Nuove metodologie di analisi genetica.
Successi e problematiche.

A.I.Vi.P.S. 6 Ottobre 2018
Sassari

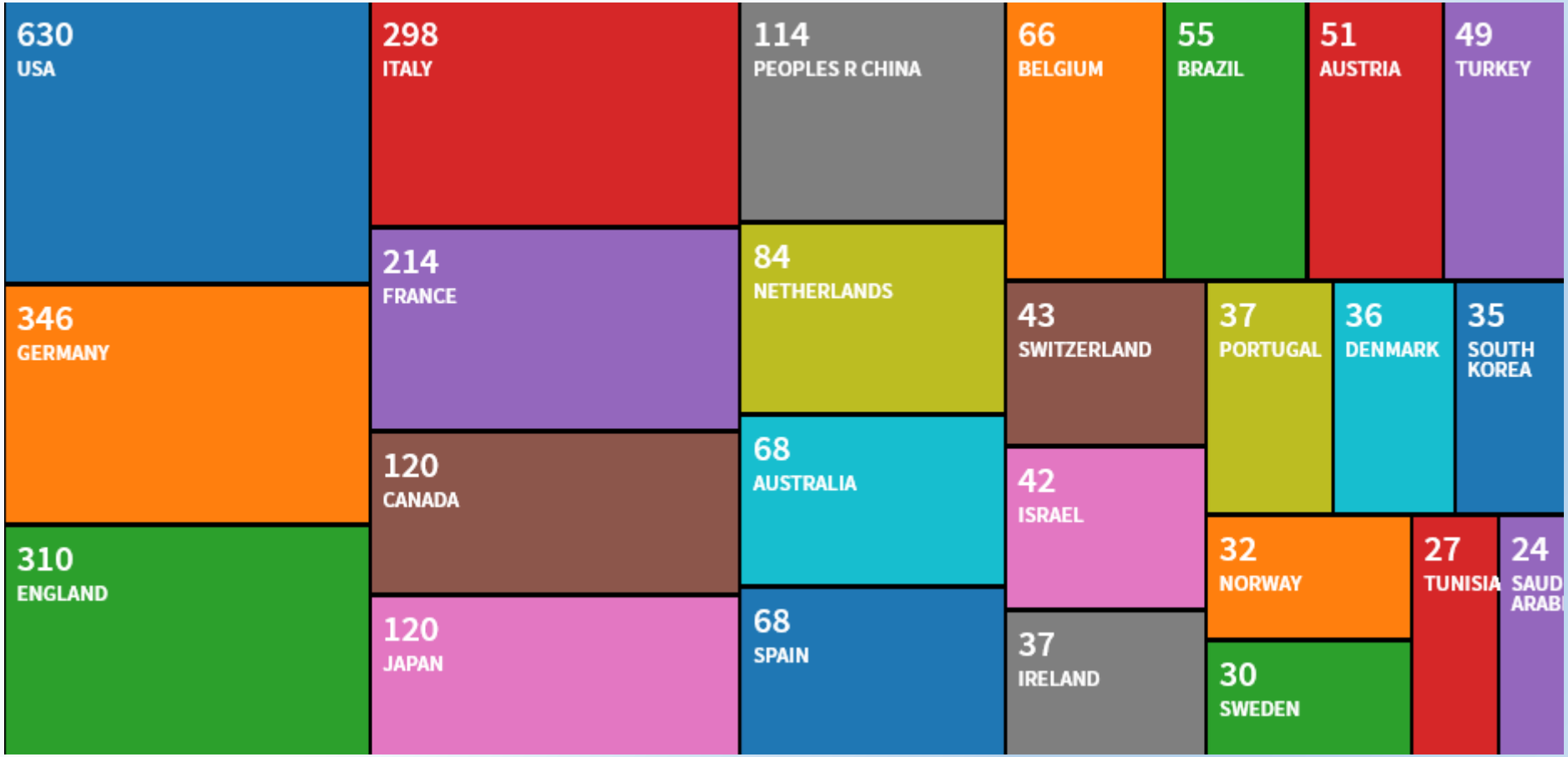
Giorgio Casari

FONDAZIONE Telethon

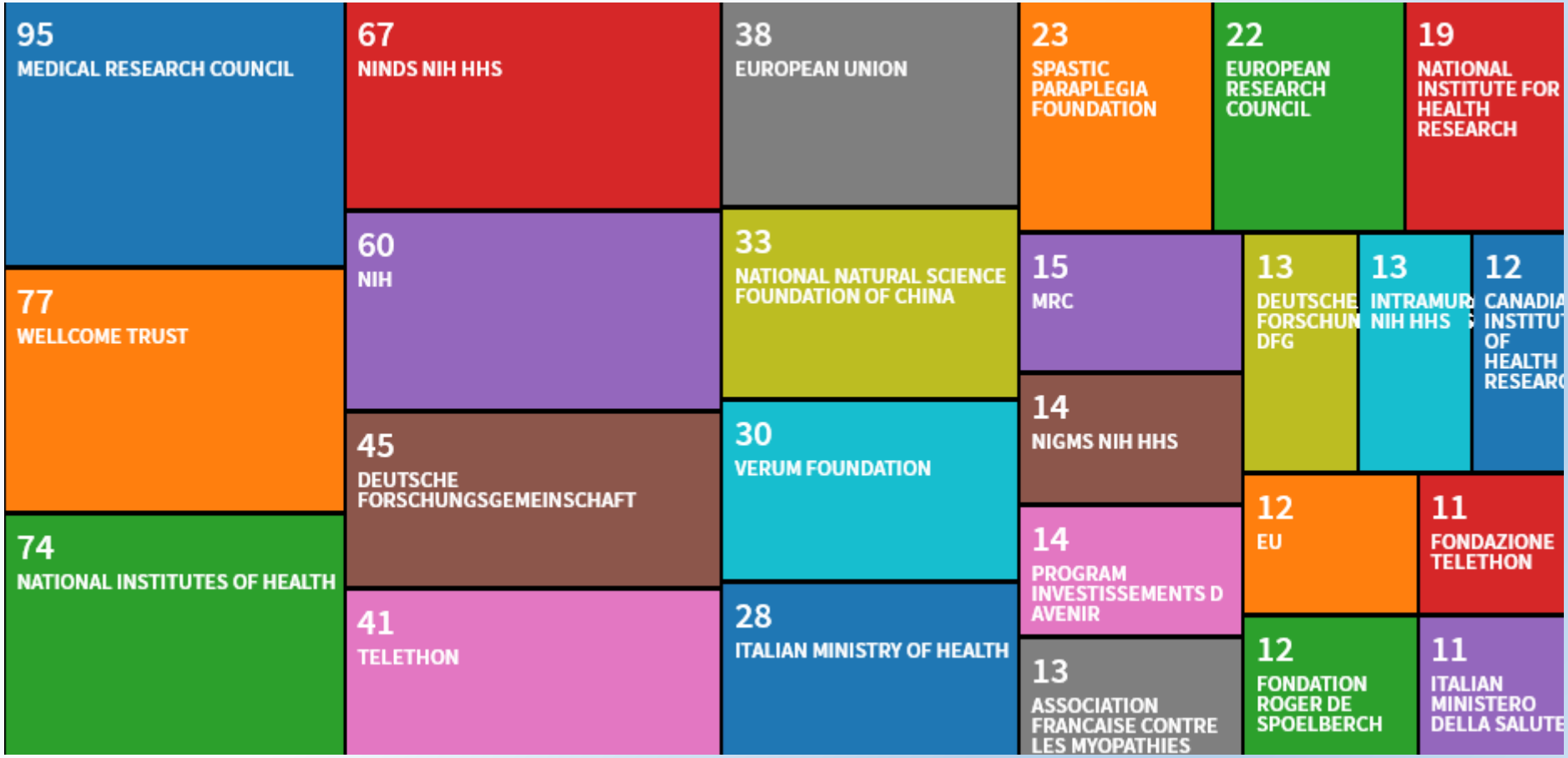
Number of papers on Hereditary Spastic Paraplegia

Science categories

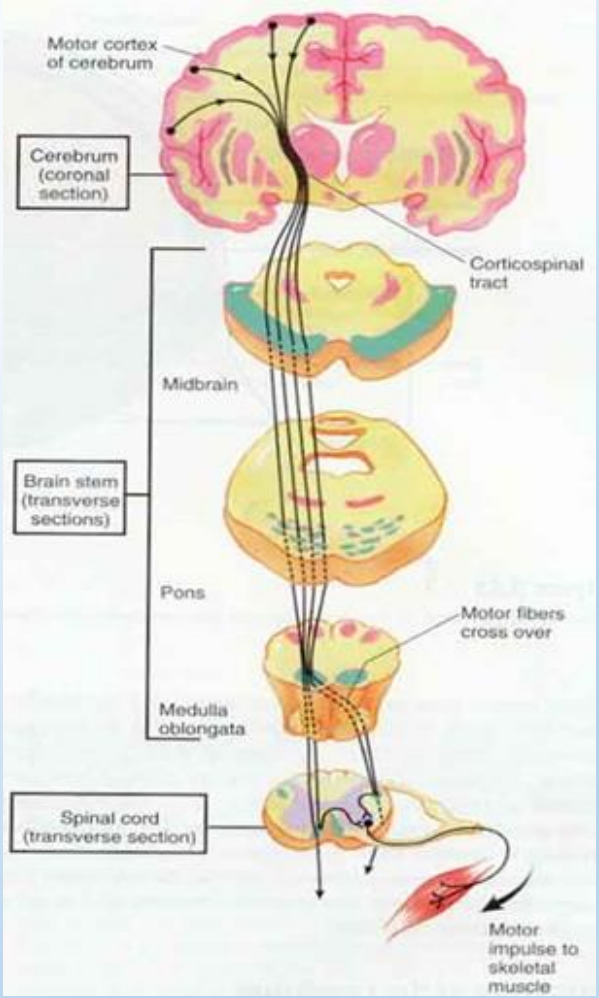
Countries studying HSP



Funding agencies

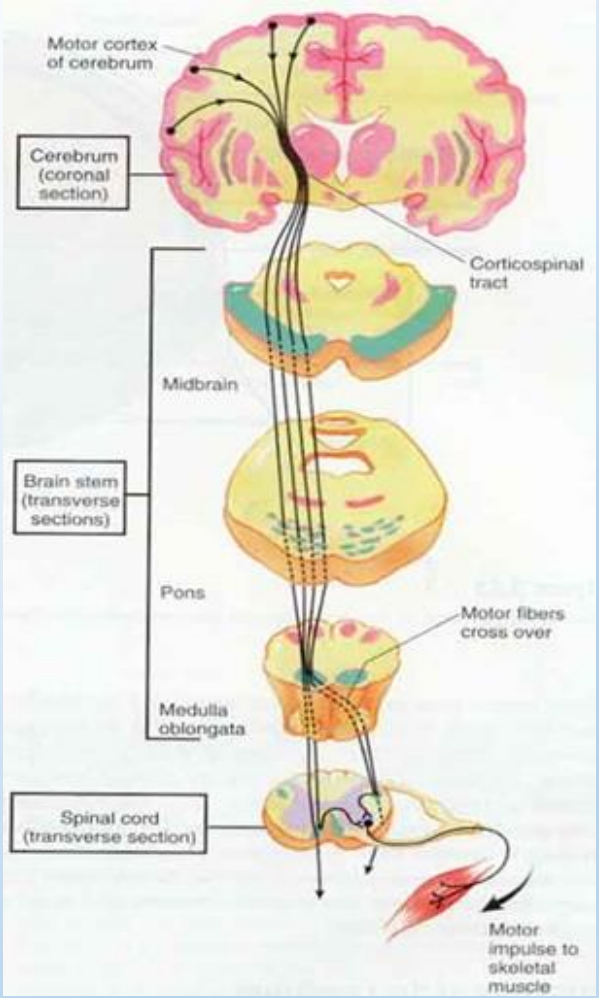


Function of HSP genes



Function of HSP genes

- ER morphogenesis
- Microtubule dynamics and transport
- Mitochondria
- Lipid metabolism
- Endosomal/ lysosomal



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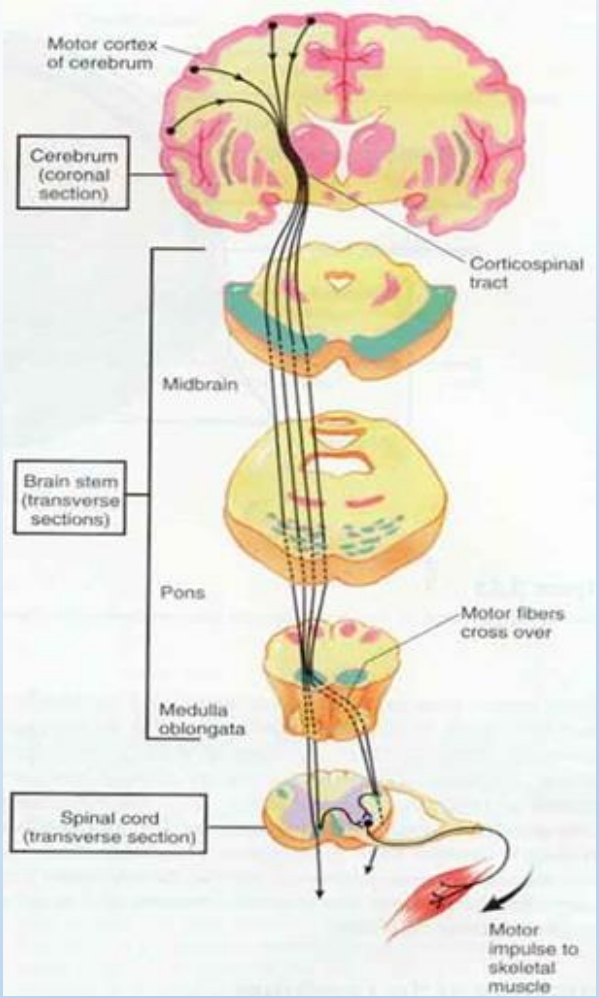
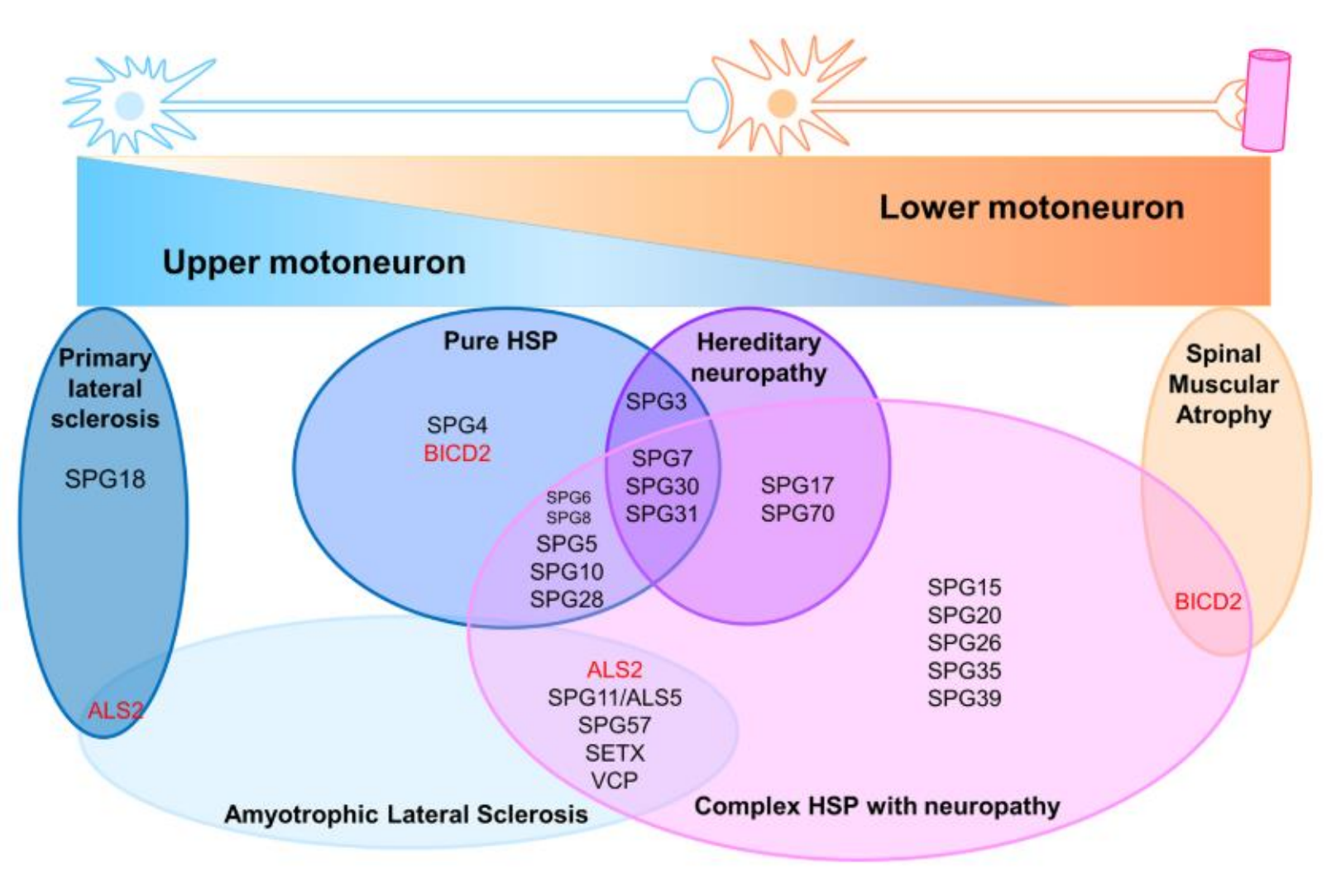


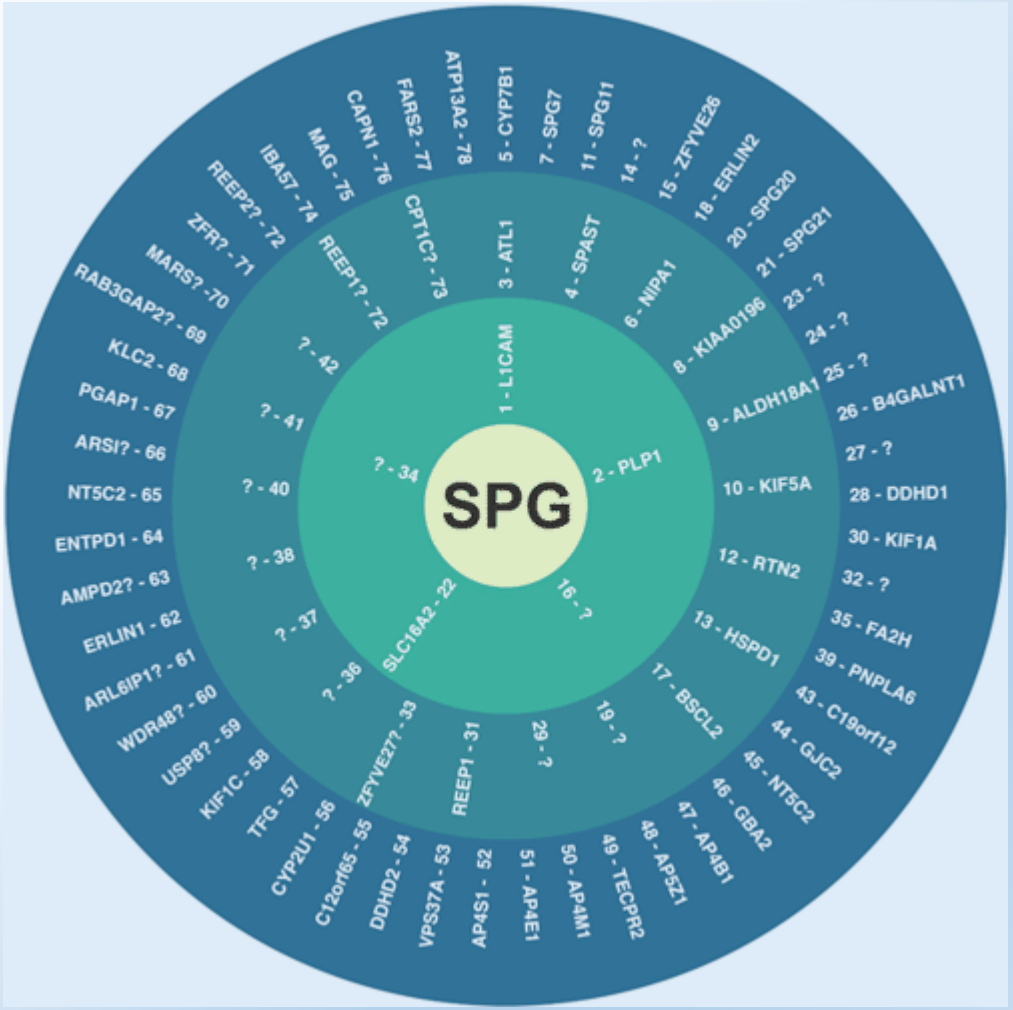
⇨ impaired cellular trafficking

Overlapping phenotypes originate from mutant HSP genes



Tesson et al., (2015) *Human Genetics*, *134*(6), 511–38.

High genetic heterogeneity of HSP



Schüle and Schöls, (2017) Nervenarzt 88:720-727

615491

SPASTIC PARAPLEGIA 79, AUTOSOMAL RECESSIVE; SPG79

Alternative titles; symbols

NEURODEGENERATION WITH OPTIC ATROPHY, CHILDHOOD-ONSET; NDGOA

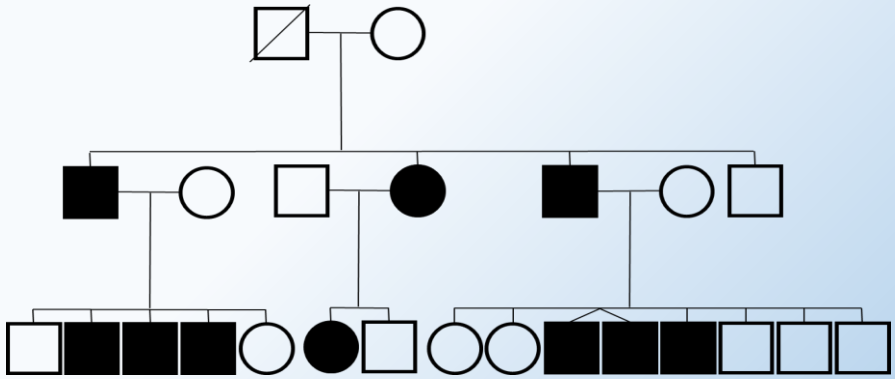
Phenotype-Gene Relationships

Location	Phenotype	Phenotype MIM number	Inheritance	Phenotype mapping key	Gene/Locus	Gene/Locus MIM number
4p13	Spastic paraplegia 79, autosomal recessive	615491	AR	3	UCHL1	191342

Molecular diagnosis of HSP

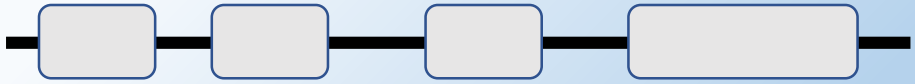
Molecular diagnosis of HSP

- Positional cloning



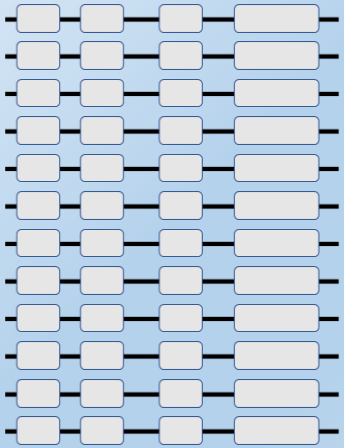
Molecular diagnosis of HSP

- Positional cloning
- Single gene testing



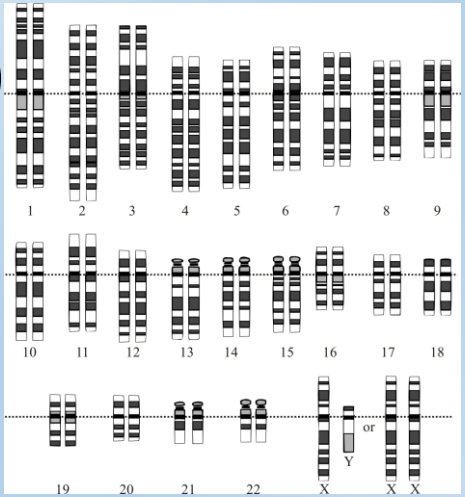
Molecular diagnosis of HSP

- Positional cloning
- Single gene testing
- HSP gene panels (80 and more genes)

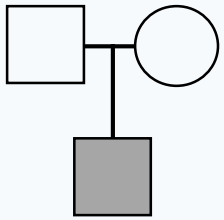


Molecular diagnosis of HSP

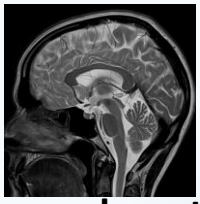
- ❑Positional cloning
- ❑Single gene testing
- ❑HSP gene panels (80 and more genes)
- ❑NGS: whole exome sequencing (WES) and whole genome seq (WGS)



The Telethon Undiagnosed Diseases Program
TUDP
Giving a name to the unknown/undiagnosed



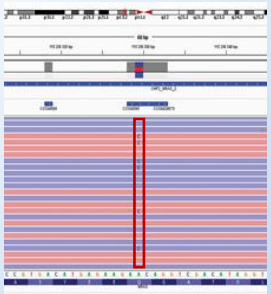
pediatric undiagnosed case



- **deep phenotyping**
- **exclusion of known genes**
- **negative CGH**

selection and prioritization trios/quartet families

very high-coverage whole exome sequencing



variant filtering and weighting functional effect

world-wide phenotype matching

The Telethon Undiagnosed Diseases Program
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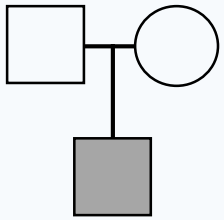
return diagnosis and counselling



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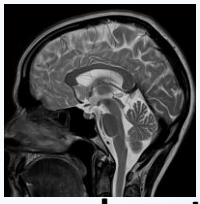

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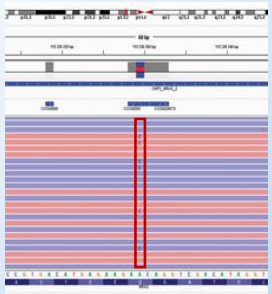


variant filtering and weighting functional effect

world-wide phenotype matching

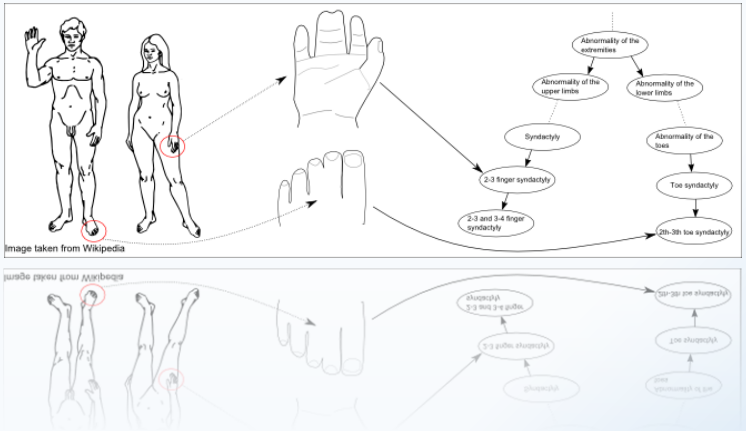
High coverage Whole Exome Sequencing

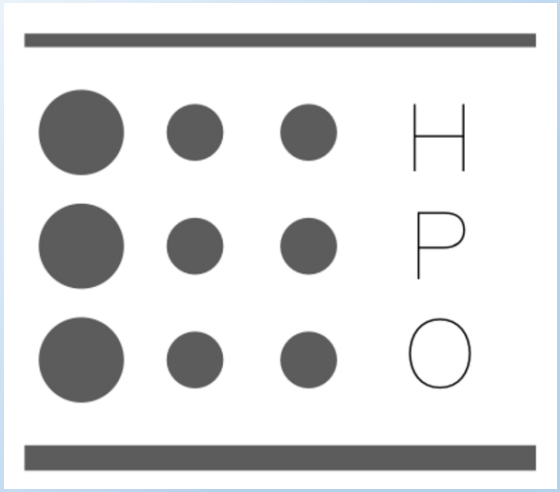
The pediatrics and clinical genetics TUDP network

Case phenotypes are defined by standardized definitions and glossary

Human Phenotype Ontology

http://human-phenotype-ontology.github.io/





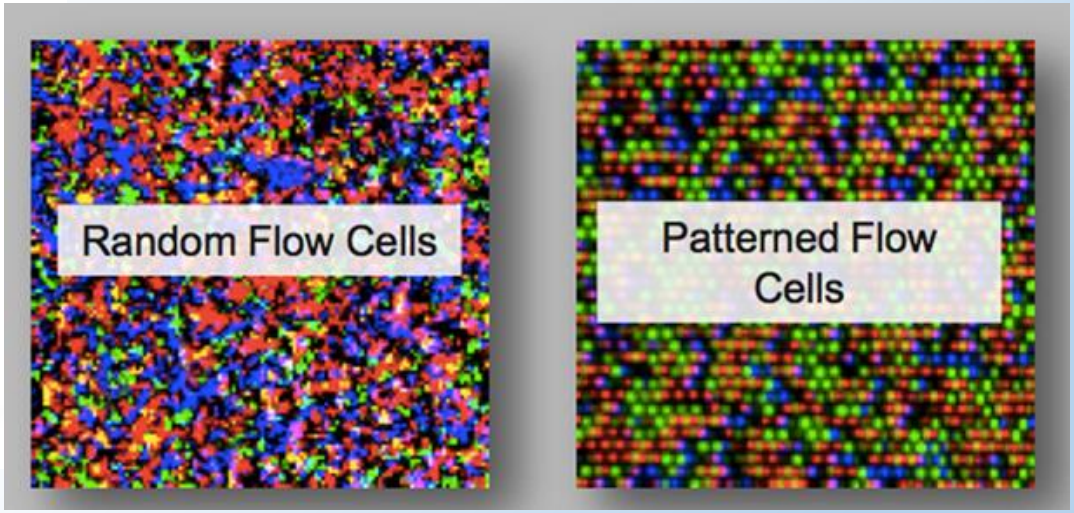
- PhenomeCentral, a repository for **secure data sharing** targeted at clinicians and scientists working in the rare disorder community.
- The PhenomeCentral collaboration model allows to **learn about the existence of similar cases** world-wide, which helps to improve understanding of disorder manifestations and confirm underlying causes.
- The **matching** algorithms -the semantic model- are **sensitive to atypical phenotypic manifestations** of disorders

https://www.phenomecentral.org/

Genomics analysis phases

- DNA extraction and library preparation
- Sequencing
- Bionformatics analysis
- Reporting genomics data
- Proritizating and selecting variants for final discussion
- Diagnosing
- Functional validation of new variants/mutations (and VUS)

High sequencing power



NovaSeq6000

6,000,000,000,000 bp/40 hours

= 40 seconds/ billion DNA bp

7$/ billion DNA bp

Four different flow cells S1-S4

What to do with new gene mutations or gene variants with unknown significance (VUS)?

- ❑ Find additional unrelated mutant patients
- ❑ Cellular models
- ❑ Animal models

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