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## EAST syndrome: Clinical, pathophysiological, and genetic aspects of mutations in *KCNJ10*

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### ABSTRACT

EAST syndrome is a recently described autosomal recessive disorder secondary to mutations in *KCNJ10* (*Kir4.1*), a gene encoding a potassium channel expressed in the brain, eye, ear and kidney. This condition is characterized by 4 cardinal features; Epilepsy, Ataxia, Sensorineural deafness, and (a renal salt-wasting) Tubulopathy, hence the acronym EAST syndrome. Here we review reported clinical manifestations, in particular the neurological signs and symptoms which typically have the most impact on the quality of life of patients. In addition we review the pathophysiology and genetic aspects of the disease. So far 14 different *KCNJ10* mutations have been published which either directly affect channel function or may lead to mislocalisation. Investigations of the pathophysiology may provide clues to potential treatments.

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ataxia; EAST syndrome; epilepsy; *KCNJ10*; kidney; *Kir4.1*; potassium channel; SeSAME syndrome; tubulopathy

### Introduction

EAST syndrome is an autosomal recessive disorder characterized by the tetrad of Epilepsy, Ataxia, Sensorineural deafness, and (a renal salt-wasting) Tubulopathy. It was first described as a distinct clinical entity in 2009 by 2 independent groups, who named this condition EAST syndrome and SeSAME syndrome (for Seizures, Sensorineural deafness, Ataxia, Mental retardation and Electrolyte imbalance), respectively.<sup>1,2</sup> However, the first clinical description may have been 16 y earlier by White et al.,<sup>3</sup> who reported a patient with epilepsy, sensorineural deafness, ataxia, mental retardation and a salt-wasting tubulopathy in addition to oculocutaneous albinism. So far 26 patients have been reported, in whom mutations in the underlying gene *KCNJ10* (also known as *Kir4.1*) have been identified.<sup>1,2,4-8</sup> This gene encodes a potassium channel. In human kidney, it locates in the distal nephron's basolateral membrane and defective function of this potassium channel leads to impaired distal salt reabsorption.<sup>1,2,8</sup> The biochemical phenotype thus resembles Gitelman syndrome, another salt-losing tubulopathy of the distal convoluted tubule, and potentially also Bartter syndromes which are due to



defective salt transport in the thick ascending limb of the loop of Henle.<sup>9-11</sup> Normotensive hypokalaemic metabolic alkalosis is the shared finding in these salt-wasting renal tubulopathies which are all inherited in an autosomal recessive manner.<sup>1,2,12</sup> A key distinguishing feature between these conditions and EAST syndrome are the additional extrarenal manifestations, especially of the neurological system in the latter.<sup>4</sup>

There is phenotypic heterogeneity as the spectrum and severity of symptoms varies across identified EAST syndrome cases and there is also intrafamilial variability as observations in families with more than one affected member illustrate the phenotypic variability that can be seen even within the same kindred with the same mutation.<sup>7</sup> There is no cure for this condition and management is focused on controlling seizure activity and on salt supplementation.<sup>1,2,4,7</sup>

### Clinical features

#### Neurological features

The neurological aspects are the most debilitating features of EAST syndrome and have the most impact on the quality of life of affected patients. While severity

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can vary, in some patients the ataxia dramatically limits the ability of both oral and written communication, leading to enormous frustration of these patients and a potentially mistaken diagnosis of mental retardation. The acronym SeSAME, which makes mental retardation a defining feature of the disease, is thus problematic. Not only has the term “mental retardation” been replaced in the United States by “intellectual disability” (“Rosa’s law”) (Pub. L. 111-256), but the difficulty of properly assessing intelligence in these patients may label them with this stigmatizing term undeservedly.

Detailed descriptions of neurological abnormalities, including brain imaging and electrophysiological investigations, have been reported in patients with confirmed EAST syndrome.<sup>1,2,4,7</sup> Typically, patients were born after uncomplicated pregnancies and with normal birth weights by normal vaginal deliveries at term. The majority were initially well with an uneventful neonatal period and first presentation to hospital was typically in the first few months of life either due to seizure activity, failure to thrive or an intercurrent illness with incidental biochemical abnormalities suggestive of a tubulopathy.<sup>4</sup>

### **Epilepsy**

Seizures are typically infantile in onset with the first fit usually occurring between 3 to 9 months of age; however, there are reports of a possible episode in the neonatal period.<sup>4,7</sup> These generalized seizures, which may be primary or secondary generalized seizures, were mostly easily controlled by a broad-spectrum anticonvulsant. In our review of affected children seen at Great Ormond Street Hospital, around half of the children were seizure free later on in childhood and adolescence and the other half subsequently developed focal seizures or continued to have tonic-clonic seizures when unwell.<sup>4</sup> Two of the patients also experienced non-convulsive and convulsive *status epilepticus* and there is reporting of a child developing a prolonged seizure activity resulting in a right-sided hemiparesis.<sup>4,7</sup> Seizure activity was controlled by a range of different anticonvulsants such as carbamazepine, oxcarbazepine, sodium valproate, lamotrigine, phenobarbital, phenytoin and diphenylhydantoin, however in a proportion of these children there was drug-resistant seizures of evident focal onset.<sup>2,4,7</sup>

Epileptic seizures were reported in all EAST patients studied except for 2 cases. Both these patients

were homozygote for the *KCNJ10* mutant p.A167V which experimentally has been shown to have around 60% residual activity.<sup>6</sup> They did however both have hearing impairment and tubulopathy, so the apparent lack of seizure activity in these patients raised interesting questions on organ specific effects of this mutation. Yet, another patient homozygote for the p.A167V exhibited the full spectrum of symptoms, including epilepsy and ataxia, raising the possibility that the epileptic activity in the other 2 patients may have not been picked up due to infrequency or subtlety.<sup>6,13</sup>

### **Ataxia and cerebellar dysfunction**

All of the children, who were old enough to have their cerebellar function assessed, presented with cerebellar dysfunction with varying severity and impediment of their daily activities.<sup>2,4,7</sup> In those that could walk, cerebellar dysfunction presented as characteristic broad-based ataxic gait, which resulted in unsteadiness and falls. Ataxia presented early in the majority of cases and was evident from when they were learning to walk, suggesting that ataxia from the outset is a feature of EAST syndrome. Other cerebellar signs include intention tremor, dysdiadochokinesis, dysmetria, truncal ataxia, titubation and slurred and scanning (explosive) speech.<sup>4,7</sup> Nystagmus appears to be an infrequent feature in these patients and has so far been only documented in one case.<sup>7</sup> There seems to be heterogeneity in muscle tone among EAST syndrome patients. Only one of the studies, which have described the clinical features in EAST syndrome cases, reported hypotonia as a shared feature of 4 patients who were referred for evaluation of Gitelman and Bartter syndromes and later found to have *KCNJ10* mutations.<sup>2</sup> Tone was more commonly reported as increased, either generally or exclusively in the lower limbs with a suggestion that there is increasing tone with age.<sup>4,7</sup> Other upper motor neuron signs exhibited included exaggerated deep tendon reflexes, ankle clonus and extensor plantar responses.<sup>4,7</sup> There were also reports of dystonic posturing in the upper limb and face in older patients.<sup>4</sup>

### **Developmental delay and intellectual disability**

Some form of neurodevelopmental delay was evident in most patients that were old enough to be assessed. Typically, there is delayed attainment of motor milestones with delays in sitting unsupported, walking

aided or walking independently.<sup>2,4,7</sup> Two children were never independently ambulant due to the severity of their ataxia.<sup>4</sup> In those that were able to walk, the age to acquire this skill ranged from around 17 months to 7 y.<sup>4,7</sup> EAST syndrome appears to be static or non-progressive in nature with no regression in attained milestones, however one patient who walked independently within the normal developmental time frame later lost the ability to walk after a prolonged fit.<sup>4</sup> This is likely to be the result of brain hypoxia from the prolonged seizure and not directly due to the *KCNJ10* mutation. Another case was evaluated on several occasions due to apparent regression in development however it is not clearly documented, whether there was actual loss of previously attained milestones.<sup>7</sup> There also appears to be phenotypic variability among siblings with EAST syndrome with regards to their motor development. In a family with 3 affected children, one sibling was able to walk at around 3.5 y of age, another at around 7.5 y and more unsteady, and another unable to walk independently at all.<sup>4</sup> With regard to language development, apart from the slurred speech and scanning dysarthria suffered by some patients, indicators of cerebellar disease, delayed onset of speech especially the ability to talk in full sentences was a common finding.<sup>2,4,7</sup>

Intellectual disability, as discussed above, can be difficult to properly assessed due to the impaired communication in severely affected patients. In our experience, there are clearly patients without intellectual disability who attend mainstream schools without special support, but these are a minority. Detailed neuropsychological assessments have not been undertaken in all patients with *KCNJ10* mutations, but most patients do require special support in school.<sup>4,14</sup>

### Neuroimaging and electrophysiological investigations

In the majority of cases, MRI of the brain detected pathological findings although normal brain MRI were reported in some.<sup>1,2,4,7</sup> However, on careful review of a cohort of children with EAST syndrome subtle changes in MRI were detected, that were previously reported as normal. This highlights the difficulties of interpreting such subtle findings as normal variant or related to disease pathology when only seen in an isolated case.<sup>1,4</sup> Importantly, automated regional quantitative volumetric analysis of the images showed

a pattern of variation from the norm involving comparatively smaller temporal cortical and larger frontal cortical regions. Such automated analysis may thus provide an alternative and potentially more objective approach to identify minor imaging abnormalities, especially in rare disease, where the radiologist is unlikely to see cohorts of patients.<sup>15</sup>

Identified aberrations on radiological review varied in severity as well as location, however, the majority showed abnormalities of the cerebellum which included subtle signal changes in the cerebellar dentate nuclei and cerebellar hypoplasia.<sup>4</sup> These changes are in keeping with ataxia, a manifestation of cerebellar dysfunction. Repeated MRI scans were conducted in 2 patients and these showed no evidence of any progressive changes supporting the concept that EAST syndrome is static.

Unlike brain MRI, almost all EEG that were conducted were unremarkable with no epileptiform discharges.<sup>1,2,4,7</sup> In one patient, there were localized spikes and sharp waves with slow activity with a subsequently normal EEG performed at later stage.<sup>4</sup> An electroencephalographic recording in another case revealed occasional sharp activity over one hemisphere with a slow and disorganized background with focal slowing. Yet the recording was reportedly carried out after a prolonged seizure that left the patient with a right sided weakness and subsequently more frequent seizures. Thus, the abnormal EEG activity could have been a consequence of the prolonged seizure, consistent also with the development of new MRI changes in that same patient in the form of a new lesion with restricted diffusion in the parietal area, which was deemed to be excitopathic.<sup>7</sup>

Electromyographic recordings were normal, as well as muscle biopsies when tested.<sup>1</sup> Nerve conduction studies on EAST patients were also normal except in one patient who was reported to have diminished nerve conduction velocities in the left peroneal and left tibial nerves. Nerve biopsy in the same patient revealed hypomyelination of the sural nerve with moderate progressive axonal neuropathy.<sup>2</sup> Interestingly, central myelination appeared normal on magnetic resonance imaging in the same patient.

### Hearing impairment

Sensorineural deafness is a consistent feature found in those with EAST syndrome.<sup>16</sup> All cases, in whom

hearing assessment could be undertaken, presented with some degree of hearing impairment except one case, who showed grossly normal hearing on audiometry and further audiological investigations were not documented.<sup>1,2,4,7</sup> Severity of hearing impairment is variable from mild, in some cases undiagnosed until formal testing was undertaken as part of the study, to severe, necessitating bilateral hearing aids.<sup>4</sup> Importantly, in those patients where serial hearing tests were available, no progression of the impairment was noted.<sup>4</sup>

### Renal

From a nephrological perspective EAST syndrome is very similar to Gitelman syndrome as biochemically there is hypokalaemic metabolic alkalosis, hypomagnesaemia and hypocalciuria as well as activation of the renin-angiotensin-aldosterone axis and loss of urinary sodium, potassium, chloride and magnesium. Both these conditions are the result of defective electrolyte transport in the distal convoluted tubule.<sup>1,2</sup> In the majority of cases initial clinical presentation of this potassium channel defect is due to neurological symptoms (see above) with the biochemical aspects of this syndrome generally often noted only later in childhood. These observations argue that the etiology of the seizures is an intrinsic consequence of *KCNJ10* dysfunction in the brain and not a secondary complication of electrolyte imbalance. However, biochemical abnormalities, especially hypokalaemic alkalosis have been reported already in infancy in some patients.<sup>2,4,7</sup>

Other symptoms suggestive of renal salt wasting include salt craving, polydipsia, polyuria and enuresis.<sup>2</sup> In our own experience, polydipsia and polyuria are rare, with most patients able to concentrate their urine normally, as evidenced by random urine osmolalities well above 800 mosm/kg. Blood pressure is typically low or at the lower range of normal.<sup>1</sup> Impaired growth and failure to thrive appears to respond well to medical management with salt supplementation and nutritional support. However, short stature as well as weight and head circumference measurements that were low for age and sex were reported later on in life in a few of the cases.<sup>2,4</sup>

There is variability in the degree of electrolyte disturbance among those with EAST syndrome, including variability within families. In some cases there is

an incidental mild hypomagnesaemia with no other biochemical disturbance and others with a more typical Gitelman-like abnormality of their electrolytes. Serial electrolyte measurements may reveal worsening and progression of the biochemical disturbance with age. In a particular study focused on the renal facets of *KCNJ10* mutations, there was a statistically significant effect of age on bicarbonate and potassium levels.<sup>7</sup> While serum bicarbonate levels increase and serum potassium decreases normally with age, in these patients, the evolving changes were noticeably greater than the expected physiological change. Bicarbonate levels are usually normal in the first few years of life followed by a rapid increment until around age 6 y with subsequent plateauing. There was insufficient data on magnesium levels to draw a conclusion on its progression [7]: in some patients renal magnesium loss became evident later on in life whereas in others hypomagnesaemia has been detected as early as the first year of life. Oral sodium, potassium or magnesium supplementation is prescribed in almost all cases where serum levels were found to be low.<sup>1,2,4,7</sup> In attempt to normalize serum potassium levels, potassium sparing diuretics have been used in some patients. However such diuretic treatment compounds the renal salt loss and thus may put patients at risk of potentially life threatening hypovolemia.<sup>4,7</sup>

When urine was tested, 24-hour urine sampling detected renal potassium and magnesium wasting, low urinary calcium-creatinine ratio and elevated 24-hour urinary aldosterone.<sup>2,4,7</sup> There was no proteinuria or glycosuria. Spot urine osmolality, a measure of the ability to concentrate urine, was frequently within high normal limits, attesting to non-compromised urinary concentrating ability. When measured, plasma renin activity (PRA) and aldosterone were always elevated.<sup>1</sup> Other serum electrolyte disturbances found rarely in patients with EAST syndrome included reduced serum sodium and chloride.<sup>4,7</sup>

Ultrasound examination of kidneys typically shows no abnormalities.<sup>1</sup> Moreover, in one patient with EAST syndrome, in whom a renal biopsy had been performed, careful electron microscopic examination revealed ultrastructural changes in the distal tubule with decreased basolateral infolding and a reduced number of mitochondria, as a morphological correlate to the impaired transport capacity in this segment.<sup>8</sup>



## Pathophysiology

### Neurological

Seizure activity may be explained by a process called potassium spatial buffering which is a mechanism that dictates the resting potential of neurons.<sup>17-19</sup> When excitation and repolarization occurs repeatedly there is considerable neuronal influx of sodium as well as potassium efflux and thus extracellular potassium build-up. This decreases the membrane potential rendering it susceptible to further excitations and so a predilection for seizure activity. It is thought that the excess extracellular potassium is taken up by glial cells via KCNJ10 potassium channels and subsequently distributed through gap junctions to minimize the risk of excessive excitability.<sup>17-20</sup> Thus any functional defect of KCNJ10 would disrupt this protective potassium “siphoning” and could explain why epilepsy is a cardinal feature of EAST syndrome.<sup>1,21</sup> The removal and redistribution of potassium by KCNJ10 also appears to occur in the Müller glia cells of the retina, which has been observed experimentally in mice.<sup>22</sup> It is likely that *KCNJ10* mutations impair the function of Müller cells also in humans as electroretinographic recordings in 4 EAST patients were reported as abnormal.<sup>23</sup>

### Inner ear

In mice *Kcnj10* is expressed in the intermediate cells of the stria vascularis of the inner ear, where they contribute to the rich potassium content of the endolymph.<sup>24</sup> These processes are imperative for the hearing mechanism as they facilitate potassium entry into the cochlear hair cells, which is needed for signal transduction.<sup>24,25</sup> This explains why hearing impairment is a feature of patients with EAST syndrome and why there is severe hearing loss in *Kcnj10* knockout mice.<sup>1,2,16,24,26</sup>

### Renal

One of the major roles of the kidneys is to maintain homeostasis of water, electrolytes and acid-base balance. KCNJ10 is a potassium channel located distal to the macula densa, in the distal convoluted tubules, connecting tubules and collecting ducts.<sup>27,28</sup> These structures, which are in the latter part of nephron, contribute to fluid and electrolyte balance and regulate pH through reabsorption of sodium, potassium, magnesium, calcium, and excretion of potassium and

hydrogen ions by energy dependent transcellular transport. These processes are regulated by hormones, most prominently aldosterone.<sup>29,30</sup>

The distal convoluted tubule and connecting tubule are characterized by numerous infoldings of their basolateral plasma membrane, where KCNJ10 potassium channels reside, as well as an increased number of mitochondria and  $\text{Na}^+/\text{K}^+$ -ATPases, all of which reflects the high transport activity in this segment.<sup>31</sup> In these parts of the nephron as well as cortical thick ascending limb, where *KCNJ10* is expressed, this potassium channel is responsible for potassium conductance and necessary for a process called pump-leak coupling.<sup>8,32,33</sup> According to this hypothesis, which was first proposed in 1958, the activity of  $\text{Na}^+/\text{K}^+$ -ATPase in epithelia, across which there is a high rate of transport, is rate limited by the availability of potassium outside the cell. Because of this constraint, potassium recycling is required and this process occurs at the basolateral membrane via potassium channels such as KCNJ10. In this way, KCNJ10 potassium channels and other potassium channels generate a hyperpolarized membrane voltage which facilitates ion transport that is voltage dependent such as sodium influx and chloride efflux.<sup>1,8,31,34</sup> The renal phenotype of EAST syndrome due to *KCNJ10* mutations is thus a result of an impairment of this process: the reduced potassium recycling affects  $\text{Na}^+/\text{K}^+$ -ATPase activity which consequent reduced transport activity.

Renal loss of sodium chloride results in activation of the renin-angiotensin-aldosterone axis leading to increased plasma levels of renin and aldosterone activity in EAST patients when measured.<sup>1</sup> Aldosterone stimulates sodium reabsorption by amiloride-sensitive epithelial sodium channels (ENaC), which are present on the apical membrane of the late distal convoluted tubule and connecting tubule. This sodium reabsorption, a compensatory mechanism to minimize the renal sodium wasting, is accompanied by potassium and hydrogen ion secretion in urine resulting in the hypokalaemic metabolic alkalosis found in patients with EAST syndrome.<sup>31</sup>

A characteristic feature seen in EAST syndrome, as well as other conditions with defective function of the distal convoluted tubule such as Gitelman syndrome, is the reduced absorption of magnesium in the distal convoluted tubule resulting in hypermagnesiuria and hypomagnesaemia, as well as increased calcium

reabsorption leading to hypocalciuria.<sup>35</sup> There are several proposed mechanisms in the literature to try to explain why there is renal loss of magnesium and retention of calcium; however this appears to be still under debate.<sup>31,36</sup>

### KCNJ10 expression

The clinical phenotype of *KCNJ10* mutations correlates with its gene expression in various organs such as the brain, ear, eye and kidney.<sup>1,16,22,24,27,28,37</sup> In the kidney, *KCNJ10* potassium channels and its homolog *KCNJ16* are located within the renal tubules.<sup>8,27,28</sup> In the mouse *kcnj10* and *kcnj16* channel proteins were localized by immunofluorescence to the basolateral membrane of the distal convoluted tubule, connecting tubule and cortical collecting duct.<sup>8</sup> *KCNJ* potassium channels are tetramers and thus consisting of 4 subunits. Association of different subunits (heteromers) can occur and electrophysiological studies on isolated tubules and cell lines clearly support the idea of *KCNJ10/KCNJ16* heteromers as the predominant in vivo conductance in the basolateral membrane of the distal convoluted tubule.<sup>8,34,38-41</sup> Thus *KCNJ16* constitutes a candidate gene for salt wasting tubulopathies.

As in rodent kidney, where *Kcnj10* is expressed pre and post macula densa,<sup>42</sup> there are some data in human kidney suggesting expression also in the basolateral membrane of cortical thick ascending limb of Henle's loop (TAL).<sup>8</sup> However, the clinical phenotype of a Gitelman-like tubulopathy suggests dysfunction

primarily in the DCT, as defective salt reabsorption is associated with hypercalciuria, i.e. a Bartter-like tubulopathy. Whether expression in TAL is too low to affect the phenotype or whether other potassium channels can compensate for loss-of-function in this segment remains to be elucidated.

Studies in mice showed that *Kcnj10* is also expressed in brain astroglia, Müller cells of the retina and the stria vascularis of the inner ear where it is needed for the generation of the endocochlear potential which is crucial for hearing.<sup>22,24</sup> Within the brain glia, *Kcnj10* is expressed predominantly in the cerebral cortex and cerebellar cortex as well as in the caudate nucleus and putamen.<sup>1,37</sup>

### KCNJ10 mutations

The *KCNJ10* gene encodes the *KCNJ10* channel, which is also known as ATP-sensitive inward rectifier potassium channel KIR4.1. It is located on chromosome 1q23.2 and so far 14 different pathogenic *KCNJ10* mutations have been described in the literature in cases with EAST syndrome.<sup>1,2,5-8,39,43</sup> EAST syndrome is an autosomal recessive disorder and all known EAST patients carry 2 mutant alleles of *KCNJ10*. As expected for most autosomal recessive conditions so far there is no reported symptomology or pathology in heterozygote relatives such as siblings and parents.<sup>1,31</sup> Patients with EAST syndrome either carry the same mutant allele on each strand of chromosome one and thus are homozygotes or carry 2 different mutant alleles, one on

**Table 1.** All known *KCNJ10* mutations published to date.

Mutation	Pathogenic Inherited State	Experimental Residual activity	Reference
p.T57I (c.170C>T)	• Homozygous missense mutation	Loss of function	7
p.R65C (c.193G>C)	• Homozygous missense mutation	< 20%	5
p.R65P (c.194G>C)	• Homozygous missense mutation	< 20%	1,2,8,38,39
	• Compound heterozygous missense / nonsense mutations with p.R199X		
p.F75C (c.224T>G)	• Homozygous missense mutation	Loss of function	6
p.F75L (c.225T>G)	• Homozygous missense mutation	< 10%	5
p.G77R (c.229G>C)	• Homozygous missense mutation	< 5%	1,8,39
p.V91fs197* (c.272delT)	• Homozygous frameshift mutation	Loss of function or < 2% residual activity	6
p.C140R (c.418T>C)	• Homozygous missense mutation	Loss of function	2,39,43
p.T164I (c.491C> T)	• Homozygous missense mutation	Loss of function	2,38,39,43
p.A167V (c.500C>T)	• Homozygous missense mutation	60%	2,6,38,39,43
	• Compound heterozygous missense mutations with p.R297C		
p.R175Q (c.524G>A)	• Homozygous missense mutation	< 5%	8
p.R199* (c.595C>T)	• Compound heterozygous missense / nonsense mutations with p.R65P	Loss of function	2,38,39,43
p.V259* (c.775delG)	• Homozygous nonsense mutation	Loss of function	5
p.R297C (c.889C>T)	• Compound heterozygous missense mutations with p.A167V	< 10%	2,5,38,39,43

each chromosome, therefore being compound heterozygotes.<sup>1,2,5-8</sup> Table 1 shows all known *KCNJ10* mutations published to date. Mutations identified so far are mainly missense mutations except for p.V259fs259\*, R199\* (so far only reported in heterozygous state) and p.V91fs197\*, which result in the generation of a premature stop codon.<sup>5</sup>

Electrophysiological studies in mammalian cells and *Xenopus* oocytes showed decreased potassium conductivity of *KCNJ10* channels with mutations associated with EAST syndrome.<sup>1,5-8,38,39,43</sup> All but one mutation, p.A167V, severely affect the function with less than 20% residual activity reported and with complete or near complete loss of function in p.R199\*, p.F75C, p.C140R, p.V91fs197\*, p.V259\*, p. T57I and p.T164I.<sup>5-7,13,38,39,43</sup> *KCNJ10* p.A167V mutations also reduce potassium conductance, however to a lesser extent, retaining 60% residual activity.<sup>6,38,39</sup> This level of activity is presumed to be present in unaffected siblings or parents of EAST patients who are heterozygous for this mutation and so initially it was thought that this mutation was not pathogenic in the homozygote state.<sup>38</sup> However homozygotes for this mutation (p.A167V) have been diagnosed with EAST syndrome and in our own analysis co-expression of this *KCNJ10* mutation with *KCNJ16*, mimicking the native channel in kidney showed a dramatic reduction in function, suggesting that the p.A167V mutation causes reduced heteromer activity.<sup>6</sup> A recent study questions this proposed pathogenic mechanism of p.A167V mutation as *KCNJ10* does not form heteromers in the brain or ear.<sup>13,41,44</sup> Therefore reduced *KCNJ10*/*KCNJ16* heteromeric activity may not be the sole pathogenic process caused by this mutation as it does not explain the non-renal symptoms seen in these EAST patients. The same study suggested experimentally that this p. A167V mutation causes the *KCNJ10* channels to not be anchored to the basolateral membrane of the renal epithelia and there is inhibition of expression. This mistrafficking and thus mislocalisation of *KCNJ10* may consequently cause loss of potassium recycling which is an essential function of the basolateral membrane of the distal nephron segment.<sup>13</sup> The divergent clinical findings in the 3 patients identified so far with homozygous A167V mutations reflect this controversy, as 2 patients show predominantly renal manifestations (i.e., consistent with the heteromer hypothesis), whereas the other patient shows the complete EAST phenotype, including the neurological

manifestations (i.e. consistent with the mistrafficking hypothesis).

## Conclusion

The constellation of epilepsy, ataxia, sensorineural deafness and salt wasting secondary to renal tubulopathy is known as EAST syndrome. This phenotype is consequent to mutations in the *KCNJ10* gene, which is located on chromosome 1 and has an autosomal recessive mode of inheritance. So far there have been 14 different *KCNJ10* mutations described in the literature and these result in reduced or total loss of function of the *KCNJ10* channel. Expression of *KCNJ10* correlates with the clinical and biochemical characteristics of EAST syndrome with *KCNJ10* potassium channels located in the brain, eye, ear and kidney. Inadequate potassium clearance by these channels in glial cells of the brain lowers seizure threshold and thus epilepsy is a hallmark of EAST syndrome. Neuroimaging in most cases revealed changes in their cerebellum highlighting cerebellar dysfunction, manifesting as ataxia, another clinical facet of *KCNJ10* mutations. Sensorineural hearing loss is the result of the failure to generate the endocochlear potential and high potassium concentration in the endolymph, a process vital for hearing. The renal salt-wasting seen in this condition is secondary to defective potassium recycling and basolateral membrane depolarization in the distal nephron leading to impaired transepithelial electrolyte transport.


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