P190 BORTEZOMIB DESENSITIZATION EFFICACY IN PEDIATRIC CARDIAC TRANSPLANTATION: A SINGLE CENTER STUDY

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Aim: Bortezomib (BTZ) is a short-lived proteasome inhibitor, having the ability to lyse plasma cells responsible for the production of antibodies (Ab). It has been reported to have inconsistent success when used as a desensitization treatment for the reduction of HLA Ab in the context of pre and post solid organ transplantation. Furthermore, a variety of undesirable toxic side effects associated with this drug have been reported. In this work we assessed the efficacy of desensitization treatment using BTZ in a small pediatric cardiac transplant cohort.

Methods: Pre, during and post BTZ desensitization treatment sera were screened for HLA Ab using One Lambda LabScreen Single Antigen Beads (SAB) according to manufacturer's instructions. SAB was performed with neat sera and dilutions in PBS. Analysis was performed using Fusion v.3.0 software.

Results: 10 cardiac transplant patients, 2 pre and 8 post, were monitored for BTZ desensitization efficacy by assessing HLA Ab surrounding the time of treatment. For the two pre-transplanted patients no significant change in HLA Ab profile was observed before and after treatment. Among the 8 post- transplanted patients analyzed, 4 demonstrated an increase in several HLA Ab for both Class I and II, whether DSA or not, while no significant change in HLA Ab profile was noted in 3 of these patients. One post-transplanted patient had a single DSA positive DQ7 demonstrating 11,000 MFI pre-treatment which decreased to become negative after BTZ treatment. Overall, BTZ treatment did not appear successful in lowering the level of HLA Ab in 9 out of the 10 patients.

Conclusions: BTZ treatment does not appear to have significant impact on long or short term reduction of HLA Class I or II Ab in this pediatric transplant patient cohort. Further studies need to be performed to determine the ability of BTZ to reduce HLA Ab in transplant patients. Moreover, these studies will further demonstrate or challenge the proposed mechanisms of action described for this drug and perhaps will help to discover currently unidentified mechanisms by which it exerts its actions.

P191 A FIRST EVALUATION OF THE *HLA-A* GENE 3′ UNTRANSLATED REGION DIVERSITY IN BRAZIL AND ITS RELATIONSHIP WITH CODING HAPLOTYPES

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Aim: Here we propose a NGS strategy to evaluate the *HLA-A* coding and 3′UTR variability and get reliable haplotypes. Then, we present the data obtained in very admixed population (Brazil).

Methods: The *HLA-A*gene segment was amplified in a unique amplicon considering 408 Brazilian individuals. Amplicons were sequenced by NGS and data processed using *hla-mapper* and a local pipeline to get genotypes and haplotypes. Allele and haplotype frequencies, together with Linkage Disequilibrium (LD) plots, were also calculated.

Results: According to the human genome draft version hg38, and known *HLA-A* transcripts available, the *HLA-A* 3′UTR comprises a segment of 433 nucleotides. However, only 70 percent of this segment is considered at the IPD-IMGT/HLA database. A total of 21 variable sites were found in the 3′UTR and, together, they were organized in 11 different haplotypes. Seven of those variants were detected outside the segment tracked by the IMGT/HLA database. We observed a high LD throughout the *HLA-A* locus and associations between each of these 3′UTR haplotypes and specific coding alleles. Thus, in many cases, it is possible to detect the *HLA-A* allele group by targeting one or two variations at the 3′UTR. Although few haplotypes were detected, they were quite frequent. It is possible that the variability detected at this segment would influence allelic expression, mainly by posttranscriptional mechanisms through microRNA binding.

Conclusions: The *HLA-A* locus is one of the most variable genes on the human genome. Most of its variability was described within exons 2 and 3, which encode the peptide-binding groove. Thus, these segment are usually the only ones targeted by many *HLA-A* studies. Little is known regarding the *HLA-A*regulatory segments variability, including its 3′ untranslated region (3′UTR). Thus, *HLA-A* 3′UTR present few but frequent haplotypes, and each of these haplotype is associated with a specific group of coding alleles, which in turn may present a similar post transcriptional profile.