Figure\_S1.tif

**Multiparameter flow cytometry to define immunophenotype of single tumor cells in mult-HCL.** Normal PBMNCs and isolated spleen cells from HCL cases were immunophenotyped by gating on live cells and staining for CD19/CD11c/CD103 and sIgH/L surface expression. (A) Following FSC/SSC gating, CD19hiCD11chicells (for 7/10 cases) or CD19hiCD11chiCD103+ cells (for 3/10 cases) were examined for sIg expression using rabbit F(ab`)2 anti-human Ig antibodies. Normal PBMNC are virtually devoid of B-cells that are dual CD11chiCD103+. (B) Representative example of immunophenotype of a mult-HCL tumor. IgD-ve mult-HCL Case 10 CD19hiCD11chiCD103+ cells display co-expression of sIgG and λ; in 100% of HCL cells, with subsets of cells also sIgA (72%) and sIgM (18%) positive.

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Figure\_S2.tif

**Anti-sIg stimulation results in endocytosis of functional BCR in IgD<sup>+</sup> mult-HCL.** Cells were stimulated with goat F(ab`)2 anti-Ig antibodies for 1 hour either at 37°C to allow endocytosis or at 4°C to prevent it. Changes in stimulated sIg expression levels were then measured by secondary staining using rabbit F(ab`)2 anti-goat F(ab`)2 (A, top panels). Changes in expression of paired sIg were measured using rabbit F(ab`)2 anti-Ig (A, lower 2 rows). Loss of functional sIg was further confirmed by measuring induction of calcium flux following endocytosis (B). Using the anti-goat secondary antibody method (A, top panels) stimulation via sIgD, λ and sIgM in Case 13 as a representative example resulted in reduction of surface expression of these specific isotypes. Stimulation of non-functional sIgA in this tumor did not result in endocytosis (A, top right panel). Using the rabbit F(ab`)2 staining method (A, lower 2 rows) stimulation of sIgD (left panels) in Case 13 resulted in loss of λ, but not κ light chain expression. Lambda stimulation, second column, resulted in loss of both surface IgD and IgM expression. IgM stimulation, third column, resulted in marginal loss of λ but not κ; surface expression. Inset of a Table of the sIg phenotype and functional sIg isotypes is included for reference purposes to evaluate data (A, lower right). The ability to induce Ca2+ flux (B) via IgD was markedly reduced following endocytosis by λ stimulation (B, left panel). Ability to induce Ca2+ flux via λ was ablated by IgD endocytosis and also greatly reduced by IgM endocytosis (B, centre panel). Similarly, IgM induced flux was completely ablated following endocytosis following anti-λ stimulation (B, right panel).

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Figure\_S3.tif

**Stimulation of BCR in mult-HCL triggers apoptosis not proliferation.** Cells were stimulated with goat F(ab`)2 anti-Ig antibodies for 4 hours at 37°C and early apoptosis was measured in CD19+CD11c+CD103+ HCL cells using Apostat (left panels). Alternatively, XTT, a tetrazolium salt that is cleaved to formazan in metabolically active cells only was used to assess viability and proliferation following anti-BCR stimuli and OD assayed at 490 nm (right panels). In 2/2 HCL cases (HCL #3, top panels; HCL #10, lower panels) ApoStat assays revealed significant increases in apoptosis in response to both functional heavy (IgG) and light (κ; HCL 3)/λ;HCL 10) chain stimulation. No apparent increase in numbers of metabolically active tumor cells was observed in response to BCR stimulation in either HCL case by XTT assay (right panels).

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**Contains supplementary materials and methods and Table S1.**

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