

Supplemental Figure 1. VH125Tg/V κ 125^{SD} B1 cell differentiation is impaired. Peritoneal lavage or spleens were harvested from n = 6 WT/B6, n = 6 VH125Tg/V κ 125^{SD}/B6, n = 3 WT/NOD, or n = 3 VH125Tg/V κ 125^{SD}/NOD 9-13 week old male and female mice and flow cytometry was used to identify CD5⁺ B1 cells. (A) The frequency of CD5⁺ cells among transgenic (black bars, IgM^{a+}, VH125Tg/V κ 125^{SD} mice) or non-transgenic (white bars, IgM^{b+}, WT mice; IgM^{a-}, VH125Tg/V κ 125^{SD} mice) B Cells (CD19⁺ IgM⁺ live lymphocytes) was assessed in peritoneal lavage. (B) Spleens were harvested and the frequency of CD5⁺ cells among total B cells (black bars, B220⁺ IgM⁺ live lymphocytes) or T1 gated B cells (white bars, B220⁺ CD21^{low} CD23^{low} IgM^{a high} live lymphocytes) was assessed in the indicated genotype. The average ± SD is shown.

Supplemental Table I. Number of non-insulin-binding or insulin-binding B cells following insulin autoantigen-targeted therapy of anti-insulin VH125Tg/Vk125^{SD}/NOD mice^a

| | Immature ^{b, c} | T1 ^{b, c} | T2 ^{b, c} | Follicular ^{b, c} | Pre-Marginal | Marginal Zone ^{b, c} |
|--------------------------------|--------------------------|--------------------|--------------------|----------------------------|----------------------|-------------------------------|
| | | | | | Zone ^{b, c} | |
| Untreated, Non-Insulin-Binding | 0.18 ± 0.07 | 18 ± 14 | 21 ± 6 | 26 ± 13 | 0.97 ± 0.99 | 33 ± 21 |
| mAb123, Non-Insulin-Binding | 0.37 ± 0.08 | 91 ± 15 | 162 ± 10 | 40 ± 29 | 6.9 ± 2.8 | 75 ± 29 |
| Untreated, Insulin-Binding | 7.0 ± 2.3 | 987 ± 776 | 834 ± 128 | 1530 ± 674 | 83 ± 83 | 3229 ± 2360 |
| mAb123, Insulin-Binding | 3.7 ± 2.1 | 285 ± 301 | 776 ± 303 | 4383 ± 1357 | 194 ± 43 | 977 ± 1290 |

^a Spleen or bone marrow was freshly isolated 2-7 d after single i.p. injection of mAb123, n = 2 untreated mice, n = 3 mAb123-treated mice

^b Gated as in Fig. 6B ^c Average number of cells x $10^3 \pm$ standard deviation. Statistical analysis was not performed.

Supplemental Table II. Number of non-insulin-binding or insulin-binding B cells following insulin autoantigen-targeted therapy of anti-insulin VH125Tg/Vk125^{SD}/RAG2-GFP/B6 mice^a

| | Immature ^{b, c, d} | T1 ^{b, c, d} | T2 ^{b, c, d} | Follicular ^{b, c, d} | Pre-Marginal | Marginal |
|--------------------------------|-----------------------------|-----------------------|-----------------------|-------------------------------|-------------------------|-------------------------|
| | | | | | Zone ^{b, c, d} | Zone ^{b, c, d} |
| Untreated, Non-Insulin-Binding | 3.3 ± 2.9 | 17 ± 13 | 67 ± 40 | 194 ± 139 | 10 ± 10 | 69 ± 42 |
| mAb123, Non-Insulin-Binding | $58 \pm 54 * *$ | $106 \pm 129 **$ | 162 ± 169 | 139 ± 100 | 8.9 ± 6.9 | 62 ± 26 |
| Untreated, Insulin-Binding | 43 ± 24 | 247 ± 258 | 1532 ± 1123 | 5366 ± 3740 | 338 ± 256 | 2513 ± 1062 |
| mAb123, Insulin-Binding | $410 \pm 256^{**}$ | 294 ± 328 | $242\pm210^{**}$ | 3181 ± 2696 | $64 \pm 77*$ | $135 \pm 98 **$ |

^a Spleen or bone marrow was freshly isolated 1d after final i.p. injection of mAb123 (injected every 2 d for 1 wk), $n \ge 6$ mice per group

^b Gated as in Fig. 7A ^c Average number of cells x $10^3 \pm$ standard deviation ^d ** p < 0.01, Mann Whitney test, two tailed, mAb123 compared to untreated

Supplemental Table III. Number of insulin-binding or non-insulin-binding B cells following short-term insulin autoantigen-targeted therapy in VH125Tg/NOD mice^a

| | Immature ^{b, c, d} | T1 ^{b, c, d} | T2 ^{b, c, d} | Follicular ^{b, c, d} | Marginal Zone ^{b, c, d} | Pancreatic Draining Lymph Nodes ^{b, c, d} |
|--|-----------------------------|-----------------------|-----------------------|-------------------------------|-------------------------------------|--|
| Untreated, Insulin-binding | 3.9 ± 1.7 | 6.3 ± 2.7 | 15.4 ± 10.3 | 25.9 ± 12.4 | 21.5 ± 8.2 | 1.9 ± 1.7 |
| $F(ab')_2$ 123, Insulin-binding | 3.8 ± 1.3 | 5.6 ± 3.9 | 8.6 ± 4.7 | 47.5 ± 22.2 | 27.0 ± 12.6 | 1.2 ± 0.65 |
| mAb123, Insulin-Binding | $1.2 \pm 0.5*$ | $0.75\pm0.58*$ | $0.63\pm0.36*$ | 29.5 ± 15.9 | $6.7 \pm 7.6^{*}$ | $0.44 \pm 0.13^{*}$ |
| Untreated, Non-Insulin-Binding | 576 ± 321 | 593 ± 257 | 1615 ± 892 | 2526 ± 1054 | 1715 ± 784 | 94 ± 36 |
| F(ab') ₂ 123, Non-Insulin-Binding | 588 ± 192 | 780 ± 435 | 1943 ± 685 | 3258 ± 943 | 2183 ± 700 | 104 ± 48 |
| mAb123, Non-Insulin-Binding | 498 ± 127 | 682 ± 164 | 1867 ± 668 | 2860 ± 1345 | 1797 ± 722 | 64 ± 12 |

^a Spleen or bone marrow was freshly isolated 1-2 d after single i.p. injection of indicated therapy, n = 5 mice per group ^b Gated as in Fig. 8B ^c Average number of cells x $10^3 \pm$ standard deviation ^d * p < 0.05, ** p < 0.01, Mann Whitney test, two tailed, as compared to untreated