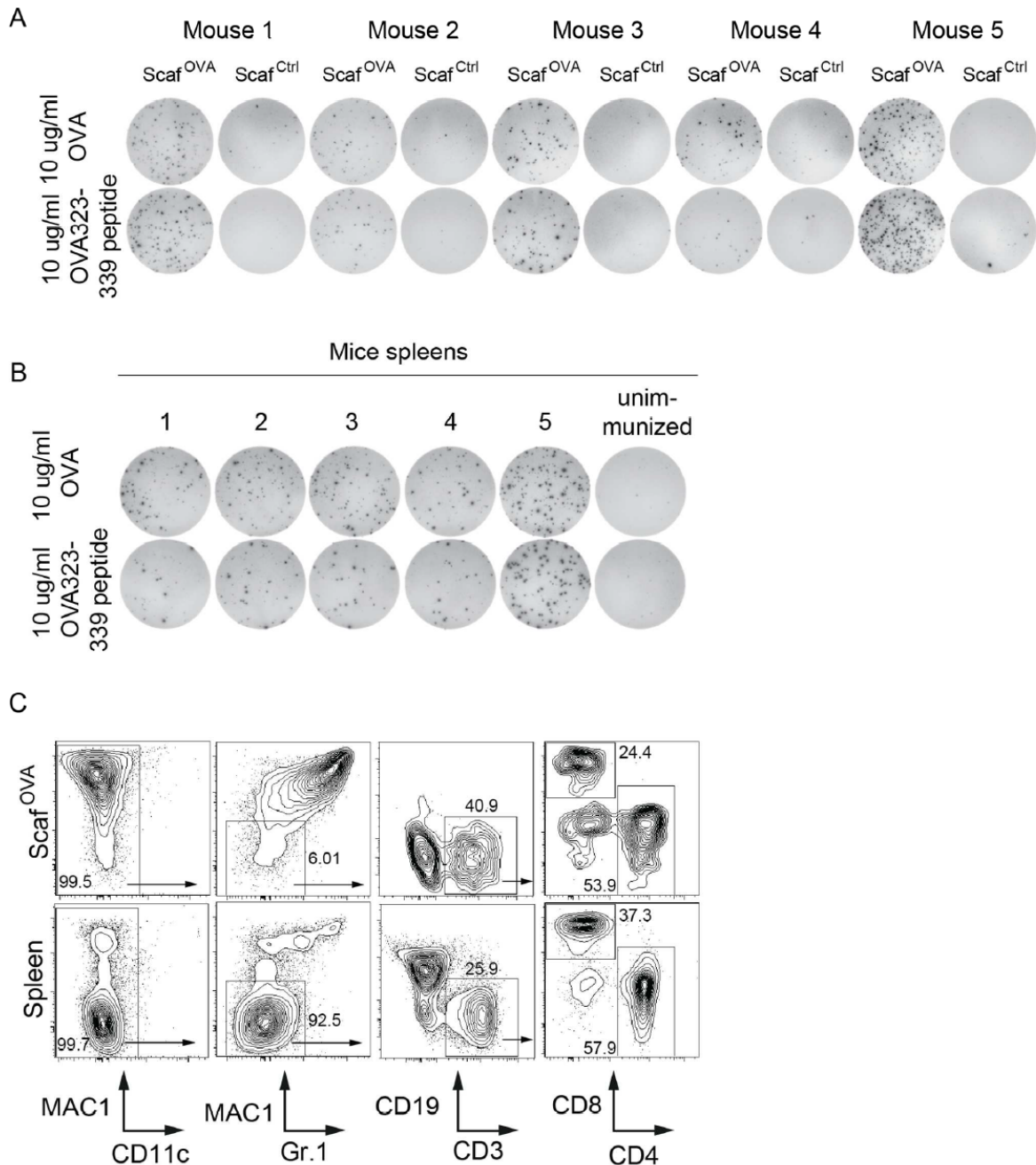


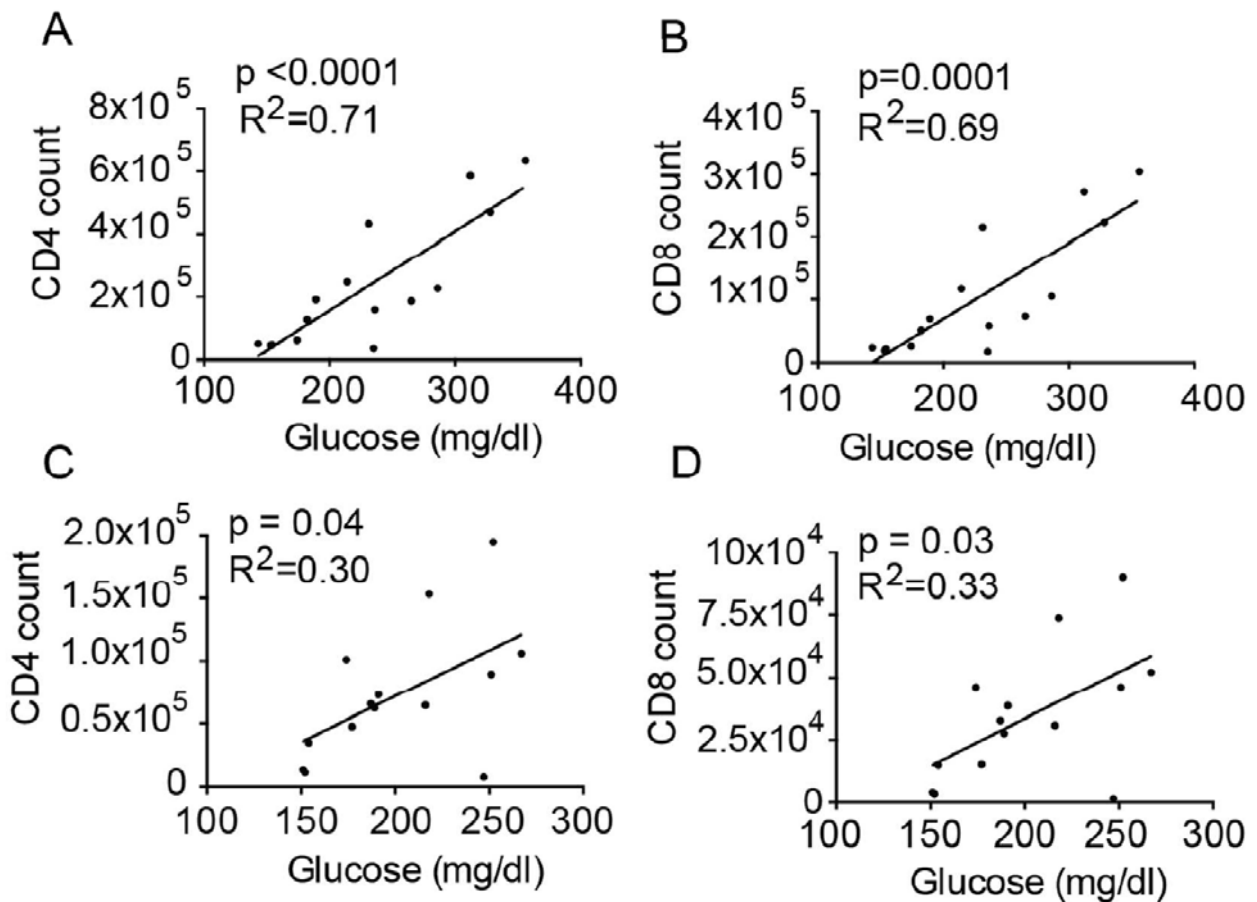
SUPPLEMENTARY DATA

Supplementary Figure 1. ELISPOT and flow cytometry assays of cells isolated from scaffolds. (A) ELISPOT wells from which the data for OVA and control scaffolds in Fig. 1 E-H are derived. (B) Representative ELISPOT T wells for spleen data in Fig. 1 E-H. The spleen data in Fig. 1 E-H is an average of four independent ELISPOT wells. (C) Representative flow cytometry analysis from which the numbers of CD4⁺ T cells are calculated in Fig. 1 G-H. Frequencies of granulocytes (Mac1⁺ CD11c⁻), monocytes (Mac1⁺ Gr1⁺), T cells (CD3⁺), and CD4⁺, CD8⁺ were determined. Live cells were gated as propidium iodide exclusion and CD45⁺.



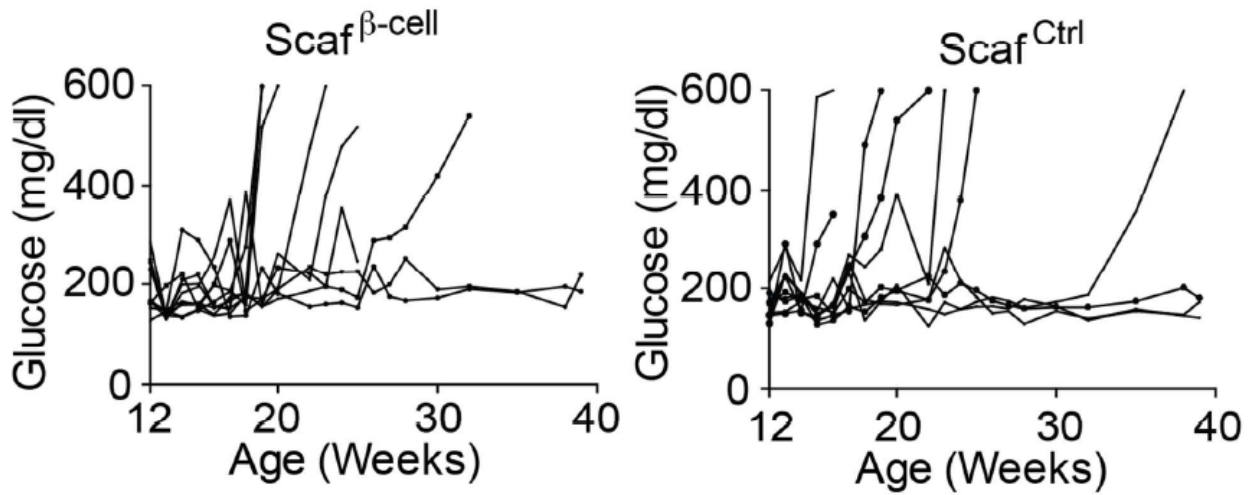
SUPPLEMENTARY DATA

Supplementary Figure 2. Numbers of infiltrating CD4⁺ and CD8⁺ T cells correlate with increased glucose measurements in both β -cell and control scaffolds. Numbers of CD4⁺ T cells (A) and CD8⁺ T cells (B) in β -cell scaffolds, and numbers of CD4⁺ T cells (C) and CD8⁺ T cells (D) in control scaffolds were determined in 15 β -cell or 14 control scaffolds explanted after 14 days in NOD mice, and were graphed against blood glucose levels (mg/dl). Best-fit lines were generated by linear regression (Prism software).



SUPPLEMENTARY DATA

Supplementary Figure 3. Diabetes development in NOD mice implanted with scaffolds. β -cell scaffolds or control scaffolds were implanted in NOD mice at 10 weeks of age and weekly glucose measurements were conducted until the age of 40 weeks.



SUPPLEMENTARY DATA

Supplementary Table 1. Lineage analysis of cells within scaffolds and spleen. Hematopoietic lineages determined from 4 β -cell and 6 control scaffolds explanted after 14 days in 14 week old NOD mice.

Sample	MAC1+ CD11c- Gr.1-	CD11c+	Gr1+ MAC1+	CD19+	CD4+	CD8+	$\gamma\delta$ +
Scaf β -cell	49.4 \pm 5.7 ^a	16.7 \pm 7.0	17.4 \pm 10.9	0.2 \pm 0.1	1.5 \pm 0.7	1.3 \pm 0.8	3.1 \pm 1.4
Scaf Ctrl	38.3 \pm 8.3	40.6 \pm 17.2	9.0 \pm 9.0	0.5 \pm 0.4	1.1 \pm 0.4	0.5 \pm 0.3	3.4 \pm 1.2
Spleen	4.9	1.6	2.3	34	31	19	1.3

^a percentages of total cells within scaffolds or spleens. Values indicate mean \pm S.D.

SUPPLEMENTARY DATA

Supplementary Table 2. TCR gene sequences that overlap between pancreases and scaffolds. TCR β protein sequences that are greatly expanded (3 standard deviations above the mean) in β -cell scaffolds or the pancreases as indicated in Fig. 5C by Greek letters I-III.

Highly expanded in pancreas (I)	TCR β V	TCR β J
CASSPGQEQYF	15	02-07
CASSLGSGNTLYF	16	01-03
CASGGTGGAREQYF	13	02-07
CASSDAGQYEQYF	13	02-07
CASSQGGPGERLFF	2	01-04
CASSDSAETLYF	19	02-03
Highly expanded in pancreas and β -cell scaffolds (II)		
CTCSATGANERLFF	1	01-04
CASGDAREQYF	13	02-07
CTCSAETVISNERLFF	1	01-04
CTCSAVSNERLFF	1	01-04
Highly expanded in β -cell scaffolds (III)		
CASSDTYQDTQYF	5	02-05
CASGDEGYEQYF	13	02-07
CASSDAGTTNERLFF	13	01-04
CAWSLQGISNERLFF	31	01-04
CTCSGQGISNERLFF	1	01-04
CASSQETANERLFF	5	01-04
CASGDEGNTLYF	13	02-04
CASSQVGHPSAETLYF	5	02-03
CASGSSYEQYF	13	02-07

Supplementary Protocol #1.

Gas Foaming-Particulate Leaching Protocols for fabricating PLG T cell Scaffolds

Specialty Materials Needed

30 μm LG30K PLG microspheres (Cat. No. LG30-8515, Phosphorex, Hopkington, MA)

8.5 mm pellet die (machined and custom made) – standardized pellet dies are also available through (Carver, Inc.)

Carver press (Carver Inc)

Pressure Vessel (Custom made) – Degassing vacuum chambers with at least 1000 PSI capacity may also be used.

Industrial CO₂ with purity of 99.5% as foaming agent.

β -cell Cell Lysate Materials

NIT-1 cells (ATCC CRL-2055)

DMEM/F12 containing 10% fetal bovine serum

NIT-1 Lysate Procedure

1. Culture NIT-1 Cells in DMEM/F12 containing 10% fetal bovine serum.
2. Passage cells at 70-80% confluency.
3. Trypsinize cells and quench the trypsin with complete medium containing 10% fetal bovine serum, wash with PBS once, and resuspend the cells in PBS.
4. Count the cells (1×10^7 cells per scaffold).
5. Sonicate the cells for 5 seconds.

Adsorb Lysate Material onto Polymer Microspheres

1. Mix 18mg per scaffold of PLG microspheres with the cell lysate at a ratio of 556,000 cells/mg of microspheres. Approximately 1ml of PBS per 3-5 scaffolds.
2. Vortex for 30 seconds to make it homogenous and let stand at RT for 15 min.
3. Vortex mixture for 15 seconds and snap freeze the microspheres/lysate mixture in liquid N₂.
4. Lyophilize cell lysate and polymer for approximately 2 days or until dry.
5. Use a spatel to crush the lyophilize material into a small particles

Preparation of 250-450 μm NaCl as Leachable Porogen

1. Clean 425 μm and 250 μm ASTM sieves (#40 and #60; W.S. Tyler Inc.) and collection pan.
2. Assemble the sieves with the 425 μm on top, 250 μm second and the collection plate on the bottom.
3. Add approximately 100 g of NaCl (Sigma Aldrich) to the top sieve.

SUPPLEMENTARY DATA

4. Slowly shake the sieves to allow the salt granules to fall through the sieves.
5. After the NaCl has been allowed to fall through the two sieves, all of the salt in the 250 μ m sieve should be collected, weighed and placed in a polypropylene container. Note: any salt captured in the 425 μ m sieve and the bottom pan can be discarded or used in another capacity.

Scaffold Formation by Tablet Compression

1. Weigh out 200mg sieved NaCl for each scaffold.
2. Mix thoroughly with spatula for 5-15min. Note: it needs to be homogenous because salt generates the pores in the biomaterial after having been eluted.
3. Weigh out approximately 262mg (18mg of polymer, 240mg of NaCl, and 3.6mg of cell lysate) of the substance for each scaffold in eppendorf tubes
4. Place mixture from each eppendorf tube in a compression pellet die and place in press.
5. Compress for 1 min at 1500 psi. Remove compressed scaffold pellet and repeat with each tube.

Gas foaming T cell Scaffolds

Step #1 Scaffold Pressurization:

Note: Wear Safety attire and be cautious when working with high pressure.

1. Open the pressure vessel.
2. Place the scaffolds to be foamed into the trays (Teflon trays can be fabricated for scaffold placement).
3. Ensure that scaffolds are not touching each other.
4. Place the trays into pressure vessel.
5. Place a spacer between each of the shelves to be used (ensure that trays are separated from each other to allow for continuous airflow and gas contact with scaffolds).
6. Seal pressure vessel so it is air tight (making sure the o-ring or sealer is in place and tighten bolts or other closing mechanisms).
7. Once completely tightened, close gas intake and outlet valves.
8. Ensure that pressure vessel intake is connected securely to industrial grade CO₂ (99.5% purity) gas tank. Pressure regulators should be fitted to monitor and control internal pressure.
9. Open the intake valve to the pressure cylinder slowly.
10. Slowly turn the regulatory control to tune the pressure in the vessel appropriately.
11. Slowly and carefully bring the pressure up to 800 PSI.
12. Allow to foam for at least 16 hours.

Step #2 Depressurization for polymer foaming

1. Ensure that pressure cylinder intake valve and valve to CO₂ source tank is closed.
2. Slowly open the outlet valve of pressure vessel to release gas at a constant rate. The pressure should be completely released in 1-2 minutes to ensure proper foaming.
3. Open vessel and place foamed devices in sealed sterile containers and store at -20°C.

NaCl particulate Leaching

1. Place 1 scaffold in 10 mL of ddH₂O at RT. Up to 5 scaffolds can be placed in 50mL leach bath.
2. Leach out NaCl for 4 hours.
3. Implant the scaffolds.