**Introduction:** CSNB2 is a non-progressive retinal disorder with clinical features that include reduced visual acuity, nystagmus and variable myopia or hypermetropia. Characteristic abnormalities are detected upon electroretinography, autofluorescence and infrared imaging. The results of these tests indicate that vision impairment in CSNB2 patients may derive from a decreased synaptic transmission between photoreceptor and second-order neurons. The Cav1.4 channel is involved in this process and CACNA1F gene encodes the pore-forming subunit α1. Since CACNA1F is located on the X chromosome, Cav1.4 channelopathies are typically affecting male patients.

**Materials and Methods:** We performed NGS of 32 known retinal disease genes on DNA of 3 male patients with CSNB2 and different symptoms of the disease. In two patients CACNA1F mRNA was also studied in peripheral lymphocytes.

**Results:** We have identified 3 unreported CACNA1F variants : in exon 4 c.425dupC, in exon 43 c.G5123C and in exon 48 c. 5800delG. The first variant causes insertion of a stop codon that could determine mRNA degradation by Nonsense Mediated Decay (NMD). However, mRNA evaluation revealed that the transcript was present. The second variant is located in a splicing site and skipping of exon 43 was demonstrated by mRNA sequencing. The third variant determinates a frameshift. In this patient mRNA was not available.

**Conclusions:** These results confirm that, in CSNB2 patients, variants in the same gene may be associated with different phenotypic characteristics. Moreover, they highlight the importance of studying gene expression and protein function to assess the biological significance of the variants detected.

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### P02.13D

### Large-scale molecular analysis of Hereditary Hearing Loss genes in Argentinean deaf patients: lookingfora needle in a haystack

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Hereditary Hearing Loss (HHL) is a common trait affecting 1 in 2000 new born children. The presence of over 100 different genes involved in HHL, lead us to go on board with Whole Exome Sequencing (WES) in order to search for the causative mutations. The main objective of this project was to diagnose Argentinean deaf families and discover novel mutations or new genes involved in pathology. We designed a flowchart to exclude all the spurious variations obtained and target for few candidates. To approach this, we filtered results from WES, and candidate variations were segregated throughout family members. Variations positively selected, were analyzed using bioinformatic predictors and tracked in public databases. Additionally, conservation studies, structure and functional domain analysis in proteins, and in-vivo studies were performed. Using this strategy we analysed 15 WES results. We identified 16 causative mutations in 12 families with syndromic and non-syndromic hearing loss (11 missense, 4 frameshift and 1 splicing site mutations). Six were novel and functional studies of some of the identified mutations, using Zebra fish models, are under way. In the remaining 3 families, variables of uncertain significance were detected (Vous). To our knowledge this is the first study using WES to diagnose deaf patients in Argentina. We show in the present study that our flowchart is advantageous and noteworthy for large-scale molecular analysis in deaf patients. These findings clearly highlight the importance of genetic studies followed by in-sílico and in-vivo validation to better understand the genetic basis of Hereditary Hearing loss.

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### P02.14A

### Personalized stem cell therapy to correct corneal defects due to a unique homozygous-heterozygous mosaicism of Ectrodactyly-Ectodermal dysplasia-Clefting syndrome

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**Introduction:** Ectrodactyly-Ectodermal dysplasia-Clefting syndrome is a rare autosomal dominant disease caused by mutations in the p63 gene. To date, approximately 40 different p63 mutations have been identified, all heterozygous. No definitive treatments are available to counteract and