Clinical Kidney Journal, 2021, vol. 14, no. 8, 1990–1993

doi: 10.1093/ckj/sfab083
Advance Access Publication Date: 28 April 2021
Letter to the Editor

LETTER TO THE EDITOR

Historical and geographical distribution of the founder mutation c.610G>A; p.Ala204Thr in the *CLCNKB* gene linked to Bartter syndrome type III in Spain

Ramón Peces [1,*], Rocío Mena[2], Carlos Peces[3], Pilar Barruz[2], Hernando Trujillo[4], Agustín Carreño[5], Laura Espinosa[6], Rafael Selgas[1], Pablo Lapunzina[2,7] and Julián Nevado[2,7,*]

[1]Servicio de Nefrología-Adultos, Hospital Universitario La Paz. IdiPAZ. Universidad Autónoma, Madrid, Spain, [2]Instituto de Genética Médica y Molecular (INGEMM), Hospital Universitario La Paz. IdiPAZ. Universidad Autónoma, Madrid, Spain, [3]Area de Tecnología de la Información, SESCAM, Toledo, Spain, [4]Servicio de Nefrología-Adultos, Hospital Universitario 12 de Octubre, Madrid, Spain, [5]Servicio de Nefrología, Hospital General Universitario de Ciudad Real, Ciudad Real, Spain, [6]Servicio de Nefrología-Infantil, Hospital Universitario La Paz. IdiPAZ. Universidad Autónoma, Madrid, Spain and [7]CIBERER, ITHACA (European Reference Network), Madrid, Spain

*These authors contributed equally to this work.
Correspondence to: Ramón Peces; E-mail: ramon.peces@salud.madrid.org

Bartter syndrome (BS) type III caused by variation within *CLCNKB* has a heterogeneous presentation, extending from severe to very mild, and is the most common in Spain [1–4]. The p. Ala204Thr variation is the most recurrent in our country and is only observed in Spanish descendants, suggesting a founder effect [1–4].

A cohort of 21 Spanish patients with a clinical history consistent with BS was analysed and 15 patients from different unrelated families were characterized as BS type III (Supplementary data, Table S1). Thirteen out of 15 (87%) are carrying the variant c.610G>A; p.Ala204Thr (Supplementary data, Figure S1) in homozygosis or as a part of a compound heterozygous. Characterization of the *CLCNKB* locus and its neighbourhood by SNP arrays showed a shared haplotype of seven single nucleotide polymorphisms (SNPs) (Figure 1) that co-segregates with this variant in the affected individuals, a fact that strongly reflects the presence of a founder mutation and excludes the alternative hypothesis of a hot spot. We noted that all the subjects' ancestry originated from an area of the centre of Spain, where all the localities were founded between 10th and 14th centuries (afterward repopulated as a result of *The Reconquest*), which are crossed by historical-pecuary routes (*Cañadas Reales*; Table 1 and Figure 2). For hundreds of years, a seasonal movement (North–South and vice versa) of people along these livestock roads took place (the transhumance), suggesting a regional founder effect of this variant. Remarkably, our cohort's grandparents were likely to have been born in the decades either side of 1920–30, so the spatial distribution of the genetic structure described in this study would reflect that the genetic structure seen within Spain around the beginning of the 20th century is a result of regional genetic isolation.

The cultural and linguistic impact of Muslim rule in the Iberian Peninsula is well-known but limited to the extent, timing and geographic spread of genetic mixing between immigrants and Iberians over several centuries after the initial Muslim conquest (711 CE) [5, 6]. During *The Reconquest*, Christian-controlled territory in the North moved gradually Southwards from the mid-8th century. The Castilian kings

Received: 25.1.2021; Editorial decision: 20.4.2021

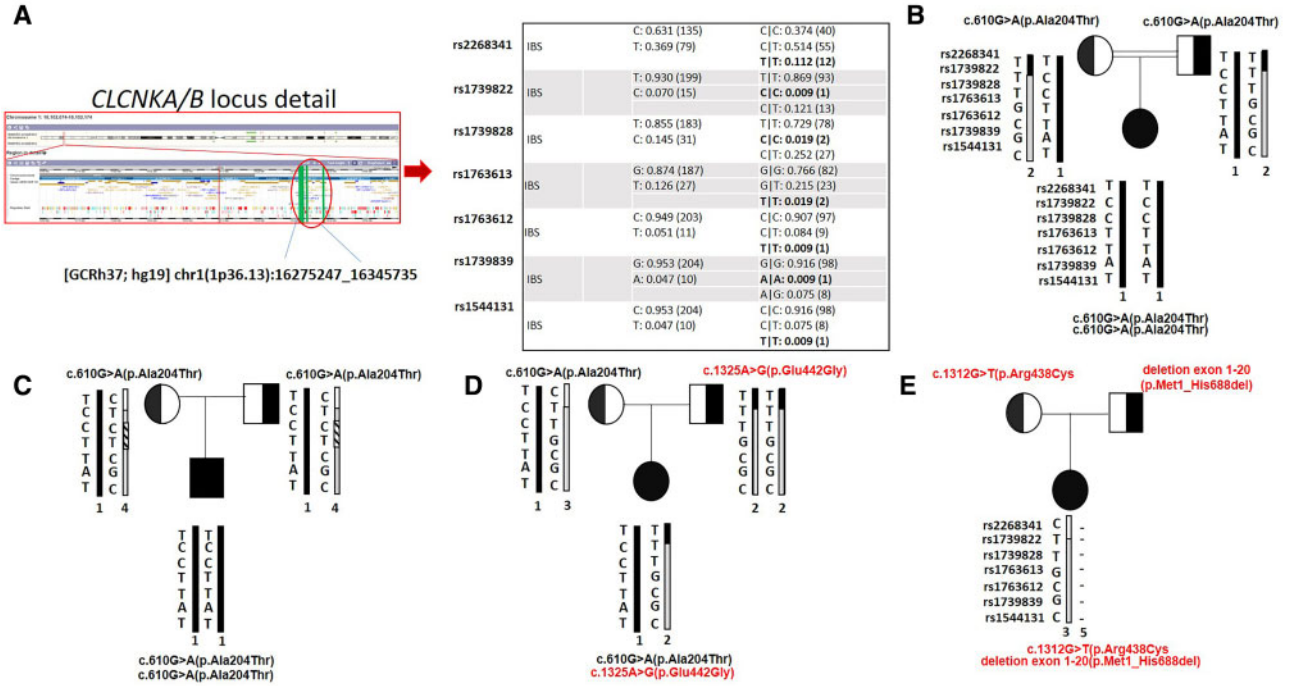

FIGURE 1: Haplotypes reconstruction (1 to 5) relative to seven markers (SNPs) regarding NM_000085.4: NM_000085.4:c.610G>A(p.Ala204Thr) mutation in the CLCNKB of three families type (as example). A) SNP markers selected and position into CLCNKA/B locus. From the top: rs2268341, rs1739822, rs1739828, rs1763613, rs1763612, rs1739839, rs1544131, and their poblational frequencies (source: gnomAD). B) Homozygous consanguineous family, C) Homozoygous non-consanguineous familiy, D) Compound heterozygous case, E) Compound heterozygous case without including c.610G>A.

Table 1. Genetic characteristics, geographical and historical ancestry origin of 13 patients with p.Ala204Thr

Case	Age/sex	Paternal mutation	Maternal mutation	Father ancestry origin/century *Cañada*	Mother ancestry origin/century *Cañada*
1	49/Male	p.Ala204Thr	p.Ala204Thr	San Pablo de los Montes (Toledo)/13th *Cañada Real Segoviana (4)*	San Pablo de los Montes (Toledo)/13th *Cañada Real Segoviana (4)*
2	26/Male	p.Ala204Thr	p.Ala204Thr	Cantalejo (Segovia)/10th *Cañada Real Soriana Occidental (6)*	Cuellar (Segovia)/11th *Cañada Real Leonesa Oriental (3)*
3	39/Male	p.Ala204Thr	p.Ala204Thr	El Casar de Escalona (Toledo)/12th *Cañada Real Leonesa Oriental (3)*	Pozuelo del Rey (Madrid)/12th *Cañada Real Galiana (7)*
4	40/Female	p.Ala204Thr	p.Ala204Thr	Urda (Toledo)/11th *Cañada Real Leonesa Oriental (3)*	Urda (Toledo)/11th *Cañada Real Leonesa Oriental (3)*
5	62/Female	p.Ala204Thr	p.Ala204Thr	Fuencarral (Madrid)/12th *Cañada Real Galiana (7)*	Medina del Campo (Valladolid)/11th *Cañada Real Leonesa Occidental (2)*
6	21/Female	p.Ala204Thr	p.Ala204Thr	Caleruega (Burgos)/10th *Cañada Real Segoviana (4)*	Caleruega (Burgos)/10th *Cañada Real Segoviana (4)*
7	39/Female	p.Ala204Thr	p.Met1_His654del	San Pedro de Gaillos (Segovia)/10th *Cañada Real Soriana Occidental (6)*	Ferrol (Galicia)
8	12/Male	p.Ala204Thr	p.Ala204Thr	Valdemanco (Madrid)/14th *Cañada Real Segoviana (4)*	Valdemanco (Madrid)/14th *Cañada Real Segoviana (4)*
9	11/Female	p.Glu442Gly	p.Ala204Thr	Madrid-Jaen	Madrid, Fuensalida (Toledo)/11th, 13th *Cañada Real Segoviana (4)* *Cañada Real Galiana (7)*
10	52/Female	p.Ala204Thr	p.Ala204Thr	Corral de Almaguer (Toledo)/13th *Cañada Real Soriana Oriental (5)*	Corral de Almaguer (Toledo)/13th *Cañada Real Soriana Oriental (5)*
11	37/Male	p.Ala204Thr	p.Ala204Thr	Almagro (Ciudad Real)/13th *Cañada Real Soriana Oriental (5)* *Cañada Real de Cuenca (8)*	Bolaños de Calatrava (Ciudad Real)/13th *Cañada Real Soriana Oriental (5)* *Cañada Real de Cuenca (8)*
12	26/Female	p.Ala204Thr	p.Ala204Thr	Guzmán (Burgos)/11th *Cañada Real Segoviana (4)*	Madrigalejo (Cáceres)/11th *Cañada Real de La Plata (1)* *Cañada Real Leonesa Occidental (2)*
13	57/Female	p.Leu439Pro	p.Ala204Thr	Mundaca (Bilbao)	Alcalá de Henares (Madrid)/11th *Cañada Real Galiana (7)*

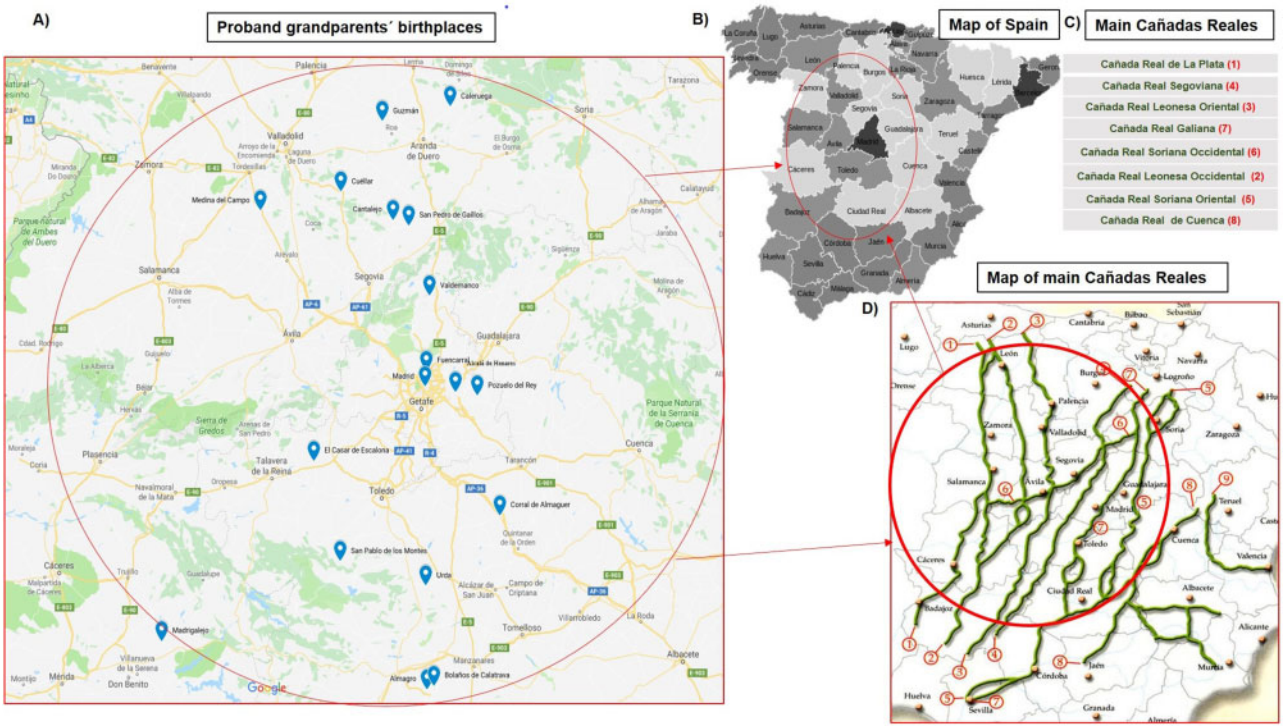

FIGURE 2: Historical-geographical relationships between Spanish type III Bartter syndrome patients studied (dot blue), who carries the NM_000085.4:c.610G>A (p.Ala204Thr) mutation in the CLCNKB gene and historical pathways (Cañadas Reales). Each individual is represented by a dot placed the centroid of their grandparents' birthplaces. The mutation can be related to a transhumance population established into or in the vicinities of pathways of historical importance (Cañadas Reales).

moved in with their armies but in what amounted to a veritable frontier movement, struggled to repopulate their new territories. Transhumance could thrive in this largely unpopulated landscape. The population movements were facilitated by the subsequent establishment of 'The Honourable Concejo de la Mesta' (year 1273 CE) by King Alfonso X (The Wise), regulating the migration of people through historical pathways (*Cañadas Reales*). Interestingly, the distribution of p.Ala204Thr in the BS population was strikingly very similar to that established for different populations and dialects presented around 1300 CE (Old Kingdom of Castile) [5, 6]. Since p.Ala204Thr is not found in populations from North Africa, Arab origin or Sephardic-Jewish ancestry, this mutation seems to have originated in the primitive population of Spain, who either remained unmixed with the Muslim invaders and Jewish people or originated after the onset of *The Reconquest* (which finished in 1492 CE) [5, 6], spreading with the re-population of the reconquered lands and establishment of the transhumance. In addition, the p.Ala204Thr variation is also present in Latin American individuals with Spanish ancestry, but exclusively in a heterozygous way (source: gnomAD). This fact, along with finding several heterozygous carriers (for p. Ala204Thr) in individuals with ancestry originated from Mestanza (a town founded in the 11th century, localized at the South of Ciudad Real) and a dot of confluence of *Cañada Real Segoviana* (4) and *Cañada Real Soriana Oriental* (5) (Figure 2), seemed to support our hypothesis. Our data are also consistent with studies establishing a strong association between specific-unique mutations and population founder effects on BS [7–9], such as in Korea and Japan [8, 9] or Costa Rican populations [10] (Supplementary data, Table S2).

Our findings support the p.Ala204Thr variant as a founder mutation originating from the central part of Spain, and it is conceivable that the greater concentration of individuals within this area was the consequence of an offspring from a common ancestor. Thus, the knowledge of the spectrum of mutations and their geographical distribution of this variant will allow us a more effective detection strategy in individuals with BS type III, or extend it to other countries with large Spanish populations.

SUPPLEMENTARY DATA

Supplementary data are available at ckj online.

ACKNOWLEDGEMENTS

The authors thank the families who contributed to this study. This study was conducted in accordance with declaration of Helsinki and all of its amendments, and national regulations.

PATIENT CONSENT

Written informed consents were obtained from the patients.

FUNDING

This study was supported in part by grants from the Research Activity Intensification Programme (IdiPAZ) to R.P. and J.N. ISCIII RETIC REDINREN RD16/0009.FEDER FUNDS.

CONFLICT OF INTEREST STATEMENT

All the authors declared no competing interests.

REFERENCES

1. Rodríguez Soriano J, Vallo A, Pérez de Nanclares G *et al.* A founder mutation in the *CLCNKB* gene causes Bartter syndrome type III in Spain. *Pediatr Nephrol* 2005; 20: 891–896
2. García Castaño A, Pérez de Nanclares G, Madariaga L *et al.* Genetics of type III Bartter syndrome in Spain, proposed diagnostic algorithm. *PLoS One* 2013; 8: e74673
3. Gorgojo JJ, Donnay S, Jeck N, Konrad M. A Spanish founder mutation in the chloride channel gene, CLCNKB, as a cause of atypical Bartter syndrome in adult age. *Horm Res* 2006; 65: 62–68
4. García Castaño A, Pérez de Nanclares G, Madariaga L *et al.* Poor phenotype-genotype association in a large series of patients with type III Bartter syndrome. *PLoS One* 2017; 12: e0173581
5. Bycroft C, Fernandez-Rozadilla C, Ruiz-Ponte C *et al.* Patterns of genetic differentiation and the footprints of historical migrations in the Iberian Peninsula. *Nat Commun* 2019; 10: 551
6. Adams SM, Bosch E, Balaresque PL *et al.* The genetic legacy of religious diversity and intolerance: paternal lineages of Christians, Jews, and Muslims in the Iberian Peninsula. *Am J Hum Genet* 2008; 83: 725–736
7. Lee BH, Cho HY, Lee H *et al.* Genetic basis of Bartter syndrome in Korea. *Nephrol Dial Transplant* 2012; 27: 1516–1521
8. Tajima T, Nawate M, Takahashi Y *et al.* Molecular analysis of the *CLCNKB* gene in Japanese patients with classic Bartter syndrome. *Endocr J* 2006; 53: 647–652
9. Nozu K, Fu XJ, Nakanishi K *et al.* Molecular analysis of patients with type III Bartter syndrome: picking up large heterozygous deletions with semiquantitative PCR. *Pediatr Res* 2007; 62: 364–369
10. Kurtz CL, Karolyi L, Seyberth HW *et al.* A common NKCC2 mutation in Costa Rican Bartter's syndrome patients: evidence for a founder effect. *J Am Soc Nephrol* 1997; 8: 1706–1711