identification of somatic mutations in patients with anca-associated vasculitis

Keywords: Vasculitis, Genetics/Epigenetics

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Background: Growing evidence reveals a pathological role of somatic mutations in various autoimmune diseases, such as the mutation in UBA1 in VEXAS syndrome, CARD11 and KLHL6 in cryoglobulinemic vasculitis, or STAT3 in Felt’s syndrome. Somatic mutations might also be involved in the pathogenesis of ANCA-associated vasculitis (AAV), which typically manifests in middle-aged and elderly individuals.

Objectives: We aimed to identify somatic mutations in patients with AAV.

Methods: We collected whole blood and obtained peripheral blood mononuclear cells (PBMCs) and neutrophils from patients with AAV in active-disease status (n=16), as well as from patients with other autoimmune diseases (n=8) as disease controls, and healthy subjects (n=10). In addition, we collected these specimens from 12 out of the 16 patients with AAV after remission induction. We performed RNA sequencing (RNA-seq) on the obtained cells and whole genome sequencing (WGS) on DNA extracted from the whole blood. Somatic mutations were detected by comparing the RNA and DNA sequences [2].

Results: After stringent quality control, we identified 108 somatic mutations across 16 patients in active-disease status. The mean coverage of RNA-Seq at the mutation site was 100.9 ± 367.8 x, and that of WGS was 14.4 ± 4.3 x, while the mean allele fraction was 22.9 ± 20.5%. One or more mutations were detected in each of the 15 (93.8%) patients. The median mutation count of each patient was 4.0, which was not significantly different from disease controls or samples after remission induction. We mapped one gene to each of the 108 mutations, resulting in 95 genes in total. Mutations for six of the 95 genes were observed in two or more patients, and two of them were related to the ubiquitin system. Of the 108 mutations, 37 were missense, and 20 were predicted to be deleterious (combined annotation-dependent depletion Phred score > 20). Among the 20 mutations, the HST2H2AC mutation (NM_003517; p.L86P) in neutrophils was observed in two patients. To evaluate the functional outcome of the 20 mutations, we analysed the data on knocking out the corresponding genes using CRISPR in K562 cells [3]. The results showed that the expression of genes related to antigen presentation was increased in HEATR1 and RPL18 knockouts. When we followed up with 12 of the 16 patients after remission induction, 81.4% of the somatic mutations were no longer detected. Additionally, 91.7% of the deleterious mutations disappeared.

Conclusion: We found somatic mutations with potential pathological effects in some patients with AAV exhibiting active-disease status. Notably, the majority of these mutations were not detected after remission induction.

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evaluation of fc gamma receptor (fcr/g) cyp genetic polymorphism and its pharmacogenomic correlation with rituximab and cyclophosphamide response in anti-neutrophil cytoplasmic antibody associated vasculitis

Keywords: Remission, Vasculitis, Genetics/Epigenetics

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Background: The antineutrophil cytoplasmatic antibody (ANCA)-associated vasculitides (AAV) are three discrete entities – Granulomatosis with polyangiitis (GPA), Microscopic polyangiitis (MPA), and Eosinophilic granulomatosis with polymyalgia (EGPA). They present with a plethora of signs and symptoms resulting in significant burden of mortality and morbidity. There is huge lacuna in the knowledge of relationship between various genes affecting pharmacogenetics of AAV and its implications in context of therapeutic complications. We thus intend to evaluate genotypic polymorphism in FcγRI family & CYP genes affecting pharmac-molecular response of Rituximab and Cyclophosphamide respectively in AAV patients undergoing induction treatment.

Objectives: To study the single nucleotide polymorphism (SNP) of the FcγRI family (FCγRIIA, FCγRIIB, FCγRIIB) in patients receiving Rituximab (RTX) and the SNPs of CYP2C19*2 (rs4244285), CYP2B6 (rs321137) polymorphisms in patients receiving Cyclophosphamide (CYC) using polymerase chain reaction. Additionally, to correlate the genetic polymorphism of AAV patients with Rituximab and Cyclophosphamide response.

Methods: This study was a prospective cohort study in which AAV patients undergoing induction treatment were enrolled from Jan 2021 to Dec 2022 in Clinical Rheumatology Department at PGIMER Chandigarh, India. Functional SNPs for FCγRIIA, FCγRIIB 695T>C, FCγRIIL 557T>G and FCγRIIB 519G>A and...