Genetic analysis in a large cohort of patients with hereditary spastic paraplegia: diagnostic challenges

Sara Morais[1,2], Ana Filipa Brandão[1,2], Ana Lopes[1,2], Rita Bastos-Ferreira[1,2], Susana Sousa[1,2], Paulo Silva[1,2], Fátima Lopes[1,2], Alexandra Lopes[1,2], Joana Damásio[1,2,3], José Leal Loureiro[4], Marina Magalhães[3,5], Miguel Leão[6], Cristina Costa[7], Ricardo Maré[8], Jorge Sequeiros[1,2], João Freixo[1,2], Jorge Oliveira[1,2]

1- CGPP-IBMC – Centro de Genética Preditiva e Preventiva, Instituto de Biologia Molecular e Celular, Universidade do Porto, Portugal; 2- i3S – Instituto de Investigação e Inovação em Saúde, Universidade do Porto, Portugal; 3- Serviço de Neurologia, Hospital de Santo António, Centro Hospitalar Universitário do Porto, Portugal; 4- Serviço de Neurologia, Centro Hospitalar de Entre o Douro e Vouga, Santa Maria da Feira, Portugal; 5- Serviço de Neurologia, Hospital Padre Américo, Centro Hospitalar do Tâmega e Sousa, Portugal; 6- Serviço de Genética, Centro Hospitalar de São João/Faculdade de Medicina da Universidade do Porto, Portugal; 7- Serviço de Neurologia, Hospital Professor Doutor Fernando Fonseca, Amadora, Portugal; 8- Serviço de Neurologia, Hospital de Braga, Portugal

Background

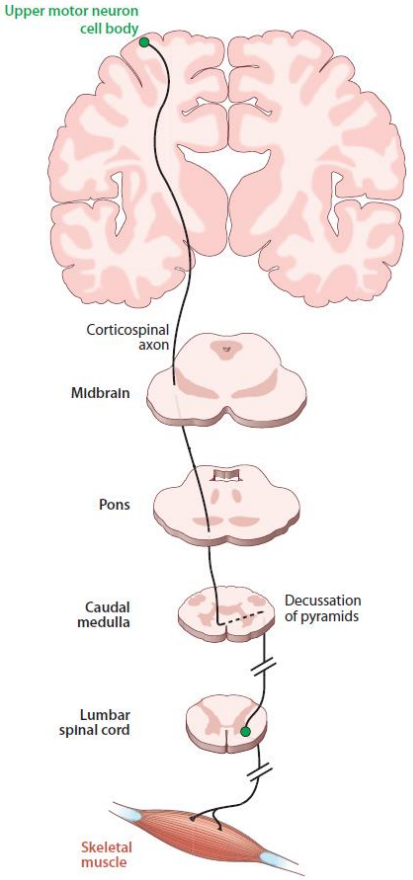
- **Hereditary Spastic Paraplegia (HSP):** group of rare neurodegenerative disorders
- Main symptoms:
  - Slowly progressive spasticity
  - Weakness of the lower limbs
  - May include additional neurologic and non-neurologic features
- Estimated prevalence in Europe: 3-10/100,000
- Pathologically characterized by axonal degeneration of the longest nerve fibers in the corticospinal tracts

Genetic aetiology

> 70 identified *loci*

still

40 - 70% of the families without a molecular diagnosis



Blackstone, C. (2012). Annu. Rev. Neurosci.

ESHG 2021 - Virtual Conference | June 12-15, 2021

Genetic analysis in a large cohort of patients with hereditary spastic paraplegia: diagnostic challenges

IBMC INSTITUTO DE BIOLOGIA MOLECULAR E CELULAR
INSTITUTE FOR MOLECULAR AND CELL BIOLOGY
CGPP CENTRO DE GENÉTICA PREDITIVA E PREVENTIVA
i3S INSTITUTO DE INVESTIGAÇÃO E INOVAÇÃO EM SAÚDE
UNIVERSIDADE DO PORTO

Aim

Molecular diagnosis of 472 HSP patients (index cases)

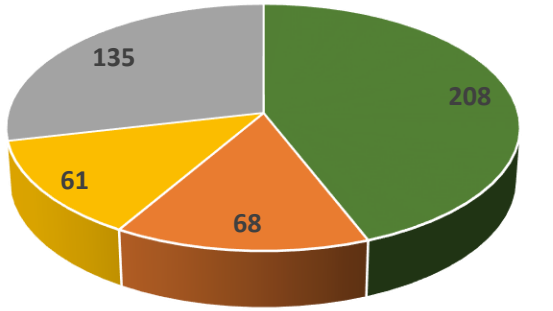
Strategy

Single-gene tests	Whole-exome sequencing based multigene panels

Results

118 patients (with molecular diagnosis)	90 patients (with molecular diagnosis)
7 patients (with variants of unknown clinical significance (VUS))	61 patients (with variants of unknown clinical significance (VUS))
11 patients (with single heterozygous variants in AR-associated genes)	50 patients (with single heterozygous variants in AR-associated genes)

Total patients (n=472)



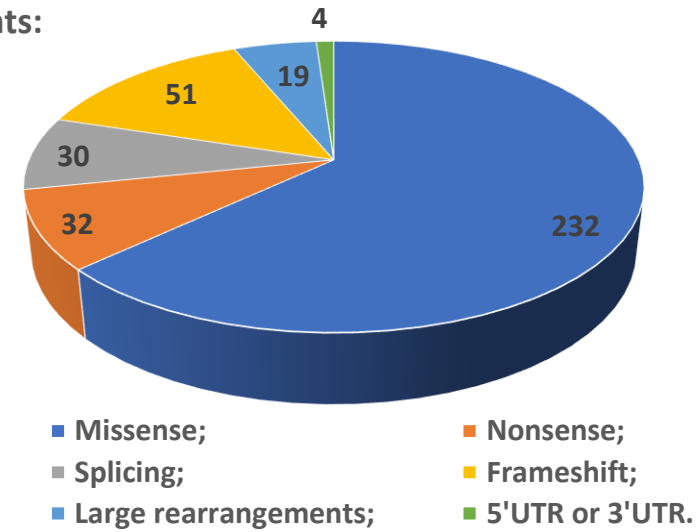
Genetic analysis in a large cohort of patients with hereditary spastic paraplegia: diagnostic challenges

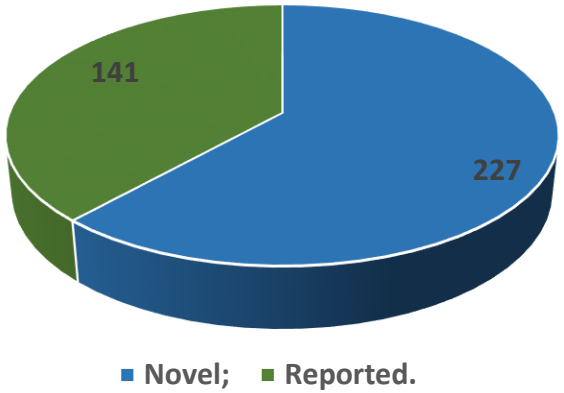
IBMC INSTITUTO DE BIOLOGIA MOLECULAR E CELULAR INSTITUTE FOR MOLECULAR AND CELL BIOLOGY
CGPP CENTRO DE GENÉTICA PREDITIVA E PREVENTIVA
i3S INSTITUTO DE INVESTIGAÇÃO E INOVAÇÃO EM SAÚDE UNIVERSIDADE DO PORTO

Results

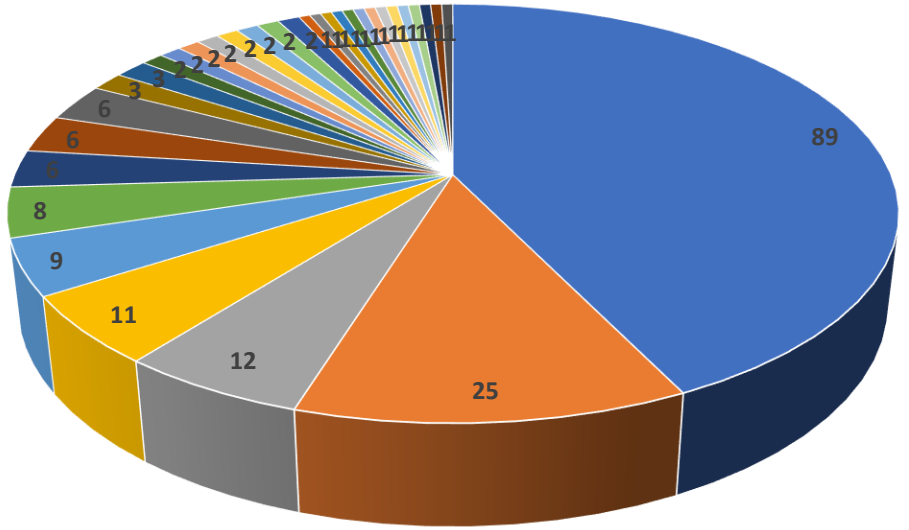
Identification of 368 different variants in 87 genes

Type of variants:





Disease-causing variants identified in 33 genes



SPAST (42.8%); SPG11 (12%); SPG7 (5.8%); ZFYVE26 (5.3%); ATL1 (4.3%);
CYP7B1 (3.8%) CAPN1 (2.9%); KIF5A (2.9%); SACS (2.9%); ARG1 (1.4%);
ATP13A2 (1.4%); B4GALNT1 (1%); BSCL2 (1%); CYP27A1 (1%); KCNA2 (1%);
KIF1A (1%); REEP1 (1%); REEP2 (1%); RNASEH2B (1%); ABCD1 (0.5%);
ALS2 (0.5%); CYP2U1 (0.5%); DDHD1 (0.5%); FA2H (0.5%); GALC (0.5%);
GBA2 (0.5%); GJC2 (0.5%); NFU1 (0.5%); NIPA1 (0.5%); PLP1 (0.5%);
SLC2A1 (0.5%); SYNE1 (0.5%); TUBB4A (0.5%).

ESHG 2021 - Virtual Conference | June 12-15, 2021

Genetic analysis in a large cohort of patients with hereditary spastic paraplegia: diagnostic challenges

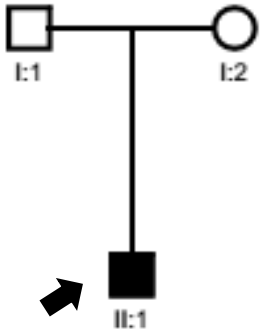
IBMC INSTITUTO DE BIOLOGIA MOLECULAR E CELULAR INSTITUTE FOR MOLECULAR AND CELL BIOLOGY
CGPP CENTRO DE GENÉTICA PREDITIVA E PREVENTIVA
i3S INSTITUTO DE INVESTIGAÇÃO E INOVAÇÃO EM SAÚDE UNIVERSIDADE DO PORTO

Results

Interesting case reports

Case #1

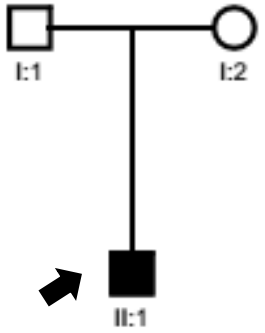
1 year-old male with pure spastic paraplegia. Normal brain MRI. Spastic hypertonia of the lower limbs.



homozygous variant in *ALS2*
NM_020919.3:c.[4261C>T];[c.4261C>T] // p.(Arg1421*)

Case #2

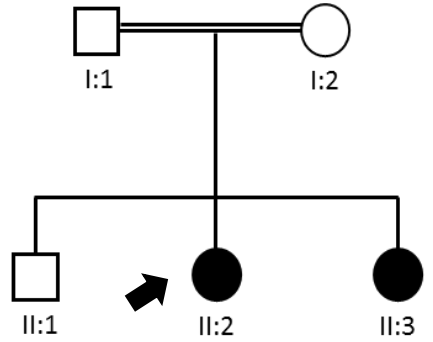
58 year-old male with spastic paraplegia.



homozygous variant in *GALC*
NM_000153.3:c.[927A>C];[927A>C] // p.(Leu309Phe)

Case #3

9 year-old female with spastic paraglegia. Polyneuropathy.



homozygous variant in *SYNE1*
NM_182961.3(SYNE1):c.[21401del]; [21401del] // p.(Ser7134Ilefs*9)

Identification of variants in **genes causing overlapping phenotypes** in patients with an apparent pure spastic paraplegia phenotype

Genetic analysis in a large cohort of patients with hereditary spastic paraplegia: diagnostic challenges

IBMC INSTITUTO DE BIOLOGIA MOLECULAR E CELULAR INSTITUTE FOR MOLECULAR AND CELL BIOLOGY
CGPP CENTRO DE GENÉTICA PREDITIVA E PREVENTIVA
i3S INSTITUTO DE INVESTIGAÇÃO E INOVAÇÃO EM SAÚDE UNIVERSIDADE DO PORTO

Conclusions

This work:

1. Contributed to the molecular, clinical and genetic heterogeneity of HSPs.

2. Demonstrates the importance of **testing genes defective in other differential diagnosis**, with overlapping clinical features (such as *SACS*, *ARG1* and *KCNA2*) – strong implications in NGS panels design.

3. Depicted a **high number of patients with VUS and/or single heterozygous variants** in genes associated to diseases with an autosomal recessive inheritance - besides the importance of identifying a missing pathogenic allele, highlighted:
   - the difficulty of interpreting VUS;
   - the importance of functional studies to clarify their causative role and, ultimately, provide a definitive diagnosis of HSP;
   - the possibility of finding, among these variants/*locus*, further modes of genetic inheritance, even of oligogenic nature.

4. The number of patients without variants reported (n=135) might indicate that a large number of HSPs causing genes and probably **novel pathogenic mechanisms** leading to the disease are yet to be discovered.

References

- Boutry et al. 2019. Curr Neurol Neurosci Rep. 19(4):18.
- Salinas et al. 2008. Lancet Neurol. 7(12):1127-38.
- Morais et al. 2017. Eur J Hum Genet. 25(11):1217-1228.
- Tesson et al. 2015. Hum. Genet. 134, 511–538.