a small percentage of DD1 patients have glomerular damage, however, and this would explain the discrepant results obtained with RAAS inhibition in such patients (van Berkel et al., 2017).

Studies on *Clcn5*$^{-/-}$ mice revealed that a high-citrate diet could reduce hypercalciuria and slow renal disease progression, even in the absence of nephrolithiasis (Cebotaru et al., 2005), but this has yet to be confirmed in humans. Nonetheless, DD1 patients are often prescribed citrates (usually potassium citrate) to prevent nephrolithiasis, improve acidosis, and thus slow CKD progression (Lieske et al., 1993; Zaniew, 2017).

Bone disease was found to respond to vitamin D supplementation in patients with high serum alkaline phosphatase levels (Wrong et al., 1994). Treatment with vitamin D requires close monitoring, however, because it can exacerbate hypercalciuria, and may increase the risk of nephrocalcinosis. That is why some authors recommend monitoring calcemia, calciuria and 25-hydroxyvitamin D to prevent this from happening (Blanchard et al., 2016).

Finally, phosphate supplementation could be used to treat hypophosphatemia or bone disease (Lieske et al., 1993; Zaniew, 2017). The most recent clinical trial focusing on this

issue was carried out at the Mayo Clinic in Minnesota from 2014 to 2019 (ClinicalTrials.gov Identifier: NCT02016235), the aim being to establish whether supplementation with phosphorus could reduce hypercalciuria. The results of this study have yet to be published.

5. Conclusions

In conclusion, proteinuria is an important clinical sign that should not be underestimated. It is well-known that the ClC-5 channel, encoded by the *CLCN5* gene, has a pivotal role in protein re-uptake by PTCs. Studies on Dent disease patients have expanded the picture, disclosing how widely the CIC-5 channel is expressed throughout the kidney, and how many processes are affected by its malfunctioning. Taken together, these data demonstrate that Dent disease type 1 should now be considered as a disorder affecting not only PTCs, but the whole kidney.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Abbreviations:

5′UTR	5′ untranslated region
BS	Bartter's syndrome
CKD	chronic kidney disease
ciPTEC	conditionally immortalized proximal-tubular epithelial cell lines
CSB	cystathionine beta-synthase
DD1	Dent disease type 1
DD2	Dent disease type 2
DD3	Dent disease type 3
FSGS	focal segmental glomerulosclerosis
GBM	glomerular basement membrane
GFB	glomerular filtration barrier

GS	Gitelman's syndrome
HEK	human embryonic kidney cells
LMW	low-molecular-weight
LMWP	low-molecular-weight proteinuria
NHERF2	Na^+-H^+ exchanger regulatory factor 2
NHE3	Na^+-H^+ exchanger isoform 3
OK	Opossum kidney
PEC	parietal epithelial cell
PTCs	proximal tubular cells
RAAS	renin-angiotensin-aldosterone system
TAL	thick ascending limb
V-ATPase	electrogenic vacuolar H^+-ATPase
WWP2	WW domain-containing protein 2

References

Accardi A, Walden M, Nguitragool W, Jayaram H, Williams C, Miller C, 2005. Separate ion pathways in a Cl−/H+ exchanger. J. Gen. Physiol 126 (6), 563–570. 10.1085/jgp.200509417. [PubMed: 16316975]

Addis M, et al., 6. 2013. An atypical Dent's disease phenotype caused by co-inheritance of mutations at CLCN5 and OCRL genes. Eur. J. Hum. Genet. EJHG 21 (6), 687–690. 10.1038/ejhg.2012.225. [PubMed: 23047739]

Anglani F, et al., 2015. Nephrolithiasis, kidney failure and bone disorders in Dent disease patients with and without CLCN5 mutations. SpringerPlus 4, 492. 10.1186/s40064-015-1294-y. [PubMed: 26389017]

Anglani F, et al., 2018. Hypercalciuria and nephrolithiasis: Expanding the renal phenotype of Donnai-Barrow syndrome. Clin. Genet 94 (1), 187–188. 10.1111/cge.13242. [PubMed: 29532936]

Anglani F, Gianesello L, Beara-Lasic L, Lieske J, 2019. Dent disease: A window into calcium and phosphate transport. J. Cell. Mol. Med 23 (11), 7132–7142. 10.1111/jcmm.14590. [PubMed: 31472005]

Armanet N, et al., 2015. Double Xp11.22 deletion including SHROOM4 and CLCN5 associated with severe psychomotor retardation and Dent disease. Mol. Cytogenet 8, 8. 10.1186/s13039-015-0107-x. [PubMed: 25670966]

Bamburg JR, 1999. Proteins of the ADF/cofilin family: essential regulators of actin dynamics. Annu. Rev. Cell Dev. Biol 15, 185–230. 10.1146/annurev.cellbio.15.1.185. [PubMed: 10611961]

Bao Y, et al., 2019. Clinical and genetic analysis of Dent disease with nephrotic range albuminuria in Shaanxi, China. Sci. China Life Sci 62 (12), 1590–1593. 10.1007/s11427-018-9829-0. [PubMed: 31701402]

Bastani B, 1997. Immunocytochemical localization of the vacuolar H(+)-ATPase pump in the kidney. Histol. Histopathol 12 (3), 769–779. [PubMed: 9225160]

Beara-Lasic L, et al., 2019. Prevalence of low molecular weight proteinuria and Dent disease 1 CLCN5 mutations in proteinuric cohorts. Pediatr. Nephrol. Berl. Ger 10.1007/s00467-019-04210-0.

Becker-Cohen R, Rinat C, Ben-Shalom E, Feinstein S, Ivgi H, Frishberg Y, 2012. Vitamin A deficiency associated with urinary retinol binding protein wasting in Dent's disease. Pediatr. Nephrol. Berl. Ger 27 (7), 1097–1102. 10.1007/s00467-012-2121-0.

Besbas N, Ozaltin F, Jeck N, Seyberth H, Ludwig M, 2005. CLCN5 mutation (R347X) associated with hypokalaemic metabolic alkalosis in a Turkish child: an unusual presentation ofDent's disease. Nephrol. Dial. Transplant. Off. Publ. Eur. Dial. Transpl. Assoc. - Eur. Ren. Assoc 20 (7), 1476–1479. 10.1093/ndt/gfh799.

Bhardwaj S, Thergaonkar R, Sinha A, Hari P, Hi C, Bagga A, 2016. Phenotype of dent disease in a cohort of Indian children. Indian Pediatr. 53 (11), 977–982. 10.1007/s13312-016-0971-4. [PubMed: 27889724]

Bignon Y, et al., 2018. A novel CLCN5 pathogenic mutation supports Dent disease with normal endosomal acidification. Hum. Mutat 39 (8), 1139–1149. 10.1002/humu.23556. [PubMed: 29791050]

Bitsori M, Vergadi E, Galanakis E, 2019. A Novel CLCN5 Splice Site Mutation in a Boy with Incomplete Phenotype of Dent Disease. J. Pediatr. Genet 8 (4), 235–239. 10.1055/s-0039-1692172. [PubMed: 31687264]

Blanchard A, et al., 2008. Effect of hydrochlorothiazide on urinary calcium excretion in dent disease: an uncontrolled trial. Am. J. Kidney Dis. Off. J. Natl. Kidney Found 52 (6), 1084–1095. 10.1053/j.ajkd.2008.08.021.

Blanchard A, et al., 2016. Observations of a large Dent disease cohort. Kidney Int. 90 (2), 430–439. 10.1016/j.kint.2016.04.022. [PubMed: 27342959]

Blanchard A, et al., 2017. Gitelman syndrome: consensus and guidance from a Kidney Disease: Improving Global Outcomes (KDIGO) Controversies Conference. Kidney Int. 91 (1), 24–33. 10.1016/j.kint.2016.09.046. [PubMed: 28003083]

Bogdanović R, Draaken M, Toromanović A, Dordević M, Stajić N, Ludwig M, 2010. A novel CLCN5 mutation in a boy with Bartter-like syndrome and partial growth hormone deficiency. Pediatr. Nephrol. Berl. Ger 25 (11), 2363–2368. 10.1007/s00467-010-1615-x.

Bökenkamp A, et al., 2009. Dent-2 Disease: A Mild Variant of Lowe Syndrome. J. Pediatr 155 (1), 94–99. 10.1016/j.jpeds.2009.01.049. [PubMed: 19559295]

Castrop H, Schießl IM, 2017. Novel routes of albumin passage across the glomerular filtration barrier. Acta Physiol 219 (3), 546–555. 10.1111/apha.12760.

Cebotaru V, et al., 2005. High citrate diet delays progression of renal insufficiency in the ClC-5 knockout mouse model of Dent's disease. Kidney Int. 68 (2), 642–652. 10.1111/j.1523-1755.2005.00442.x. [PubMed: 16014041]

Ceol M, et al., 2012. Involvement of the tubular ClC-type exchanger ClC-5 in glomeruli of human proteinuric nephropathies. PloS One 7 (9), e45605. 10.1371/journal.pone.0045605. [PubMed: 23029130]

Ceol M, Gianesello L, Tosetto E, Priante G, Del Prete D, Anglani F, 2020. CLCN5 5'UTR isoforms in human kidneys: differential expression analysis between controls and patients with glomerulonephritis. J. Investig. Med. Off. Publ. Am. Fed. Clin. Res 10.1136/jim-2019-001205.

Christensen EI, 1982. Rapid membrane recycling in renal proximal tubule cells. Eur. J. Cell Biol 29 (1), 43–49. [PubMed: 7151825]

Christensen EI, et al., 2003. Loss of chloride channel ClC-5 impairs endocytosis by defective trafficking of megalin and cubilin in kidney proximal tubules. Proc. Natl. Acad. Sci. U. S. A 100 (14), 8472–8477. 10.1073/pnas.1432873100. [PubMed: 12815097]

Christensen EI, Birn H, 2002. Megalin and cubilin: multifunctional endocytic receptors. Nat. Rev. Mol. Cell Biol 3 (4), 256–266. 10.1038/nrm778. [PubMed: 11994745]

Christensen EI, Verroust PJ, Nielsen R, 2009. Receptor-mediated endocytosis in renal proximal tubule. Pflugers Arch. 458 (6), 1039–1048. 10.1007/s00424-009-0685-8. [PubMed: 19499243]

Christensen EI, Nielsen R, Birn H, 2013. From bowel to kidneys: the role of cubilin in physiology and disease. Nephrol. Dial. Transplant. Off. Publ. Eur. Dial. Transpl. Assoc. - Eur. Ren. Assoc 28 (2), 274–281. 10.1093/ndt/gfs565.

Clague MJ, Urbé S, Aniento F, Gruenberg J, 1994. Vacuolar ATPase activity is required for endosomal carrier vesicle formation. J. Biol. Chem 269 (1), 21–24. [PubMed: 8276796]

Copelovitch L, Nash MA, Kaplan BS, 2007. Hypothesis: Dent disease is an under-recognized cause of focal glomerulosclerosis. Clin. J. Am. Soc. Nephrol. CJASN 2 (5), 914–918. 10.2215/CJN.00900207.

Cramer MT, Charlton JR, Fogo AB, Fathallah-Shaykh SA, Askenazi DJ, Guay-Woodford LM, 2014. Expanding the phenotype of proteinuria in Dent disease. A case series. Pediatr. Nephrol. Berl. Ger 29 (10), 2051–2054. 10.1007/s00467-014-2824-5.

D'Antonio C, Molinski S, Ahmadi S, Huan L-J, Wellhauser L, Bear CE, 2013. Conformational defects underlie proteasomal degradation of Dent's disease-causing mutants of ClC-5. Biochem. J 452 (3), 391–400. 10.1042/BJ20121848. [PubMed: 23566014]

Danyel M, Suk EK, Raile V, Gellermann J, Knaus A, and Horn D, "Familial Xp11.22 microdeletion including SHROOM4 and CLCN5 is associated with intellectual disability, short stature, microcephaly and Dent disease: a case report," BMC Med. Genomics, vol. 12, no. 1, p. 6, 10 2019, doi: 10.1186/s12920-018-0471-6. [PubMed: 30630535]

De Matteis MA, Staiano L, Emma F, Devuyst O, 2017. The 5-phosphatase OCRL in Lowe syndrome and Dent disease 2. Nat. Rev. Nephrol 13 (8), 455–470. 10.1038/nrneph.2017.83. [PubMed: 28669993]

Dent CE, Friedman M, 1964. HYPERCALCURIC RICKETS ASSOCIATED WITH RENAL TUBULAR DAMAGE. Arch. Dis. Child 39, 240–249. 10.1136/adc.39.205.240. [PubMed: 14169453]

Devuyst O, Christie PT, Courtoy PJ, Beauwens R, Thakker RV, 1999. Intra-renal and subcellular distribution of the human chloride channel, CLC-5, reveals a pathophysiological basis for Dent's disease. Hum. Mol. Genet 8 (2), 247–257. 10.1093/hmg/8.2.247. [PubMed: 9931332]

Devuyst O, Thakker RV, 2010. Dent's disease. Orphanet J. Rare Dis 5, 28. 10.1186/1750-1172-5-28. [PubMed: 20946626]

Dutzler R, Campbell EB, MacKinnon R, 2003. Gating the selectivity filter in ClC chloride channels. Science 300 (5616), 108–112. 10.1126/science.1082708. [PubMed: 12649487]

Edvardsson VO, et al., 2013. Hereditary causes of kidney stones and chronic kidney disease. Pediatr. Nephrol 28 (10), 1923–1942. 10.1007/s00467-012-2329-z. [PubMed: 23334384]

Ferraro PM, D'Addessi A, Gambaro G, 2013. When to suspect a genetic disorder in a patient with renal stones, and why. Nephrol. Dial. Transplant. Off. Publ. Eur. Dial. Transpl. Assoc. - Eur. Ren. Assoc 28 (4), 811–820. 10.1093/ndt/gfs545.

Fervenza FC, 2013. A patient with nephrotic-range proteinuria and focal global glomerulosclerosis. Clin. J. Am. Soc. Nephrol. CJASN 8 (11), 1979–1987. 10.2215/CJN.03400313.

Fisher SE, et al., 1994. Isolation and partial characterization of a chloride channel gene which is expressed in kidney and is a candidate for Dent's disease (an X-linked hereditary nephrolithiasis). Hum. Mol. Genet 3 (11), 2053–2059. [PubMed: 7874126]

Fisher SE, Vanbakel I, Lloyd SE, Pearce SHS, Thakker RV, Craig IW, 1995. Cloning and Characterization of CLCN5, the Human Kidney Chloride Channel Gene Implicated in Dent Disease (an X-Linked Hereditary Nephrolithiasis). Genomics 29 (3), 598–606. 10.1006/geno.1995.9960. [PubMed: 8575751]

Forino M, Graziotto R, Tosetto E, Gambaro G, D'Angelo A, Anglani F, 2004. Identification of a novel splice site mutation of CLCN5 gene and characterization of a new alternative 5' UTR end of ClC-5 mRNA in human renal tissue and leukocytes. J. Hum. Genet 49 (1), 53–60. 10.1007/s10038-003-0108-1. [PubMed: 14673707]

Friedrich T, Breiderhoff T, Jentsch TJ, 1999. Mutational analysis demonstrates that ClC-4 and ClC-5 directly mediate plasma membrane currents. J. Biol. Chem 274 (2), 896–902. 10.1074/jbc.274.2.896. [PubMed: 9873029]

Frishberg Y, et al., 2009. Dent's disease manifesting as focal glomerulosclerosis: Is it the tip of the iceberg? Pediatr. Nephrol. Berl. Ger 24 (12), 2369–2373. 10.1007/s00467-009-1299-2.

Fujimoto LM, Roth R, Heuser JE, Schmid SL, 2000. Actin assembly plays a variable, but not obligatory role in receptor-mediated endocytosis in mammalian cells. Traffic Cph. Den 1 (2), 161–171. 10.1034/j.1600-0854.2000.010208.x.

Fyfe JC, et al., 2004. The functional cobalamin (vitamin B12)-intrinsic factor receptor is a novel complex of cubilin and amnionless. Blood 103 (5), 1573–1579. 10.1182/blood-2003-08-2852. [PubMed: 14576052]

Gabriel SS, et al., 2017. Bone marrow transplantation improves proximal tubule dysfunction in a mouse model of Dent disease. Kidney Int. 91 (4), 842–855. 10.1016/j.kint.2016.11.016. [PubMed: 28143656]

Gianesello L, et al., 2017,. Albumin uptake in human podocytes: a possible role for the cubilin-amnionless (CUBAM) complex. Sci. Rep 7 (1). 10.1038/s41598-017-13789-z.13705, 20. [PubMed: 28154414]

Gianesello L, et al., 2020. Genetic Analyses in Dent Disease and Characterization of CLCN5 Mutations in Kidney Biopsies. Int. J. Mol. Sci 21 (2). 10.3390/ijms21020516.

Gianesello L, Ceol M, Priante G, Anglani F, Prete DD, 2016. Proteins Lost and Found: Not Only a Tubular Matter. J Clin Nephrol Kidney Dis 1 (1), 1002.

Gianesello L et al., "Human Parietal Epithelial Cells Express Tubular Protein Uptake System in Vivo," Nephrol. Dial. Transplant, vol. 33, no. suppl_1, pp. i71–i72, 5 2018, doi: 10.1093/ndt/gfy104.FP070.

Gorvin CM, et al., 4. 2013. Receptor-mediated endocytosis and endosomal acidification is impaired in proximal tubule epithelial cells of Dent disease patients. Proc. Natl. Acad. Sci. U. S. A 110 (17), 7014–7019. 10.1073/pnas.1302063110. [PubMed: 23572577]

Grand T, et al., 2009. Novel CLCN5 mutations in patients with Dent's disease result in altered ion currents or impaired exchanger processing. Kidney Int. 76 (9), 999–1005. 10.1038/ki.2009.305. [PubMed: 19657328]

Grand T, et al., 2011. Heterogeneity in the processing of CLCN5 mutants related to Dent disease. Hum. Mutat 32 (4), 476–483. 10.1002/humu.21467. [PubMed: 21305656]

Grieschat M, Alekov AK, 2012. Glutamate 268 regulates transport probability of the anion/proton exchanger ClC-5. J. Biol. Chem 287 (11), 8101–8109. 10.1074/jbc.M111.298265. [PubMed: 22267722]

Grimm RH, Svendsen KH, Kasiske B, Keane WF, Wahi MM, 1997. Proteinuria is a risk factor for mortality over 10 years of follow-up. MRFIT Research Group. Multiple Risk Factor Intervention Trial. Kidney Int. Suppl 63, S10–S14.

Güngör T, et al., 2020. A case of Type 1 Dent disease presenting with isolated persistent proteinuria. Turk Pediatri Ars 9 (55), 72–75. 10.5152/TurkPediatriArs.2018.6540.

Günther W, Lüchow A, Cluzeaud F, Vandewalle A, Jentsch TJ, 1998. ClC-5, the chloride channel mutated in Dent's disease, colocalizes with the proton pump in endocytotically active kidney cells. Proc. Natl. Acad. Sci. U. S. A 95 (14), 8075–8080. 10.1073/pnas.95.14.8075. [PubMed: 9653142]

Günther W, Piwon N, Jentsch TJ, 2003. The ClC-5 chloride channel knock-out mouse - an animal model for Dent's disease. Pflugers Arch. 445 (4), 456–462. 10.1007/s00424-002-0950-6. [PubMed: 12548389]

Günthner R, Wagner M, Thurm T, Ponsel S, Höfele J, Lange-Sperandio B, 2018. Identification of co-occurrence in a patient with Dent's disease and ADA2-deficiency by exome sequencing. Gene 649, 23–26. 10.1016/j.gene.2018.01.060. [PubMed: 29391272]

Guven A, et al., 2017. Mutational analysis of PHEX, FGF23 and CLCN5 in patients with hypophosphataemic rickets. Clin. Endocrinol. (Oxf.) 87 (1), 103–112. 10.1111/cen.13347. [PubMed: 28383812]

Hara-Chikuma M, Wang Y, Guggino SE, Guggino WB, Verkman AS, 2005. Impaired acidification in early endosomes of ClC-5 deficient proximal tubule. Biochem. Biophys. Res. Commun 329 (3), 941–946. 10.1016/j.bbrc.2005.02.060. [PubMed: 15752747]

Harvey KF, Kumar S, 1999. Nedd4-like proteins: an emerging family of ubiquitin- protein ligases implicated in diverse cellular functions. Trends Cell Biol. 9 (5), 166–169. 10.1016/s0962-8924(99)01541-x. [PubMed: 10322449]

Hayama A, Uchida S, Sasaki S, Marumo F, 2000. Isolation and characterization of the human CLC-5 chloride channel gene promoter. Gene 261 (2), 355–364. 10.1016/s0378-1119(00)00493-5. [PubMed: 11167024]

Herak-Kramberger CM, Brown D, Sabolić I, 1998. Cadmium inhibits vacuolar H (+)-ATPase and endocytosis in rat kidney cortex. Kidney Int. 53 (6), 1713–1726. 10.1046/j.1523-1755.1998.00914.x. [PubMed: 9607204]

Hodgin JB, Corey HE, Kaplan BS, D'Agati VD, 2008. Dent disease presenting as partial Fanconi syndrome and hypercalciuria. Kidney Int. 73 (11), 1320–1323. 10.1038/sj.ki.5002785. [PubMed: 18235437]

Hoopes RR, et al., 2004. Evidence for genetic heterogeneity in Dent's disease. Kidney Int. 65 (5), 1615–1620. 10.1111/j.1523-1755.2004.00571.x. [PubMed: 15086899]

Hoopes RR, et al., 2005. Dent Disease with mutations in OCRL1. Am. J. Hum. Genet 76 (2), 260–267. 10.1086/427887. [PubMed: 15627218]

Hryciw DH, et al., 2004. Nedd4–2 functionally interacts with ClC-5: involvement in constitutive albumin endocytosis in proximal tubule cells. J. Biol. Chem 279 (53), 54996–55007. 10.1074/jbc.M411491200. [PubMed: 15489223]

Hryciw DH, et al., 2006. Regulation of albumin endocytosis by PSD95/Dlg/ZO-1 (PDZ) scaffolds. Interaction of Na+-H+ exchange regulatory factor-2 with ClC-5. J. Biol. Chem 281 (23), 16068–16077. 10.1074/jbc.M512559200. [PubMed: 16601121]

Hryciw DH, Wang Y, Devuyst O, Pollock CA, Poronnik P, Guggino WB, 2003. Cofilin interacts with ClC-5 and regulates albumin uptake in proximal tubule cell lines. J. Biol. Chem 278 (41), 40169–40176. 10.1074/jbc.M307890200. [PubMed: 12904289]

Hryciw DH, Ekberg J, Pollock CA, Poronnik P, 2006. ClC-5: a chloride channel with multiple roles in renal tubular albumin uptake. Int. J. Biochem. Cell Biol 38 (7), 1036–1042. 10.1016/j.biocel.2005.09.009. [PubMed: 16226913]

Hryciw DH, Jenkin KA, Simcocks AC, Grinfeld E, McAinch AJ, Poronnik P, 2012. The interaction between megalin and ClC-5 is scaffolded by the Na+-H+ exchanger regulatory factor 2 (NHERF2) in proximal tubule cells. Int. J. Biochem. Cell Biol 44 (5), 815–823. 10.1016/j.biocel.2012.02.007. [PubMed: 22349218]

Silva IV, et al., 2003. The ClC-5 knockout mouse model of Dent's disease has renal hypercalciuria and increased bone turnover". J. Bone Miner. Res. Off. J. Am. Soc. Bone Miner. Res 18 (4), 615–623. 10.1359/jbmr.2003.18.4.615.

Igarashi T, et al., 1998. Functional characterization of renal chloride channel, CLCN5, mutations associated with Dent'sJapan disease. Kidney Int. 54 (6), 1850–1856. 10.1046/j.1523-1755.1998.00203.x. [PubMed: 9853249]

Jentsch TJ, Günther W, Pusch M, Schwappach B, 1995. Properties of voltage-gated chloride channels of the ClC gene family. J. Physiol 482, 19S–25S. 10.1113/jphysiol.1995.sp020560. [PubMed: 7730971]

Jentsch TJ, Stein V, Weinreich F, Zdebik AA, 2002. Molecular structure and physiological function of chloride channels. Physiol. Rev 82 (2), 503–568. 10.1152/physrev.00029.2001. [PubMed: 11917096]

Kaneko K, Hasui M, Hata A, Hata D, Nozu K, 2010. Focal segmental glomerulosclerosis in a boy with Dent-2 disease. Pediatr. Nephrol. Berl. Ger 25 (4), 781–782. 10.1007/s00467-009-1362-z.

Kerjaschki D, Farquhar MG, 1982. The pathogenic antigen of Heymann nephritis is a membrane glycoprotein of the renal proximal tubule brush border. Proc. Natl. Acad. Sci. U. S. A 79 (18), 5557–5561. 10.1073/pnas.79.18.5557. [PubMed: 6752952]

Klein R, Moss SE, Klein BE, 1993. Is gross proteinuria a risk factor for the incidence of proliferative diabetic retinopathy? Ophthalmology 100 (8), 1140–1146. 10.1016/s0161-6420(93)31514-9. [PubMed: 8341493]

Kriz W, Lemley KV, 2017. Mechanical challenges to the glomerular filtration barrier: adaptations and pathway to sclerosis. Pediatr. Nephrol. Berl. Ger 32 (3), 405–417. 10.1007/s00467-016-3358-9.

Li F, et al., 2016. Dent Disease in Chinese Children and Findings from Heterozygous Mothers: Phenotypic Heterogeneity, Fetal Growth, and 10 Novel Mutations. J. Pediatr 174, 204–210.e1. 10.1016/j.jpeds.2016.04.007. [PubMed: 27174143]

Lieske JC, Milliner DS, Beara-Lasic L, Harris P, Cogal A, and Abrash E, "Dent Disease," in GeneReviews®, Adam MP, Ardinger HH, Pagon RA, Wallace SE, Bean LJ, Stephens K, and Amemiya A, Eds. Seattle (WA): University of Washington, Seattle, 1993.

Lim Woo-Hyun, Choi Eue-Keun, Han Kyung-Do, Rhee Tae-Min, Lee Hyun-Jung, Lee So-Ryoung, Kang Si-Hyuck, Cha Myung-Jin, Oh Seil, 2017. Proteinuria Detected by Urine Dipstick Test as a Risk Factor for Atrial Fibrillation: A Nationwide Population-Based Study. Sci. Rep 7 (1). 10.1038/s41598-017-06579-0.

Lim B, Yang J, Do W, Fogo AB, 2016. Pathogenesis of Focal Segmental Glomerulosclerosis. J Pathol. Transl. Med 50 (6), 405. [PubMed: 27744657]

Lloyd SE, et al., 1996. A common molecular basis for three inherited kidney stone diseases. Nature 379 (6564), 445–449. 10.1038/379445a0. [PubMed: 8559248]

Lloyd SE, et al., 1997. Characterisation of renal chloride channel, CLCN5, mutations in hypercalciuric nephrolithiasis (kidney stones) disorders. Hum. Mol. Genet 6 (8), 1233–1239. 10.1093/hmg/6.8.1233. [PubMed: 9259268]

Lourdel S, Grand T, Burgos J, González W, Sepúlveda FV, Teulon J, 2012. ClC-5 mutations associated with Dent's disease: a major role of the dimer interface. Pflugers Arch. 463 (2), 247–256. 10.1007/s00424-011-1052-0. [PubMed: 22083641]

Ludwig M, et al., 2003. Four additional CLCN5 exons encode a widely expressed novel long CLC-5 isoform but fail to explain Dent's phenotype in patients without mutations in the short variant. Kidney Blood Press. Res 26 (3), 176–184. 10.1159/000071883. [PubMed: 12886045]

Ludwig M, et al., 2005. Functional evaluation of Dent's disease-causing mutations: implications for ClC-5 channel trafficking and internalization. Hum. Genet 117 (2–3), 228–237. 10.1007/s00439-005-1303-2. [PubMed: 15895257]

Luyckx VA, et al., 1998. Intrarenal and subcellular localization of rat CLC5. Am. J. Physiol 275 (5), F761–F769. 10.1152/ajprenal.1998.275.5.F761. [PubMed: 9815133]

Mansour-Hendili L, et al., 2015. Mutation Update of the CLCN5 Gene Responsible for Dent Disease 1. Hum. Mutat 36 (8), 743–752. 10.1002/humu.22804. [PubMed: 25907713]

Matsumoto A, et al., 2018. Severe Osteomalacia with Dent Disease Caused by a Novel Intronic Mutation of the CLCN5 gene. Intern. Med. Tokyo Jpn 57 (24), 3603–3610. 10.2169/internalmedicine.1272-18.

Maunsbach AB, 1966. Albumin Absorption by Renal Proximal Tubule Cells. Nature 212 (5061), 546–547. 10.1038/212546a0. [PubMed: 5970207]

Mellman I, Fuchs R, Helenius A, 1986. Acidification of the endocytic and exocytic pathways. Annu. Rev. Biochem 55, 663–700. 10.1146/annurev.bi.55.070186.003311. [PubMed: 2874766]

Meyer S, Savaresi S, Forster IC, Dutzler R, 2007. Nucleotide recognition by the cytoplasmic domain of the human chloride transporter ClC-5. Nat. Struct. Mol. Biol 14 (1), 60–67. 10.1038/nsmb1188. [PubMed: 17195847]

Mohammad-Panah R, et al., 2003. The chloride channel ClC-4 contributes to endosomal acidification and trafficking. J. Biol. Chem 278 (31), 29267–29277. 10.1074/jbc.M304357200. [PubMed: 12746443]

Nakamura H, Kitazawa K, Honda H, Sugisaki T, 2005. Roles of and correlation between alpha-smooth muscle actin, CD44, hyaluronic acid and osteopontin in crescent formation in human glomerulonephritis. Clin. Nephrol 64 (6), 401–411. 10.5414/cnp64401. [PubMed: 16370152]

Neagoe I, Stauber T, Fidzinski P, Bergsdorf E-Y, Jentsch TJ, 2010. The late endosomal ClC-6 mediates proton/chloride countertransport in heterologous plasma membrane expression. J. Biol. Chem 285 (28), 21689–21697. 10.1074/jbc.M110.125971. [PubMed: 20466723]

Novarino G, Weinert S, Rickheit G, Jentsch TJ, 2010. Endosomal chloride-proton exchange rather than chloride conductance is crucial for renal endocytosis. Science 328 (5984), 1398–1401. 10.1126/science.1188070. [PubMed: 20430975]

Obermüller N, Gretz N, Kriz W, Reilly RF, Witzgall R, 1998. The swelling-activated chloride channel ClC-2, the chloride channel ClC-3, and ClC-5, a chloride channel mutated in kidney stone disease, are expressed in distinct subpopulations of renal epithelial cells. J. Clin. Invest 101 (3), 635–642. 10.1172/JCI1496. [PubMed: 9449697]

Okamoto T, Tajima T, Hirayama T, Sasaki S, 2012. A patient with Dent disease and features of Bartter syndrome caused by a novel mutation of CLCN5. Eur. J. Pediatr 171 (2), 401–404. 10.1007/s00431-011-1578-3. [PubMed: 21932010]

Oken DE, Flamenbaum W, 1971. Micropuncture studies of proximal tubule albumin concentrations in normal and nephrotic rats. J. Clin. Invest 50 (7), 1498–1505. 10.1172/JCI106635. [PubMed: 5090064]

Okkenhaug H, Weylandt K-H, Carmena D, Wells DJ, Higgins CF, Sardini A, 2006. "The human ClC-4 protein, a member of the CLC chloride channel/transporter family, is localized to the endoplasmic reticulum by its N-terminus", FASEB. J. Off. Publ. Fed. Am. Soc. Exp. Biol 20 (13), 2390–2392. 10.1096/fj.05-5588fje.

Pham P-C, et al., 2004. Hypertonicity increases CLC-5 expression in mouse medullary thick ascending limb cells. Am. J. Physiol. Renal Physiol 287 (4), F747–F752. 10.1152/ajprenal.00229.2003. [PubMed: 15161605]

Picollo A, Pusch M, 2005. Chloride/proton antiporter activity of mammalian CLC proteins ClC-4 and ClC-5. Nature 436 (7049), 420–423. 10.1038/nature03720. [PubMed: 16034421]

Piwon N, Günther W, Schwake M, Bösl MR, Jentsch TJ, 2000. ClC-5 Cl− -channel disruption impairs endocytosis in a mouse model for Dent's disease. Nature 408 (6810), 369–373. 10.1038/35042597. [PubMed: 11099045]

Platt C, Jadresic L, Dudley J, Hartley JL, 2014. Dent's disease complicated by an acute Budd-Chiari syndrome. BMJ Case Rep. 2014. 10.1136/bcr-2013-200937.

Prabakaran T, et al., 2011. Receptor-mediated endocytosis of α-galactosidase A in human podocytes in Fabry disease. PloS One 6 (9), e25065. 10.1371/journal.pone.0025065. [PubMed: 21949853]

Prabakaran T, Christensen EI, Nielsen R, Verroust PJ, 2012. Cubilin is expressed in rat and human glomerular podocytes. Nephrol. Dial. Transplant. Off. Publ. Eur. Dial. Transpl. Assoc. - Eur. Ren. Assoc 27 (8), 3156–3159. 10.1093/ndt/gfr794.

Pusch M, Ludewig U, Jentsch TJ, 1997. Temperature dependence of fast and slow gating relaxations of ClC-0 chloride channels. J. Gen. Physiol 109 (1), 105–116. 10.1085/jgp.109.1.105. [PubMed: 8997669]

Pusch M, Zifarelli G, 2015. ClC-5: Physiological role and biophysical mechanisms. Cell Calcium 58 (1), 57–66. 10.1016/j.ceca.2014.09.007. [PubMed: 25443653]

Raja KA, et al., 2002. Responsiveness of hypercalciuria to thiazide in Dent's disease. J. Am. Soc. Nephrol. JASN 13 (12), 2938–2944. 10.1097/01.asn.0000036869.82685.f6. [PubMed: 12444212]

Reed AAC, et al., 2010. CLC-5 and KIF3B interact to facilitate CLC-5 plasma membrane expression, endocytosis, and microtubular transport: relevance to pathophysiology of Dent's disease. Am. J. Physiol. Renal Physiol 298 (2), F365–F380. 10.1152/ajprenal.00038.2009. [PubMed: 19940036]

Reinhart SC, et al., 1995. Characterization of carrier females and affected males with X- linked recessive nephrolithiasis. J. Am. Soc. Nephrol. JASN 5 (7), 1451–1461. [PubMed: 7703383]

Remuzzi A, Sangalli F, Fassi A, Remuzzi G, 2007. Albumin concentration in the Bowman's capsule: Multiphoton microscopy vs micropuncture technique. Kidney Int. 72 (11), 1410–1411. 10.1038/sj.ki.5002501. [PubMed: 18004314]

Rickheit G, et al., 2010. Role of ClC-5 in renal endocytosis is unique among ClC exchangers and does not require PY-motif-dependent ubiquitylation. J. Biol. Chem 285 (23), 17595–17603. 10.1074/jbc.M110.115600. [PubMed: 20351103]

Sahali D, Mulliez N, Chatelet F, Dupuis R, Ronco P, Verroust P, 1988. Characterization of a 280-kD protein restricted to the coated pits of the renal brush border and the epithelial cells of the yolk sac. Teratogenic effect of the specific monoclonal antibodies. J. Exp. Med 167 (1), 213–218. 10.1084/jem.167.1.213. [PubMed: 2891781]

Sakamoto H, et al., 1999. Cellular and subcellular immunolocalization of ClC-5 channel in mouse kidney: colocalization with H + -ATPase. Am. J. Physiol 277 (6), F957–F965. 10.1152/ajprenal.1999.277.6.F957. [PubMed: 10600943]

Samardzic M, Pavicevic S, Ludwig M, Bogdanovic R, 2011. Effect of growth hormone replacement therapy in a boy with Dent's disease: a case report. J. Med. Case Reports 5, 400. 10.1186/1752-1947-5-400.

Sancakli O, Kulu B, and Sakallioglu O, "A novel mutation of Dent's disease in an 11-year-old male with nephrolithiasis and nephrocalcinosis," Arch. Argent. Pediatr, vol. 116, no. 3, pp. e442–e444, 01 2018, doi: 10.5546/aap.2018.eng.e442. [PubMed: 29756720]

Santo Y, et al., 2004. Examination of megalin in renal tubular epithelium from patients with Dent disease. Pediatr. Nephrol. Berl. Ger 19 (6), 612–615. 10.1007/s00467-004-1445-9.

Satoh N, et al., 2016. A pure chloride channel mutant of CLC-5 causes Dent's disease via insufficient V-ATPase activation. Pflugers Arch. 468 (7), 1183–1196. 10.1007/s00424-016-1808-7. [PubMed: 27044412]

Scheel O, Zdebik AA, Lourdel S, Jentsch TJ, 2005. Voltage-dependent electrogenic chloride/proton exchange by endosomal CLC proteins. Nature 436 (7049), 424–427. 10.1038/nature03860. [PubMed: 16034422]

Scheinman SJ, Pook MA, Wooding C, Pang JT, Frymoyer PA, Thakker RV, 1993. Mapping the gene causing X-linked recessive nephrolithiasis to Xp11.22 by linkage studies. J. Clin. Invest 91 (6), 2351–2357. 10.1172/JCI116467. [PubMed: 8099916]

Schmieder RE, et al., 2011. Changes in Albuminuria Predict Mortality and Morbidity in Patients with Vascular Disease. J. Am. Soc. Nephrol 22 (7), 1353–1364. 10.1681/ASN.2010091001. [PubMed: 21719791]

Schmieder S, Bogliolo S, Ehrenfeld J, 2007. N-glycosylation of the Xenopus laevis ClC-5 protein plays a role in cell surface expression, affecting transport activity at the plasma membrane. J. Cell. Physiol 210 (2), 479–488. 10.1002/jcp.20882. [PubMed: 17111367]

Schwake M, Friedrich T, Jentsch TJ, 2001. An internalization signal in ClC-5, an endosomal Cl-channel mutated in dent's disease. J. Biol. Chem 276 (15), 12049–12054. 10.1074/jbc.M010642200. [PubMed: 11116157]

Sethi SK, Ludwig M, Kabra M, Hari P, Bagga A, 2009. Vitamin A responsive night blindness in Dent's disease. Pediatr. Nephrol. Berl. Ger 24 (9), 1765–1770. 10.1007/s00467-009-1198-6.

Sheffer-Babila S, Chandra M, Speiser PW, 2008. Growth hormone improves growth rate and preserves renal function in Dent disease. J. Pediatr. Endocrinol. Metab. JPEM 21 (3), 279–286. 10.1515/jpem.2008.21.3.279. [PubMed: 18540256]

Slattery C, et al., 2011. Na+-H+ exchanger regulatory factor 1 (NHERF1) PDZ scaffold binds an internal binding site in the scavenger receptor megalin. Cell. Physiol. Biochem. Int. J. Exp. Cell. Physiol. Biochem. Pharmacol 27 (2), 171–178. 10.1159/000325219.

Smith AJ, Lippiat JD, 2010. Direct endosomal acidification by the outwardly rectifying CLC-5 Cl(−)/H(+) exchanger. J. Physiol 588 (Pt 12), 2033–2045. 10.1113/jphysiol.2010.188540. [PubMed: 20421284]

Smith AJ, Reed AAC, Loh NY, Thakker RV, Lippiat JD, 2009. Characterization of Dent's disease mutations of CLC-5 reveals a correlation between functional and cell biological consequences and protein structure. Am. J. Physiol. Renal Physiol 296 (2), F390–F397. 10.1152/ajprenal.90526.2008. [PubMed: 19019917]

Solanki AK, et al., 2018. A Novel CLCN5 Mutation Associated With Focal Segmental Glomerulosclerosis and Podocyte Injury. Kidney Int. Rep 3 (6), 1443–1453. 10.1016/j.ekir.2018.06.003. [PubMed: 30426109]

Steinmeyer K, Schwappach B, Bens M, Vandewalle A, Jentsch TJ, 1995. Cloning and functional expression of rat CLC-5, a chloride channel related to kidney disease. J. Biol. Chem 270 (52), 31172–31177. 10.1074/jbc.270.52.31172. [PubMed: 8537381]

Stolte H, Schurek H-J, Alt JM, 1979. Glomerular albumin filtration: A comparison of micropuncture studies in the isolated perfused rat kidney with in vivo experimental conditions. Kidney Int. 16 (3), 377–384. 10.1038/ki.1979.140. [PubMed: 529684]

Tang X, et al., 2016. Functional and transport analyses of CLCN5 genetic changes identified in Dent disease patients. Physiol. Rep 4 (8). 10.14814/phy2.12776.

Tanuma A et al., "Functional characterization of a novel missense CLCN5 mutation causing alterations in proximal tubular endocytic machinery in Dent's disease," Nephron Physiol, vol. 107, no. 4, pp. p87–97, 2007, doi: 10.1159/000111253. [PubMed: 18025833]

Thakker RV, 1997. Chloride channels cough up. Nat. Genet 17 (2), 125–127. 10.1038/ng1097-125. [PubMed: 9326919]

Thakker RV, 2000. Pathogenesis of Dent's disease and related syndromes of X-linked nephrolithiasis. Kidney Int. 57 (3), 787–793. 10.1046/jM523-1755.2000.00916.x. [PubMed: 10720930]

Tojo A, Endou H, 1992. Intrarenal handling of proteins in rats using fractional micropuncture technique. Am. J. Physiol 263 (4 Pt 2), F601–F606. 10.1152/ajprenal.1992.263.4.F601. [PubMed: 1415732]

Tosetto E, et al., 2009. Locus heterogeneity of Dent's disease: OCRL1 and TMEM27 genes in patients with no CLCN5 mutations. Pediatr. Nephrol. Berl. Ger 24 (10), 1967–1973. 10.1007/s00467-009-1228-4.

Tosetto E, et al., 2009. Novel mutations of the CLCN5 gene including a complex allele and A 5' UTR mutation in Dent disease 1. Clin. Genet 76 (4), 413–416. 10.1111/j.1399-0004.2009.01212.x. [PubMed: 19673950]

Tosetto E, Casarin A, Salviati L, Familiari A, Lieske JC, Anglani F, 2014. Complexity of the 5'UTR region of the CLCN5 gene: eleven 5'UTR ends are differentially expressed in the human kidney. BMC Med. Genomics 7, 41. 10.1186/1755-8794-7-41. [PubMed: 25001568]

Tsai W-C, et al., 2016. Risk Factors for Development and Progression of Chronic Kidney Disease: A Systematic Review and Exploratory Meta-Analysis. Medicine (Baltimore) 95 (11), e3013. 10.1097/MD.0000000000003013. [PubMed: 26986114]

Tung C-W, Hsu Y-C, Shih Y-H, Chang P-J, Lin C-L, 2018. Dipstick Proteinuria and Reduced Estimated Glomerular Filtration Rate as Independent Risk Factors for Osteoporosis. Am. J. Med. Sci 355 (5), 434–441. 10.1016/j.amjms.2017.12.011. [PubMed: 29753373]

van Berkel Y, Ludwig M, van Wijk JAE, Bökenkamp A, 2017. Proteinuria in Dent disease: a review of the literature. Pediatr. Nephrol. Berl. Ger 32 (10), 1851–1859. 10.1007/s00467-016-3499-x.

Vandewalle A, et al., 2001. Tissue distribution and subcellular localization of the ClC-5 chloride channel in rat intestinal cells. Am. J. Physiol. Cell Physiol 280 (2), C373–C381. 10.1152/ajpcell.2001.280.2.C373. [PubMed: 11208533]

Waldegger S, Jentsch TJ, 2000. From tonus to tonicity: physiology of CLC chloride channels. J. Am. Soc. Nephrol. JASN 11 (7), 1331–1339. [PubMed: 10864591]

Wang SS, et al., 2000. Mice lacking renal chloride channel, CLC-5, are a model for Dent's disease, a nephrolithiasis disorder associated with defective receptor-mediated endocytosis. Hum. Mol. Genet 9 (20), 2937–2945. 10.1093/hmg/9.20.2937. [PubMed: 11115837]

Wang A, et al., 2018. Proteinuria and risk of stroke in patients with hypertension: The Kailuan cohort study. J. Clin. Hypertens. Greenwich Conn 20 (4), 765–774. 10.1111/jch.13255.

Wang X et al., "Glomerular Pathology in Dent Disease and Its Association with Kidney Function," Clin. J. Am. Soc. Nephrol. CJASN, vol. 11, no. 12, pp. 2168–2176, 07 2016, doi: 10.2215/CJN.03710416. [PubMed: 27697782]

Wellhauser L, Luna-Chavez C, D'Antonio C, Tainer J, Bear CE, 2011. ATP induces conformational changes in the carboxyl-terminal region of ClC-5. J. Biol. Chem 286 (8), 6733–6741. 10.1074/jbc.M110.175877. [PubMed: 21173145]

Wen M, Shen T, Wang Y, Li Y, Shi X, Dang X, 2018. Next-Generation Sequencing in Early Diagnosis of Dent Disease 1: Two Case Reports. Front. Med 5, 347. 10.3389/fmed.2018.00347.

Wojciechowski D, et al., 2018. Barttin Regulates the Subcellular Localization and Posttranslational Modification of Human Cl−/H+ Antiporter ClC-5. Front. Physiol 9, 1490. 10.3389/fphys.2018.01490. [PubMed: 30405442]

Wong W, Poke G, Stack M, Kara T, Prestidge C, Flintoff K, 2017. Phenotypic variability of Dent disease in a large New Zealand kindred. Pediatr. Nephrol. Berl. Ger 32 (2), 365–369. 10.1007/s00467-016-3472-8.

Wrong OM, Norden AG, Feest TG, 1994. Dent's disease; a familial proximal renal tubular syndrome with low-molecular-weight proteinuria, hypercalciuria, nephrocalcinosis, metabolic bone disease, progressive renal failure and a marked male predominance. QJM Mon. J. Assoc. Physicians 87 (8), 473–493.

Wu F, et al., 2003. Modeling study of human renal chloride channel (hCLC-5) mutations suggests a structural-functional relationship. Kidney Int. 63 (4), 1426–1432. 10.1046/j.1523-1755.2003.00859.x. [PubMed: 12631358]

Wu F et al., "Mutational analysis of CLC-5, cofilin and CLC-4 in patients with Dent's disease," Nephron Physiol, vol. 112, no. 4, pp. p53–62, 2009, doi: 10.1159/000225944. [PubMed: 19546591]

Ye Q, et al., 2019. Multicenter study of the clinical features and mutation gene spectrum of Chinese children with Dent disease. Clin. Genet 10.1111/cge.13663.

Yin J, Kuang Z, Mahankali U, Beck TL, 2004. Ion transit pathways and gating in ClC chloride channels. Proteins 57 (2), 414–421. 10.1002/prot.20208. [PubMed: 15340928]

Zaniew M, et al., 2017. Dent disease in Poland: what we have learned so far? Int. Urol. Nephrol 49 (11), 2005–2017. 10.1007/s11255-017-1676-x. [PubMed: 28815356]

Zdebik AA, et al., 2008. Determinants of anion-proton coupling in mammalian endosomal CLC proteins. J. Biol. Chem 283 (7), 4219–4227. 10.1074/jbc.M708368200. [PubMed: 18063579]

Zhang Y, Fang X, Xu H, Shen Q, 2017. Genetic Analysis of Dent's Disease and Functional Research of CLCN5 Mutations. DNA Cell Biol 36 (12), 1151–1158. 10.1089/dna.2017.3731. [PubMed: 29058463]

Zhao X et al., "Albumin induces CD44 expression in glomerular parietal epithelial cells by activating extracellular signal-regulated kinase 1/2 pathway.," J Cell Physiol, vol. 234, no. 5, p. 7224, 5 2019. [PubMed: 30362534]

Zifarelli G, Pusch M, 2009. Intracellular regulation of human ClC-5 by adenine nucleotides. EMBO Rep. 10 (10), 1111–1116. 10.1038/embor.2009.159. [PubMed: 19713962]

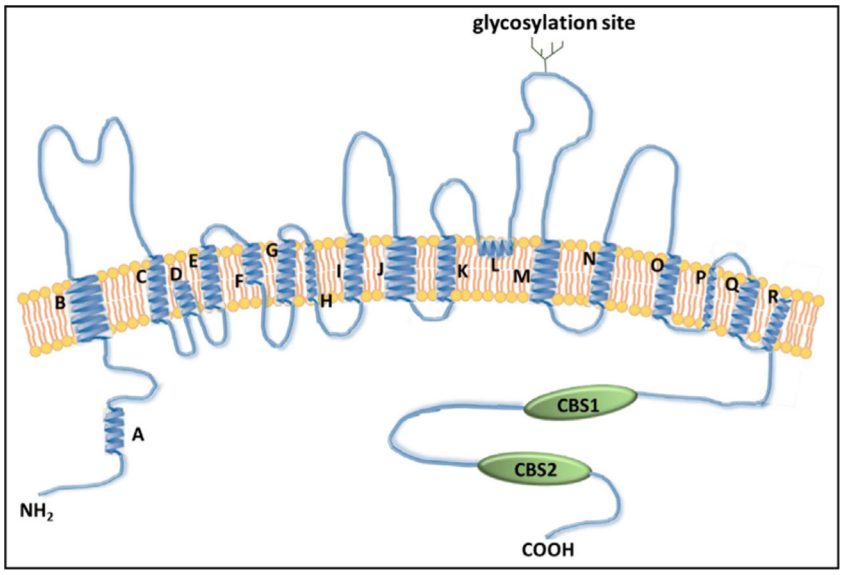

Fig. 1. ClC-5 protein structure.

Schematic representation of the ClC-5 protein showing the 18 alpha-helixes (from A to R), the two CBS domains, and the main glycosylation site.

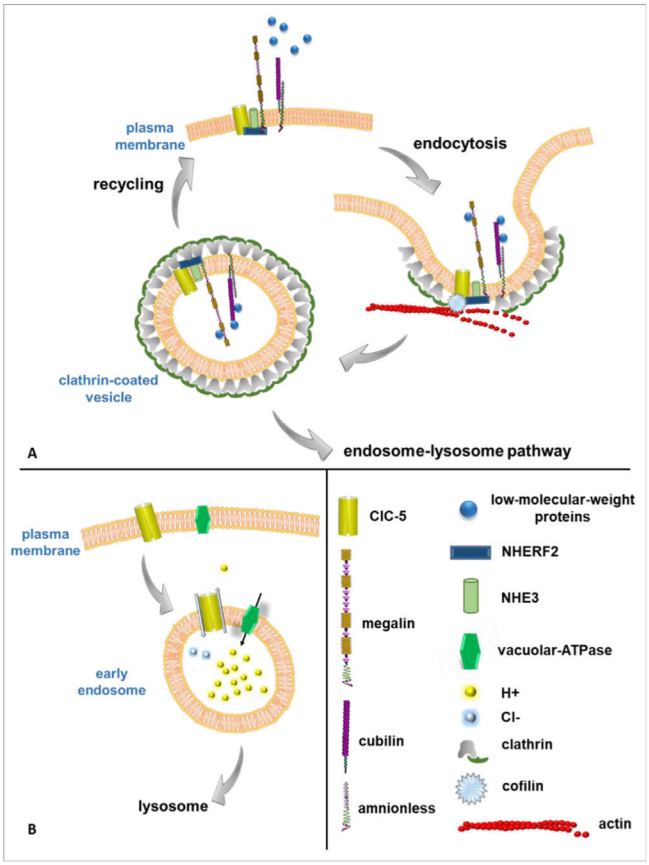

Fig. 2. ClC-5 and its protein partners.

ClC-5 is involved in two different processes conducted by PTCs: the endocytosis of low-molecular-weight proteins (A); and endosomal-lysosomal acidification (B). These two processes are closely connected, and require collaboration between ClC-5 and several other actors with distinctive features.

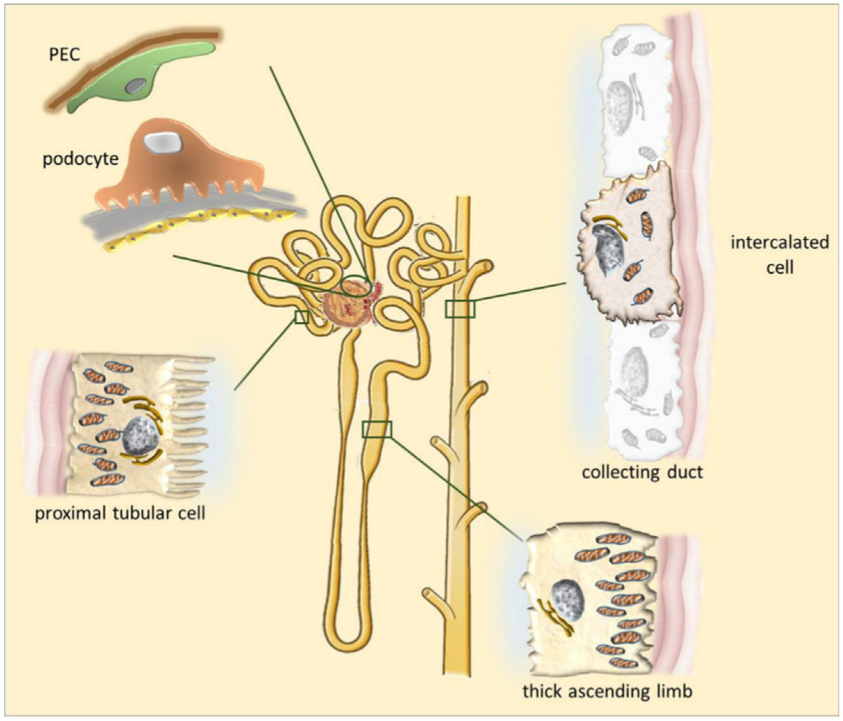

Fig. 3. ClC-5 expression in the kidney.

ClC-5 is expressed in various tubular segments (proximal tubule, thick ascending limb of Henle's loop, alpha-intercalated cells of the collecting duct), and in glomerular cells (podocytes and parietal epithelial cells [PECs]).

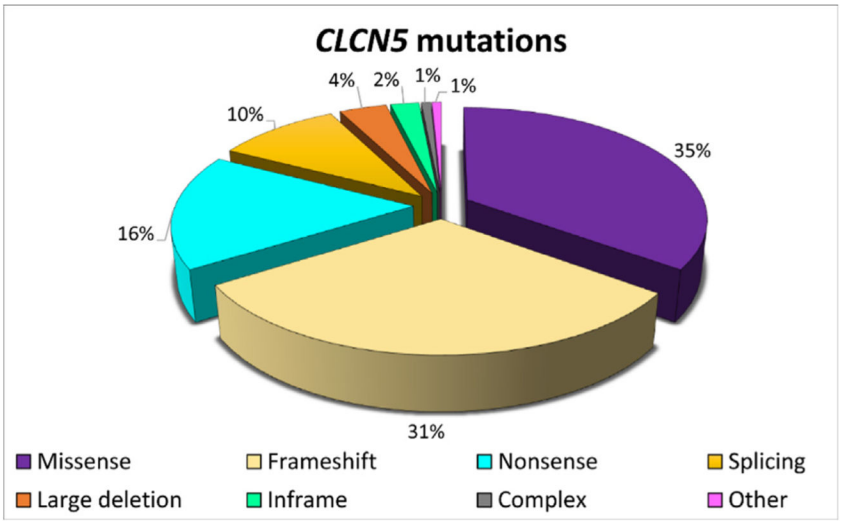

Fig. 4. Distribution of CLCN5 gene mutations.

The graph shows the pro-portions of the different ***CLCN5*** mutations identified. Other: Alu insertion and 5′UTR mutations.

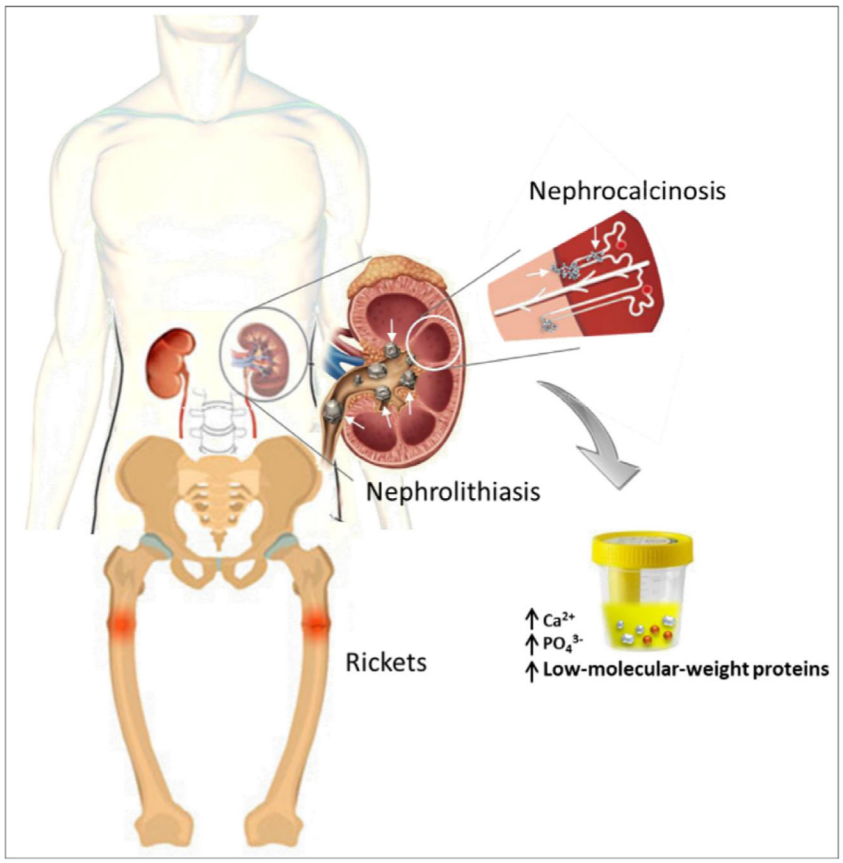

Fig. 5. Dent disease 1, clinical phenotype.

DD1 is associated with the variable presence of a number of clinical signs.

Table 1

Basic properties of *CLCN5* and its protein product ClC-5.

	Homo sapiens	***Mus musculus***
Gene symbol	*CLCN5*	*CLCN5*
Chromosome	Xp11.23	X A1.1; X 3.21
Ensembl ID	ENSG00000171365	ENSMUSG00000004317
Uniprot ID	P51795	Q9WVD4
Exon numbers	12, coding exons: 11	12, coding exons: 11
Protein length (in aa's)	746	746
Protein mass (KDa)	83	83