Use of whole genome sequencing to identify highly recurrent somatic mutations in Waldenström’s macroglobulinemia.

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Abstract

Background

Waldenström’s Macroglobulinemia (WM) is an IgM secreting lymphoplasmacytoma. The genetic basis for this disease remains to be clarified.

Methods

We performed whole genome sequencing (WGS) using CD19+ selected bone marrow lymphoplasmacytoma cells (LPC) from 30 WM patients. For 10 of these patients, paired CD19+ depleted peripheral blood samples were used for WGS as normal controls. Prior to validation, somatic and germline classifications were determined by the data from the 10 paired samples. Affected genes observed only in the 20 unpaired samples are listed as unclassified.

Results

The most common somatic variants identified and validated by Sanger sequencing included MYD88 L265P, the c-terminal domain of CXCR4, and ARID1A.

A somatic T/C mutation in MYD88 resulting in a L252P substitution was observed in 26/30 samples with one additional sample providing read level evidence. All results were validated by Sanger sequencing of both tumor and germline controls. This mutation was first observed in activated b-cell type diffuse large cell lymphoma (Ngo et al, Nature 2011) where it was shown to constitutively activates the IRAK4/IRAK1 signaling cascade (shown left). We further observed a conserved 3p acquired uniparental disomy event in four patients making L252P effectively homozygous in these individuals (shown below).

CXCR4 WHIM Syndrome Mutations in WM

Warts, Hypogammaglobulinemia, Infection, and Myelokathexis (WHIM) Syndrome is an autosomal dominant disorder that can be caused by tonic CXCR4 signaling due to impaired receptor internalization. In this study we confirmed 8/30 (27%) of patients had somatic CXCR4 mutations identical or functionally equivalent to those found in WHIM syndrome families.

CXCR4 mutations found in WM Patients

Five unique non-synonymous somatic mutations were found in a total of 8/30 (27%) of samples (A). Every finding was validated by Sanger sequencing. Representative traces for each position can be seen in (B). All variants resulted in a truncation of the cytosolic tail containing the regulatory phospho-serine leaving the seven transmembrane helix region involved in signaling and ligand binding intact (C). Protein sequence is for the canonical full-length transcript.

Summary

Using WGS and confirmatory Sanger sequencing, we have identified several somatic variants with oncogenic function, the most common of which include MYD88 L265P, the c-terminal domain of CXCR4, and ARID1A.

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