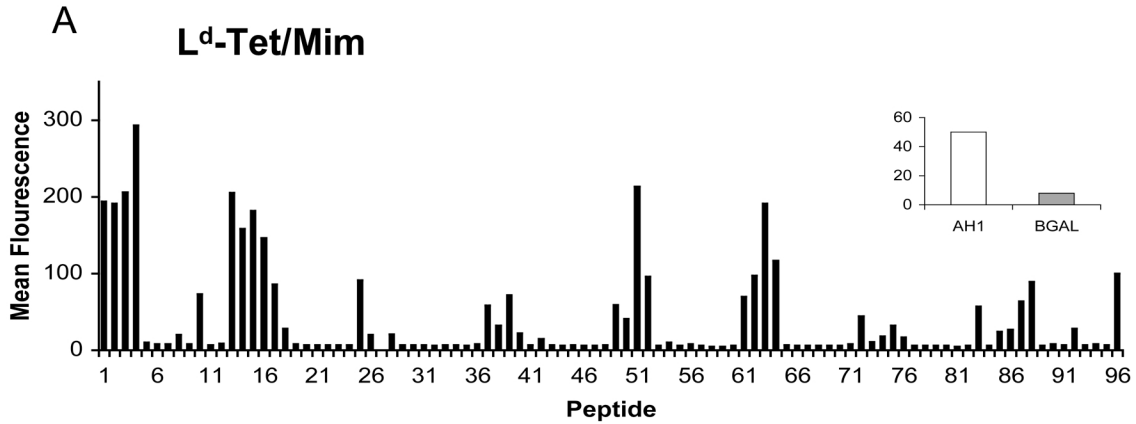
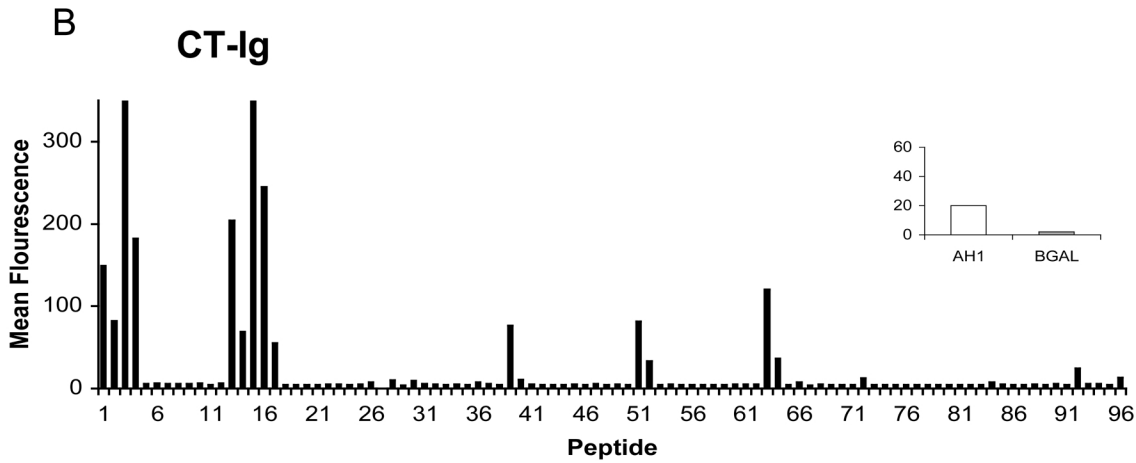
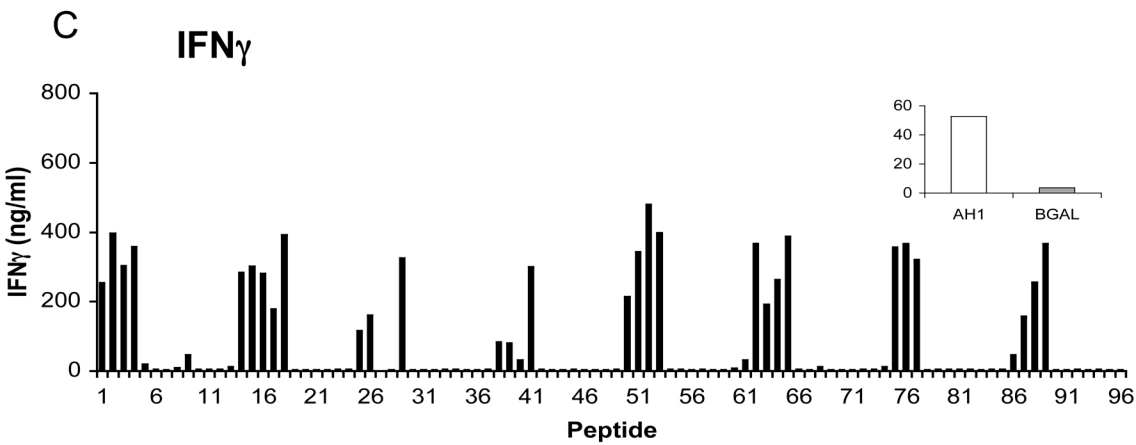

A
Ld-Tet/Mim
Mean Flourescence
0
100
200
300
1 6 11 16 21 26 31 36 41 46 51 56 61 66 71 76 81 86 91 96
Peptide
0
20
40
60
AH1
BGAL


B
CT-Ig
Mean Flourescence
0
100
200
300
1 6 11 16 21 26 31 36 41 46 51 56 61 66 71 76 81 86 91 96
Peptide
0
20
40
60
AH1
BGAL


C
IFNγ
IFNγ (ng/ml)
0
200
400
600
800
1 6 11 16 21 26 31 36 41 46 51 56 61 66 71 76 81 86 91 96
Peptide
0
20
40
60
AH1
BGAL

Supplemental figure 1. In vitro screening of candidate mimotopes with a range of binding properties identified for the AH1-specific T cell clone using a combinatorial peptide library. 96 peptides from the combinatorial peptide library were synthesized using standard Fmoc chemistry and a Spyder multiple-peptide synthesizer. Control assays were performed with the AH1 peptide and the negative control βgal peptide (inset). Synthesis of mimotope 27 was unsuccessful and therefore no related data were available in these assays. Large scale synthesis of the mimotope 27 was chosen for further study based on the amino acid sequence of the peptide, not empiric data. (A) L^d-tet was incubated with an excess of the indicated peptide. The T cell clone was stained with L^d-tet and the mean fluorescence intensity was graphed as determined by flow cytometry. (B) T2-L^d cells were incubated with the mimotopes and stained with a divalent single chain TCR from the T cell clone that recognizes AH1 (CT-Ig). TCR binding was visualized using a biotinylated antibody against IgG1 and avidin-PE. (C) IFNγ production from the T cell clone was measured by ELISA following incubation with 1 μM peptide at 37˚C for 24 h.