**Clonotypic IgH VDJ Sequences in Waldenstrom's Macroglobulinemia Indicate its Origin from an Unusual Memory B Cell Subset**

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Waldenstrom's macroglobulinemia is characterized by pleiomorphic B lineage cells in the bone marrow (BM) and monoclonal IgM in the serum. The unique IgH VDJ sequence that characterizes the malignant clone in each WM patient, termed clonotypic, was identified for 11 WM patients. For 10/11 WM patients (91%), the clonotypic IgH was of the VH3 family, with predominantly long CDR3 regions, characteristic of those found in antigen-stimulated populations. Others have shown a VH3 predominance in some IgM B cell malignancies (IgM myeloma, some CLL, follicular lymphoma) but unlike WM, these malignancies have relatively short CDR3 regions. Clonotypic VDJ regions in WM had extensive somatic mutation with mainly replacement mutations, although for one patient a germline VH3 sequence was identified as clonotypic. For 7/7 patients, clonotypic IgM transcripts were readily detected in both blood and BM, clearly identifying a blood-borne compartment of WM. For 4/4 patients, clonotypic IgM was accompanied by clonotypic IgD in blood and/or BM, but post-switch clonotypic isotypes were not detected. In all WM blood and BM cells analyzed, $CD20^+$ B cells were detectable and expressed surface IgM. In most cases these cells also expressed surface IgD. The majority of WM patients lacked detectable $CD138^+$ plasma cells in either blood or BM. Although the extensive somatic mutation indicates that the WM cell of origin is likely to be a post-germinal center memory B cell, within the IgM compartment, $CD27^+$ $IgM^+$ B cells are usually the minor subset. Work by others indicates that the normal $IgM^+D^+$ $CD27^-$ B cell counterparts to the putative WM cells are also preferentially VH3, but in contrast to WM are naive cells having unmutated VDJ sequences. Sorted $CD27^-$ $IgM^+D^+$ B cells from one patient were shown to express clonotypic transcripts. Longitudinal analysis suggests that phenotypic identification of B cells in blood of WM patients is insufficient to monitor the malignancy. At a time when serum IgM had decreased and clonotypic transcripts were very weak, the number of $CD20^+$ $IgM^+D^+$ B cells increased dramatically. The lack of clonotypic transcripts suggests that the majority of the circulating B cells in this patient were polyclonal and were likely not part of the WM clone, indicating that monitoring of clonotypic IgH provides the most accurate identifier of WM cells. Work is in progress to identify clonotypic IgH *in situ* using BM core biopsies, as a means to determine which B lineage cells persist throughout therapy and which clonal components expand at the time of relapse. *Supported by the Research Fund for Waldenstrom's.*