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Experimental Nephrology and Genetics: Case Study of Genetic Interest

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Medullary Sponge Kidney and Its Relationship with Primary Distal Renal Tubular Acidosis: Case Reports and a Comprehensive Genetics-First Approach

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Abstract
Medullary sponge kidney (MSK) is a description of radiographic features. However, the pathogenesis of MSK remains unclear. MSK is supposed to be the cause of secondary distal renal tubular acidosis (dRTA), although there are case reports suggesting that MSK is a complication of primary dRTA. In addition to these reports, we report 3 patients with metabolic acidosis and MSK, in whom primary dRTA is confirmed by molecular genetic analyses of *SLC4A1* and *ATP6V1B1* genes. With a comprehensive genetics-first approach using the 100,000 Genomes Rare Diseases Project dataset, the association between MSK and primary dRTA is examined. We showed that many patients with MSK phenotypes are genetically tested with a gene panel which does not contain dRTA-associated genes, revealing opportunities for missed genetic diagnosis. Our cases highlight that the radiological description of MSK is not a straightforward disease or clinical phenotype. Therefore, when an MSK appearance is noted, a broader set of causes should be considered including genetic causes of primary dRTA as the underlying reason for medullary imaging abnormalities.

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Introduction

Medullary sponge kidney (MSK) is described as a rare malformative kidney condition that is associated with nephrocalcinosis and nephrolithiasis, precalyceal cystic anomalies, and tubular disorders (e.g., secondary incomplete distal renal tubular acidosis [dRTA], hypocitraturia, urinary concentration defects, and hypercalciuria) [1, 2]. MSK is established radiographically with intravenous urography or urographic computed tomography scan as the gold standard. However, in clinical practice today these studies are no longer conducted, and most patients have a suggestion of MSK by ultrasound, especially in childhood. The following tetrad on ultrasound is described: hypoechoic medullary areas, hyperechoic spots, microcystic dilatation of papillary zone, and multiple calcifications in each papilla [3]. However, several of these features can fit with known kidney diseases, including renal cystic diseases and other renal ciliopathies, renal dysplasia, and diseases associated with nephrocalcinosis. As MSK is a radiographic description, it is important to distinguish underlying treatable kidney diseases.

Although the radiologic features are known for MSK, the pathogenesis remains unclear. However, there is evidence for a genetic etiology, as demonstrated by reports of family clustering with an autosomal dominant inheritance [4]. In addition, a link with glial cell-derived neurotrophic factor polymorphisms as risk factor for MSK with variable expression and incomplete penetrance has been reported [5]. Evidence for monogenic causes of MSK is limited; however, recently two observed cases and a similar case in the literature indicate that MSK could be a consequence of pathogenic variation of the H+ proton pump subunit *ATP6V1B1* and *ATP6V0A4* genes [6]. We here describe 3 additional patients with MSK and primary dRTA confirmed by genetic analyses of *SLC4A1* and *ATP6V1B1* genes. To further explore the association between these genetic variants and MSK, we also performed a genetics-first approach in the 100,000 Genomes Project (100KGP). The 100KGP is a British initiative that involved sequencing more than 85,000 individuals with rare diseases and cancers, including both patients and unaffected family members, to explore the role of genes in health and disease. In this initiative, whole-genome sequencing data are linked to electronic health record data from the National Health Service (NHS) in England. The rare disease program of 100KGP currently includes over 70,000 individuals, among which there are more than 4,000 individuals with renal and urinary tract disorders. Apart from studying the prevalence of the MSK and dRTA association, we also utilized this same dataset to investigate the differential diagnosis of medullary phenotypes and the genetic testing requested in patients with these medullary phenotypes. This can provide insights into whether physicians are considering primary dRTA as a potential diagnosis of medullary phenotypes and which causal genes are identified in patients with medullary phenotypes.

Case Reports

Case 1

A 41-year-old man was referred for genetic analysis and counseling about reproductive options to the Department of Human Genetics at the Amsterdam University Medical Center, The Netherlands, almost a year after kidney transplantation. He was diagnosed with MSK after an episode of urolithiasis in Hungary 14 years ago. A few years later, he came to The Netherlands where he presented again with an obstructive ureter stone. The diagnosis of MSK was confirmed based on extensive medullary calcifications on CT scan and a normal anion gap metabolic acidosis (MA) at random blood gas analysis. There was a progressive worsening of renal function resulting in end-stage kidney disease. While there were no known family members with kidney disease, it is worth noting that he had no siblings, and he also had no contact with his father or his father's family. Genetic analyses of an exome-based gene panel of 495 kidney disease-associated genes (online suppl. File 1; for all online suppl. material, see https://doi.org/10.1159/000538037) identified a known pathogenic missense variant in *SLC4A1* (NM_000342.4 c.1765C>T p.Arg589Cys) [7]. Additional biochemical analysis showed no associated hemolytic anemia/spherocytosis in this patient.

Case 2

A 30-year-old woman was referred to the Genetics Department of the University Medical Center Utrecht, The Netherlands, for preconceptional counseling of MSK and a positive family history for this disease. When she was 20 years old, she had presented with back pain. On CT urography, there was an interpolar concrement in the right kidney and a hyperdense medulla, which was reported as MSK. There was also hypokalemia and renal tubular acidosis, for which treatment with potassium citrate had been started. There was a positive family history for MSK and dRTA in an autosomal dominant pattern. The mother of our patient has a moderately to severely decreased kidney function. She has two brothers with nephrolithiasis. Mother's mother is on renal replacement therapy, and she was diagnosed with MSK as well. Previous genetic evaluation in the proband had consisted of just single-gene testing of *UMOD* (which was negative) in 2012. Genetic analysis was performed on DNA of her mother in 2015 using a targeted panel for renal cystic disease (online suppl. File 1), which was also negative. Ten years later, the proband came to our hospital and renal tubular acidosis was confirmed by a urinary acidification test.

A further, genetic analysis (exome-based gene panels, online suppl. File 1) identified a heterozygous missense variant in *SLC4A1* (NM_000342.4: c.2706T>A p.Asp902Glu), which was classified as a variant of unknown clinical significance (VUS) [7]. Given the

clinical phenotype of dRTA, the classification was upgraded from a VUS to a likely pathogenic variant. With this variant, the patient was counseled that there is a 50% change of an affected child and that treatment is advised to reduce the risk of nephrocalcinosis, nephrolithiasis, and chronic kidney disease (CKD). Furthermore, the patient was informed about reproductive options and family members about risk for disease and screening options. There was no associated hemolytic anemia/spherocytosis.

Case 3

A 10-year-old girl was referred to the Pediatric Nephrology Department, University Medical Center Utrecht, The Netherlands, for follow-up of her clinical and biochemical diagnosis of renal tubular acidosis. She had presented at the age of 2 years with a febrile urinary tract infection in the referring hospital. In the workup after that infection, a sensorineural hearing loss was identified. Laboratory investigations showed hyperchloremic MA, hypokalemia, high urine pH, and nephrocalcinosis; a diagnosis of dRTA was confirmed. On kidney ultrasound scanning, there were hypoechoic areas, hyperechoic spots, and microcystic dilatation of papillary zone. On X-ray, there were multiple calcifications suggestive of a diagnosis of MSK.

Molecular analysis in 2010 of genes known (online suppl. File 1) to be involved in renal tubular acidosis with deafness showed the presence of a homozygous pathogenic variant of the *ATP6V1B1* gene (NM_001692.3: c.1155dup p.Ile385fs) [7]. During follow-up with problems of adherence, she underwent frequent episodes of ureteral and vesical urolithiasis.

Methods: Genetics-First Approach in 100KGP

We used the 100KGP to study the phenotypes described in patients with (likely) pathogenic variants in dRTA genes (*SLC4A1*, *ATP6V0A4*, and *ATP6V1B1*) to estimate the prevalence of dRTA with MSK in these patients. Inclusion and genotyping of participants in the 100KGP, managed by Genomics England Limited (GEL), were previously described [8].

First, the multi-sample variant call format dataset release (aggV2), containing genome-wide sequencing data of 78,195 participants, was used to search for participants carrying rare variants in *SLC4A1*, *ATP6V0A4*, and *ATP6V1B1*. In December 2022, we extracted high and moderate impact rare variants using the Combined Annotation-Dependent Depletion (CADD) score and minor allele frequency (MAF). We used a scaled CADD score of >20 (or no CADD score if not applicable) and an MAF <0.0001 for *SLC4A1* and MAF <0.005 for (potential) biallelic variants in *SLC4A1*, *ATP6V0A4*, and *ATP6V1B1*. We then assessed variants identified in participants with a Human Phenotype Ontology (HPO) term related to MSK, cystic kidney disease (CyKD), or dRTA (online suppl. File 2), and/or participants with a recruited disease of renal and urinary tract disorders or falling in the category of ultra-rare disorders. The variants were assessed using the American College of Medical Genetics and Genomics (ACMG) criteria [7]; details on phenotype, family history, requested gene panels, and genetic testing results were extracted (release v16). Phenotype counts that would identify <5 individuals in the 100KGP dataset were either masked or a broader description of the phenotype was extracted to adhere to privacy policies.

Second, we used the 100KGP dataset to explore the differential diagnosis of any type of medullary phenotype and which gene panels are requested in these patients. We extracted phenotypes, acquired diseases, requested gene panels, including positive results (case annotated as solved with [likely] pathogenic variant), from patients with ICD10 code Q61.5 (medullary cystic kidney [MCK] disease) and HPO terms HP:0012408 (medullary nephrocalcinosis [MN]) and HP:0008659 (multiple small medullary cysts). It should be noted that the HPO term HP:0008659 is described in the HPO database as "multiple small medullary renal cysts" with the synonyms "medullary cystic disease" and "medullary sponge kidney disease" with the comment "this feature is the cardinal sign of medullary cystic disease, also known as medullary sponge disease."

Results: Genetics-First Approach in 100KGP

Using this genetics-first approach in the 100KGP dataset, we were able to identify patients with *SLC4A1*, *ATP6V0A4*, and *ATP6V1B1* variants (Fig. 1; Table 1). After filtering (online suppl. File 3), 8 patients from five families with rare heterozygous *SLC4A1* variants remained with a phenotype suspect for dRTA. Three of these patients from 1 family had previously been diagnosed with a pathogenic *SLC4A1* variant and dRTA and two (both patients with a VUS) had renal tract calcification (RTC). We identified 1 patient with a homozygous pathogenic variant in *SLC4A1,* with a phenotype described as CyKD with CKD stage 4. This variant was not previously identified in the patient. For *ATP6V0A4,* 4 participants from 3 families were identified with homozygous pathogenic variants, which had previously been diagnosed with dRTA caused by the identified variant. Additional patients that passed our filtering criteria had a VUS, including the patient with *ATP6V1B1,* and a phenotype not likely fitting with dRTA.

In two families with a pathogenic variant in one of the dRTA genes, MCKs were described. Through Genomics England Clinical Collaboration request, we contacted the clinical teams for additional phenotyping. In the family with a pathogenic heterozygous SLC4A1 variant (No. 1 in Table 1), an approximately 40-year-old woman had an MA from infancy, hypocitraturia, and a CKD stage 2. Her two sons (born between 2005 and 2015) had polyuria. One of them also had an MA and CKD stage 2. Interestingly, renal ultrasound and CT reported MN/MSK in the mother and her sons carrying the heterozygous *SLC4A1* variant. None of them had spherocytosis.

The patient (no. 6 in Table 1) with a biallelic pathogenic variant in *SLC4A1* and CyKD is an approximately 50-year-old male which presented initially in 2005 with impaired kidney function and bilateral small renal calculi. He always had a tendency to a low serum potassium level. His venous bicarbonate levels have generally been in the range of 22–25 mmol/L and on two occasions below 20 mmol/L with now an eGFR of 27 mL/min/1.73 m^2. Establishing family history has been impossible. Reviewing his imaging showed multiple small renal cysts and, on occasion, renal calculi. Magnetic resonance cholangiopancreatography recently showed bilateral renal cysts, both cortical and medullary, some of which appear associated with the renal collecting system. So, this patient may have a dRTA phenotype which is relatively mild and could be the consequence of decreased kidney function. Similarly, the imaging, which is not ideal, could be compatible with MSK.

To explore the genetic differential diagnosis of medullary phenotypes, we selected all patients with any medullary phenotype described in their medical history (n = 161) (online suppl. File 4). Their whole-genome sequencing data harbored no additional variants in the dRTA genes, whereas other causative genes were

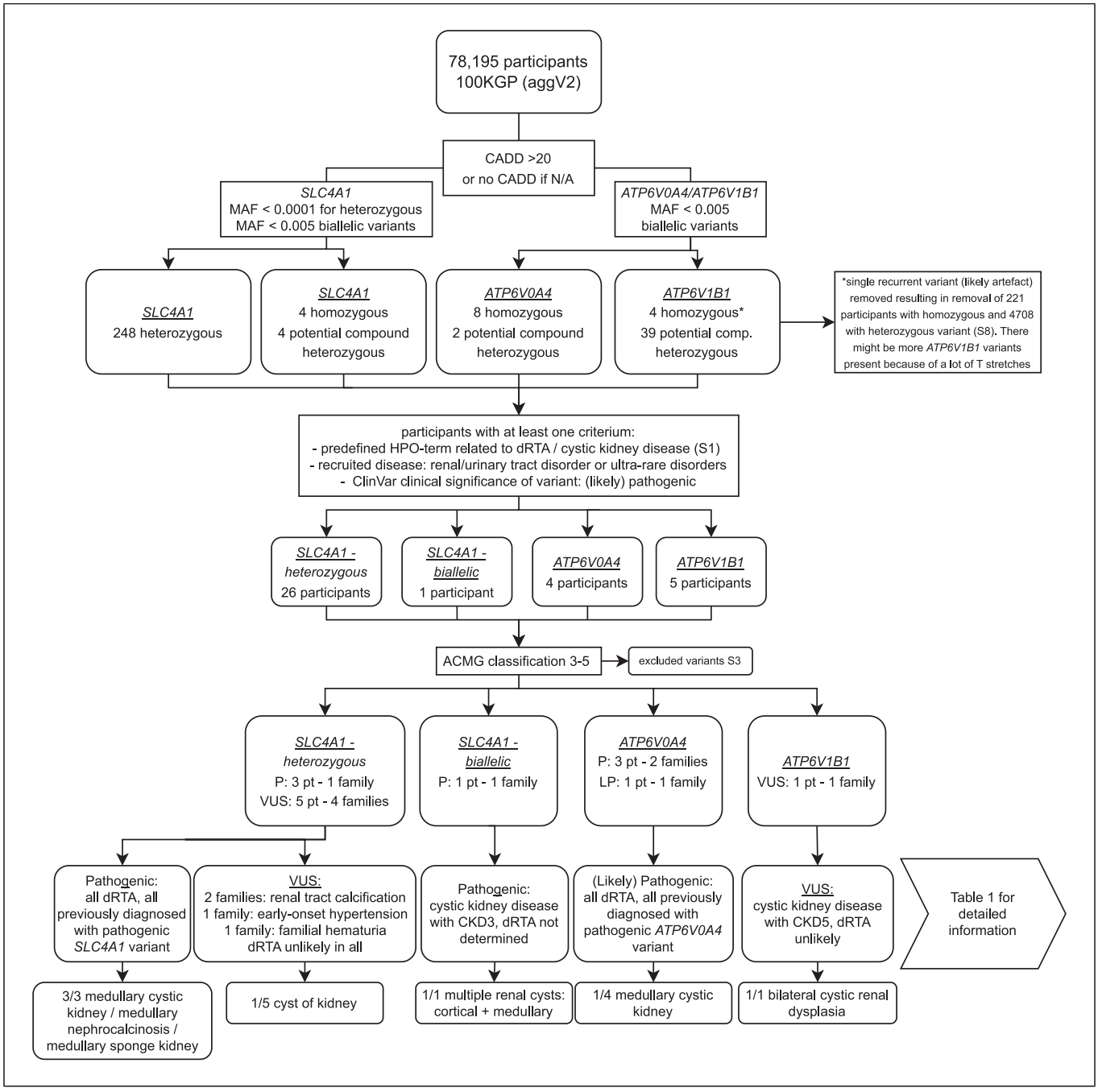

Fig. 1. Flowchart with filtering strategy for dRTA genes in the 100KGP.

identified in these families (Table 2). In 39 families totally, a causative variant was identified, most often involving a ciliary gene.

The 161 patients with a medullary phenotype had a presentation that is most often described by their doctor as either CyKD, RTCs, congenital anomalies of the kidney and urinary tract, and/or renal tubular acidosis (online suppl. File 5). When we look at what gene panels were requested and applied for these patients, a variety of gene panels are found, with the "CyKD" panel being requested the most often (online suppl. File 6). Interestingly, very few of these panels include the known and described dRTA-related genes. From the requested gene panels, only "RTC (or nephrolithiasis/nephrocalcinosis)," "renal tubular acidosis," and "renal tubulopathies" include genes *SLC4A1*, *ATP6V0A4*, and *ATP6V1B1*.

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Table 1. Variants in dRTA genes + phenotype of patients in 100KGP resulting from filtering strategy, including requested gene panels

No.	ACMG classification; zygosity	Variant (cDNA/ predicted protein structure)	Pathogenicity predictions			P/F	100KGP exit questionnaire	Approx. year of birth patient	Phenotype	Comment on dRTA/ MSK phenotype	Recruited disease	Family history	Requested gene panels	Gene of interest included in panel	Additional patients in 100KGP with this variant
			CADD	predictions#	gnomAD										
SLC4A1, NM_000342.4*															
1	Pathogenic; het	c.1765C>T/ p.Arg589Cys	33	3/3 damaging	0	3/1	Likely pathogenic variant	1980–85; 2005–10; 2010–15	3/3 MCK/ MN/MSK; MA; CKD st. 2	Suspect	RTC (or NorN)	Positive (mother and sibling with condition, 1 more affected sibling without DNA available)	RTC (or NorN) + RTA	Yes, in both panels	No
2	VUS; het	c.1535T>C/ p.Leu512Pro	29.7	3/3 damaging	0	1/1	Negative	1970–75	HT, MCK	Nonsuspect	Extreme early-onset HT	No known affected family members	Extreme early-onset HT	No	No
3	VUS; het	c.2350C>G/ p.Pro784Ala	27.4	3/3 damaging	0	2/1	Negative	1950–55; 1975–80	Proteinuria hematuria. No dRTA, MN, or MSK	Nonsuspect	Familial hematuria	Positive (segregates with condition)	Proteinuric renal disease + hematuria	No	1 family member +1 unrelated (breast cancer, HT)
4	VUS; het	c.760G>C/p. Glu254Gln	24	3/3 benign	0	1/1	Negative	1975–80	Calculus of kidney, recurrent UTI	Possibly suspect. Missing data	RTC (or NorN)	No known affected family members	NorN + familial nonsyndromic CHD	Yes	No
5	VUS; het	c.1832C>T/ p.Pro611Leu	25.8	2/3 damaging	×1 het	1/1	Negative	1985–90	NorN, calculus of kidney	Possibly suspect. Missing data	RTC (or NorN)	Positive (father + sibling affected, no DNA available)	RTC (NorN)	Yes	No
6	Pathogenic; hom	c.2573C>A/ p.Ala858Asp	27.4	3/3 damaging	×18 het	1/1	Negative	1970–75	MCK CKD st. 4 DM 2; hypokalemia	Possibly suspect. Missing data	CyKD	Negative (no DNA available from family members for segregation)	CyKD	No	×3 het., ×0 hom. or compound het.
ATP6V0A4, NM_020632.3**															
7	Likely pathogenic; hom	c.1346G>A/ p.Arg449His	28.4	3/3 damaging	×3 het	1/1	Likely pathogenic variant	1990–95	RTA; MCK		RTA	Positive (father reported as affected, no data/DNA available)	RTC (or NorN) + CyKD + RTA	Yes	No
8	Pathogenic; hom	c.1691+1G>A/ splice site variant	31	Splice donor variant	×3 het	1/1	Fits with full phenotype	2010–15	RTA		RTA	Negative	NorN + tubulopathies	Yes	No

Table 1 (continued)

No.	ACMG classification; zygosity	Variant (cDNA/ predicted protein structure)	Pathogenicity predictions			P/F	100KGP exit questionnaire	Approx. year of birth patient	Phenotype	Comment on dRTA/ MSK phenotype	Recruited disease	Family history	Requested gene panels	Gene of interest included in panel	Additional patients in 100KGP with this variant
			CADD	predictions[#]	gnomAD										
9	Pathogenic; hom	c.1312_1313del/ p.Asp438GlnfsTer2	NA	NA	0	2/ 1	Fits twice with full phenotype	2005–10; 2005–10	Hearing loss		RTA	Positive (affected sibling has same variant)	NorN + tubulopathies	Yes	Yes, affected sibling
ATP6V1B1, NM_001692.4***															
10	VUS; hom	c.1051C>T/ p.Pro351Ser	27.2	3/3 damaging	×13 het; ×0 hom	1/ 1	Negative	10–15	Bilateral cystic dysplasia, small VSD, CKD st. 5		CyKD	Negative, parents both het. carrier	CyKD + ciliopathy disorders	no	×8 het. present in dataset

CADD, Combined Annotation-Dependent Depletion; P/F, patient(s)/families; VUS, variance of unknown significance; het., heterozygous; hom., homozygous; MCK, medullary cystic kidney; MN, medullary nephrocalcinosis; MSK, medullary sponge kidney; MA, metabolic acidosis; CKD st., chronic kidney disease stage; HT, hypertension; UTI, urinary tract infection; DM, diabetes mellitus; RTC, renal tract calcification; CyKD, cystic kidney disease; CHD, congenital heart disease; NorN, nephrolithiasis or nephrocalcinosis; ACMG, American College of Medical Genetics and Genomics. More detailed data could not be provided due to 100KGP privacy policy. *All *SLC4A1* variants concern ENST00000262418.12 and ENSP00000262418.6. **All *ATP6V0A4* variants concern ENST00000310018.7 and ENSP00000308122.2. ***ATP6V1B1 variant concerns ENST00000234396.10 and ENSP00000234396.4. # Pathogenicity prediction tools: SIFT, MutationTaster, Polyphen-2.

Discussion

In recent reviews, dRTA was mentioned as secondary to MSK [9, 10]. However, in one of the earliest named reports it was concluded that renal acidification defects play an important role in the pathogenesis of MSK and so MSK might be secondary to dRTA [11]. This was supported by a recent case report of 2 patients with MSK and a mutation of the H + -ATPase genes, *ATP6V1B1* and *ATP6V0A4*, both known to cause primary dRTA [6]. One comment on this report spoke of a puzzling association of MSK and dRTA [12]. Although we are cautious in considering our cases as definite MSK patients, they previously have been diagnosed with MSK and years thereafter molecular analysis revealed mutations in either of the genes *ATP6V1B1* and *SLC4A1* which cause primary dRTA.

dRTA is a rare inherited disease characterized by the inability of the distal nephron to maximally increase the urinary secretion of protons in the presence of MA. The diagnosis is established in patients with biallelic causative variants in *ATP6V0A4*, *ATP6V1B1*, *FOXI1*, and *WDR72* genes in an autosomal recessive trait [9]. Variants in these last two have only been rarely described [13]. Heterozygous or biallelic pathogenic variants in the *SLC4A1* gene also cause dRTA and are also/can be associated with spherocytosis [9]. In addition, there is an autosomal dominant form of *SLC4A1* which is often identified by family screening rather than obvious symptoms [14].

Patients with autosomal recessive dRTA may develop symptoms very early, even during infancy. Symptoms include failure to thrive, vomiting, polydipsia, polyuria, feeding problems, and episodes of dehydration. In addition, there is a hyperchloremic MA, hypokalemia, hypercalciuria, and hypocitraturia. Together, these urinary changes favor early-onset MN and nephrolithiasis [9] resulting in CKD in most adult subjects [15].

The clinical and biochemical phenotype of individuals with heterozygous variants in *SLC4A1* gene is typically less severe during childhood compared to those with variants in other causative genes [9]. This observation is consistent with subjects 1 and 2 described above, who presented with distal acidification defect, but did not exhibit failure to thrive, growth retardation, or other problems in childhood. However, as they (case 1) or their family members (case 2) aged, they developed kidney failure, which could have been caused by an untreated RTA with calcinosis. Additionally, a family from 100KGP with the same heterozygous *SLC4A1* variant exhibited features of MSK and dRTA. The case of the patient with biallelic variant in *SLC4A1* and CyKD is

Table 2. Causative genes identified in patients with any medullary phenotype in the 100KGP (n = 161)

Identified gene in solved family	MCK (ICD10)	MN (HPO)	Multiple small medullary cysts (HPO)	Total per phenotype	Phenotype group solved gene	Total solved per phenotype group
ATP6V0A4	1			1	dRTA	2
SLC4A1	1			1	dRTA	
PKD1	1		14	15	Ciliopathy – ADPKD	23
PKD2			6	6	Ciliopathy – ADPKD	
DNAJB11			2	2	Ciliopathy – ADPKD	
CEP290	2			2	Ciliopathy – AR	9
EYS	1			1	Ciliopathy – AR	
PKHD1			2	2	Ciliopathy – AR	
NPHP1	1		1	2*	Ciliopathy – AR	
NPHP4	1			1	Ciliopathy – AR	
SDCCAG8	1			1	Ciliopathy – AR	
TMEM67			1	1	Ciliopathy – AR	
EYA1	1			1	CAKUT	2
SALL1		1		1	CAKUT	
SLC7A9		1		1	Cystinuria	1
COL4A4		1		1	Alport	1
Maternal UPD15 (Prader-Willi)	1			1	Other	1
Total result	11	3	25	40*	Total number of solved families	39

*Patients with multiple phenotypes; total number of patients is 39.

less clear-cut. There is no evidence for dRTA, and there are only features of MSK on magnetic resonance cholangiopancreatography. This homozygous variant was previously associated with a more severe phenotype with failure to thrive [16]. Therefore, the significance of this finding in this patient is subject to debate.

Our cases highlight that the radiological description of MSK is not a straightforward disease or entity. Therefore, when MSK is mentioned in a case file, one should consider broader medullary phenotypes among which a diagnosis of dRTA is primary cause for secondary medullary imaging abnormalities and consider additional tests (e.g., genetic testing). In our cases, we saw that MSK can be a confusing description, and apart from dRTA, physicians have also considered ciliopathies, because of the renal cysts that can be part of the MSK spectrum, autosomal dominant tubulointerstitial kidney disease given the medullary localization, and the dominant inheritance as highlighted by case 2. Very recently, it was shown that also *PKHD1* variants (both biallelic and in some cases monoallelic) can lead to a spectrum of kidney phenotypes that may resemble MSK as well as atypical CyKD [17, 18]. An association with MSK and *HNF1B* has also been described [19]. Since *UMOD*, *HNF1B*, and *MUC1* cases can exhibit medullary cysts when pathogenic variants are present, and these genes are not listed in Table 2, we explored the phenotypes described in solved patients with a causative (likely) pathogenic variant in one of these genes within the 100KGP. These did include (general) renal cysts but were not always further specified (online suppl. File 7).

One might consider adding the dRTA genes *SLC4A1*, *ATP6V1B1*, and *ATP6V0A4*, to renal ciliopathy (CyKD) and autosomal dominant tubulointerstitial kidney disease gene panels to prevent missing this phenocopy. This was hitherto not the case in our center or in the well-known PanelApp (see Data Availability).

Our genetics-first approach in 100KGP did not identify definite undiscovered dRTA patients with a phenotype suggestive for MSK. We did find two families with MCKs that had previously been diagnosed with a causative pathogenic variant in a dRTA gene. We also identified a biallelic pathogenic *SLC4A1* variant in a CyKD patient, without evidence for dRTA. The variant is a known pathogenic variant with a high prevalence in South-East Asian populations [16, 20]. Further phenotyping of this patient and of patients with a VUS would be interesting. Although based on the reported phenotypes in some patients with a VUS, an undiscovered dRTA is unlikely.

A limitation of the 100KGP dataset is that the somewhat unspecific HPO term HP:0008659 (multiple small medullary renal cysts, with synonyms MCK and MSK disease) is used, which might be linked by physicians to patients that do not necessarily have a radiologic description fitting with MSK. The fact that HPO uses these different descriptions as synonyms for this single HPO term highlights there is ambiguity about whether or not MSK is a separate entity. It should also be noted that in the HPO database this term is not yet linked to the dRTA genes. Taking this limitation into account, our genetics-first approach showed the overall rarity of dRTA as a cause for MSK. Repeating this analysis in a clearly defined unselected MSK cohort would give a better estimate of the prevalence of monogenic causes, including dRTA, for MSK.

Herein, in three cases we show that patients with causal variants in primary dRTA genes can have renal imaging with nephrocalcinosis and additional features yielding the term MSK. In addition, the extensive genetics-first approach showed that many patients with medullary phenotypes that might well overlap in some with the term MSK are genetically tested with a gene panel not containing the dRTA genes. As the diagnosis of primary dRTA has consequences for health and treatment, it is recommended to perform genetic analysis or at least perform a clinical workup (blood gas, potassium, urinary pH) when there is a radiological suspicion of MSK. Given the phenotypic overlap of MSK with other medullary phenotype descriptions, we recommend a broad differential diagnosis – and consequently, critical appraisal of panel content in case of genetic testing – when medullary abnormalities are identified in a patient.

Our cases highlight that the radiological description of MSK is not a straightforward disease or clinical phenotype. Therefore, when an MSK appearance is noted, a broader set of causes should be considered including genetic causes of dRTA as the underlying reason for medullary imaging abnormalities.

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Statement of Ethics

This research is performed in accordance with the World Medical Association Declaration of Helsinki. In accordance with the rules laid down in the Dutch law, the Medical Research Involving Human Subjects Act (WMO), this research is exempted from requiring ethics approval. Subjects of this study have given their informed consent to publish their case.

Conflict of Interest Statement

The authors have no conflicts of interest to declare.

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Author Contributions

Gerrit van den Berg wrote the original draft. Laura R. Claus was a major contributor to writing the manuscript and performed genetic analysis. Bert van der Zwaag interpreted genetic data and made substantively revision. Phillis Lakeman, Lotte Kaasenbrood, and John A. Sayer were providing case data and made substantively revision. Genomics England research consortium was providing genomic data and reviewed the work critically. Marc R. Lilien was providing case data and contributed to the conception and design of the work, as well as critically reviewing. Albertien M. van Eerde was providing case data, contributed to the conception and design of the work, and supervised data analysis and interpretation.

Data Availability Statement

All data generated or analyzed during this study are included in this published article [and its supplementary information files]. Genomics England PanelApp is available at https://panelapp.genomicsengland.co.uk.

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