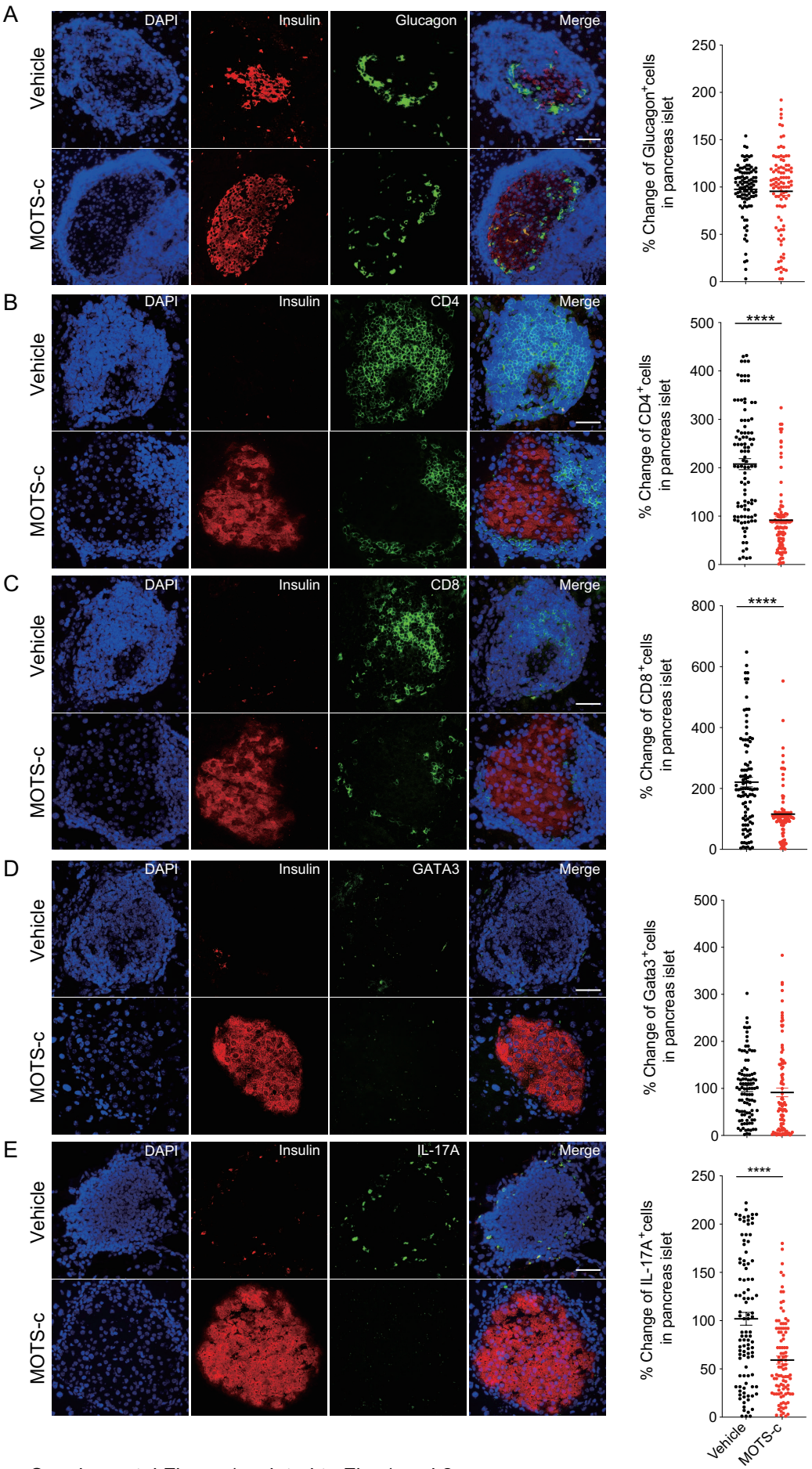
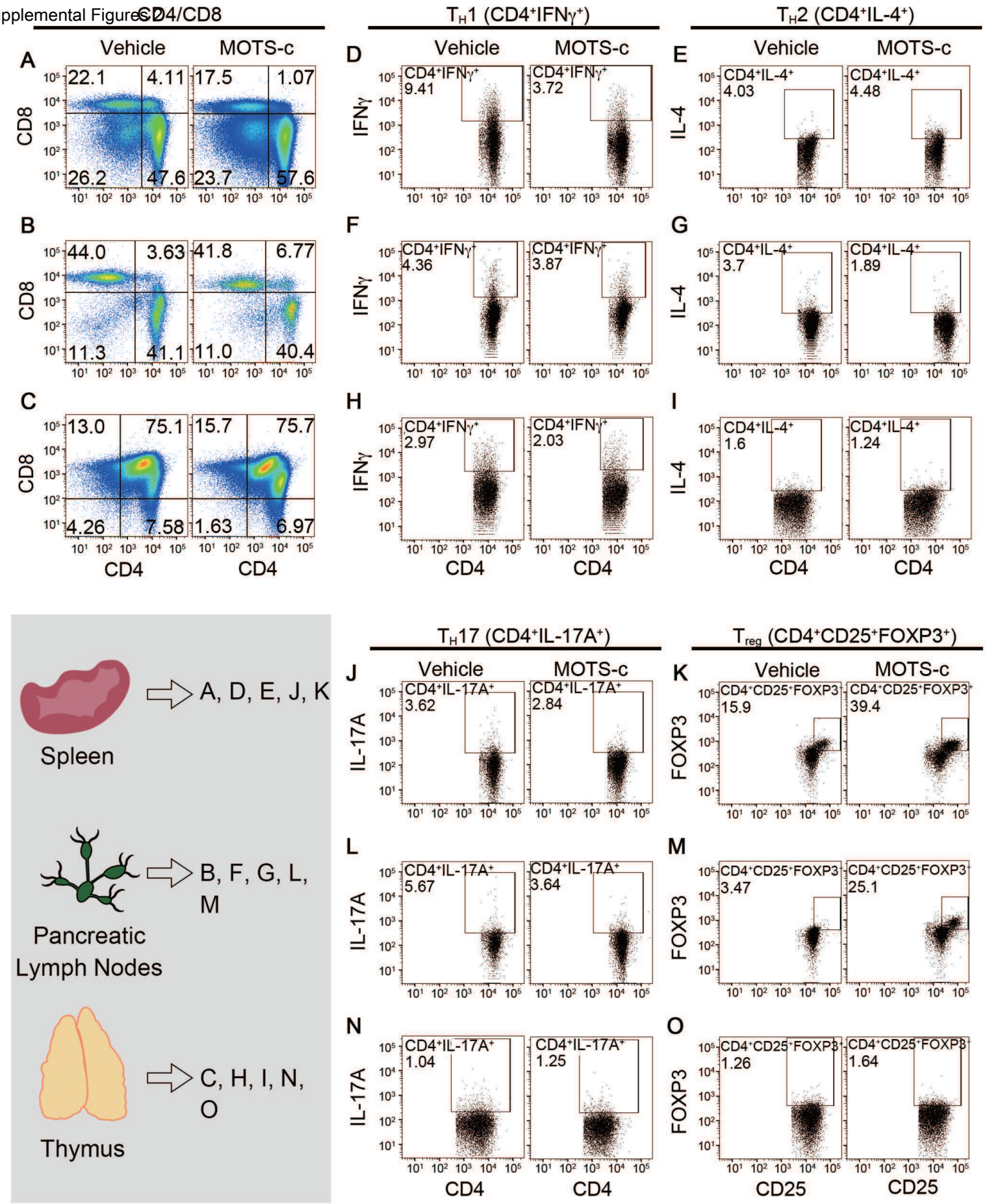


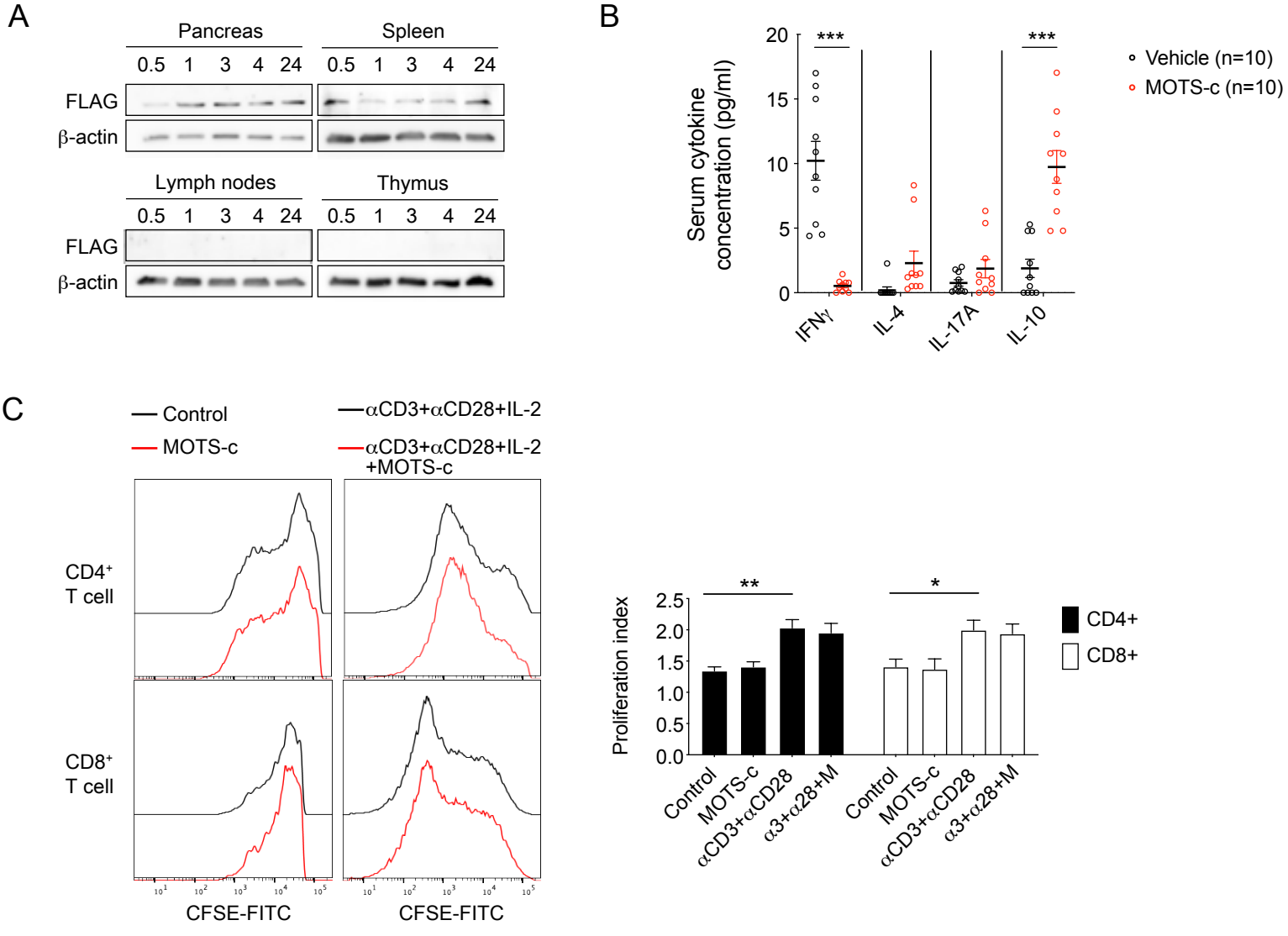
Supplemental Figure 1, related to Fig. 1 and 2.
18 weeks old-NOD mice pancreas sections were stained with insulin (red); and (**A**) glucagon, (**B**) CD4, (**C**) CD8, (**D**) GATA3, and (**E**) IL-17A (green) were performed. IF staining on pancreatic sections were prepared as described and the representative data and graphs from ten pancreases with ten sections for each group (n=100). Error bars are S.E.M, ****p<0.0001, Student's t-test. Scale bars: 50 μm.



Supplemental Figure 2, related to Fig. 2.

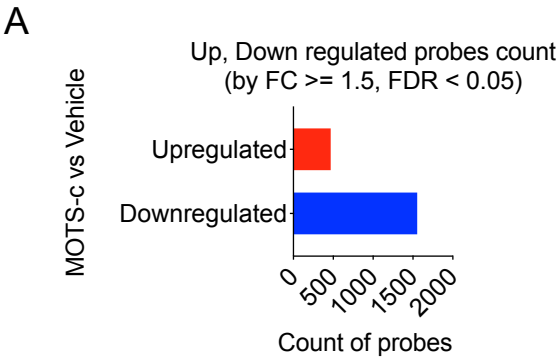
Representative graphs for NOD $CD4^+$ and $CD8^+$ T cells (n=10) in (**A**) spleen, (**B**) pancreatic lymph nodes, and (**C**) thymus. NOD $CD4^+$ T cells were subdivided into T_H1 ($CD4^+IFN\gamma^+$), T_H2 ($CD4^+IL\text{-}4^+$), T_H17 ($CD4^+IL\text{-}17A^+$), and T_{reg} ($CD4^+CD25^+FOXP3^+$) in (**D, E, J, K**) spleen, (**F, G, L, M**) pancreaic lymph nodes, and (**H, I, N, O**) thymus.

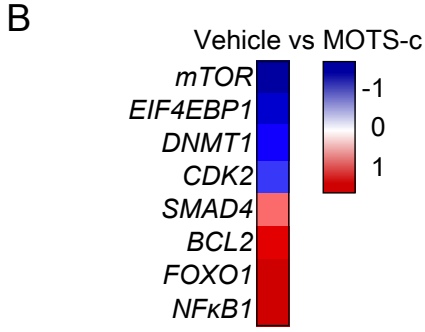
Supplemental Figures 3

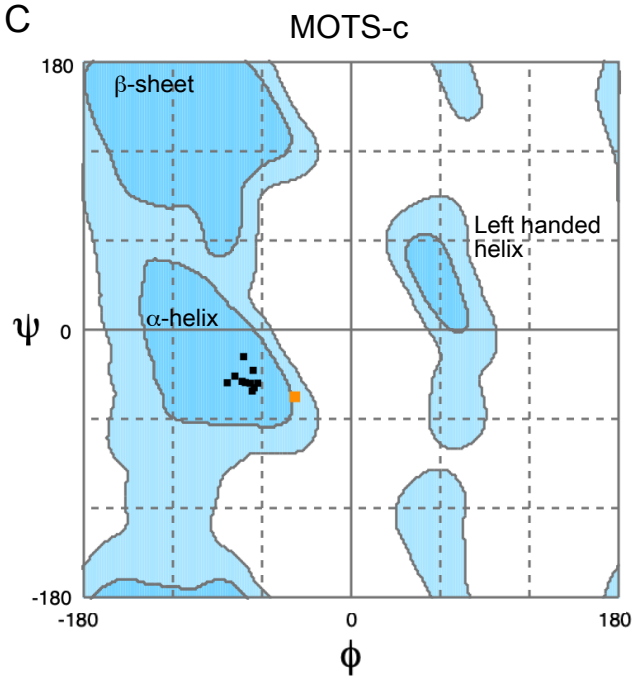


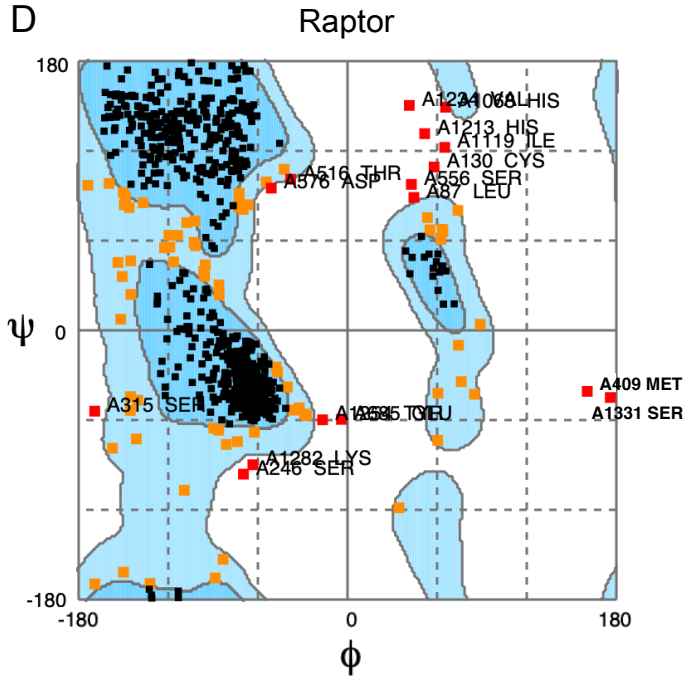
Supplemental Figure 3, related to Fig. 3. MOTS-c increases IL-10 production and CD206 expression in CD11b$^+$ cells.
A. C57BL/6J mice were i.v. injected with flagged-MOTS-c (10 mg/kg) in time dependent manner. Pancreas, spleen, lymph nodes, and thymus were isolated from each mouse to detect FLAG in whole cell lysates. Data are from one represenative of five independent experiments.
B. 18-week-old NOD serum were analyzed for immune cell related cytokines (n=10); two-way ANOVA, ***p<0.001.
C. For proliferation assay, CFSE-stained CD4$^+$ and CD8$^+$ T cells isolated from NOD mice were stimulated with CD3 and CD28 antibodies (2 ug/ml each) for 3 days to evaluate T cell proliferation in the presence or absence of MOTS-c (10 uM); data are mean values ±SEM of three independent experiments.

Supplemental Figures 4









Number of resides in favoured region (~98.0% expected) : 13(92.9%)
Number of residues in allowed region (~2.0% expected): 1 (7.1%)
Number of residues in outlier region: 0 (0%)

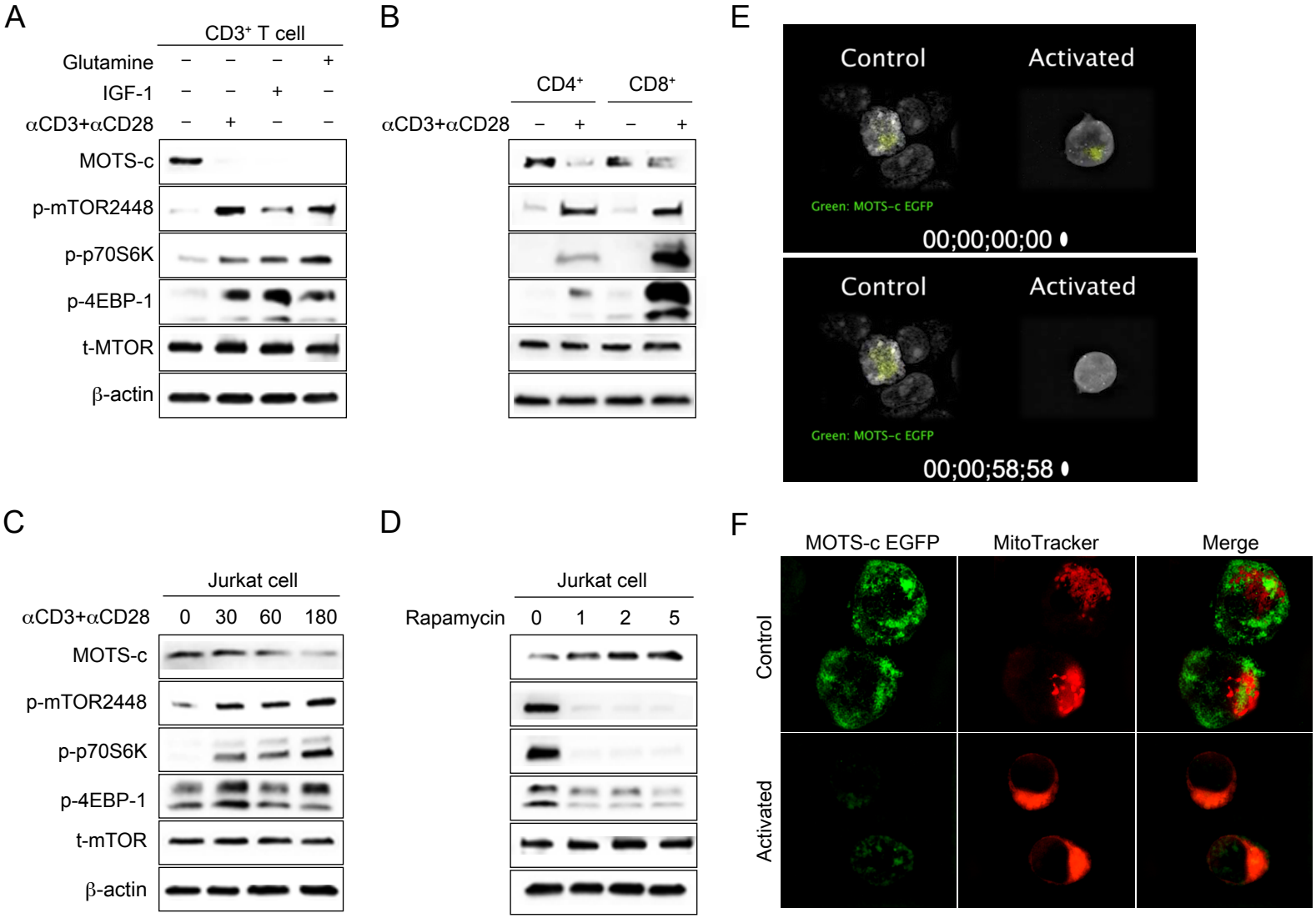
Number of resides in favoured region (~98.0% expected): 937 (89.8%)
Number of residues in allowed region (~2.0% expected): 81 (7.8%)
Number of residues in outlier region: 26 (2.5%)

Supplemental Figure 4, related to Fig. 3

A. Microarray analyses on NOD splenocytes treated with either vehicle or MOTS-c for 18 weeks of age (n=3). Graph shows significantly up- or down-regulated probes count with fold change of 1.5 or higher and FDR less than 0.05.

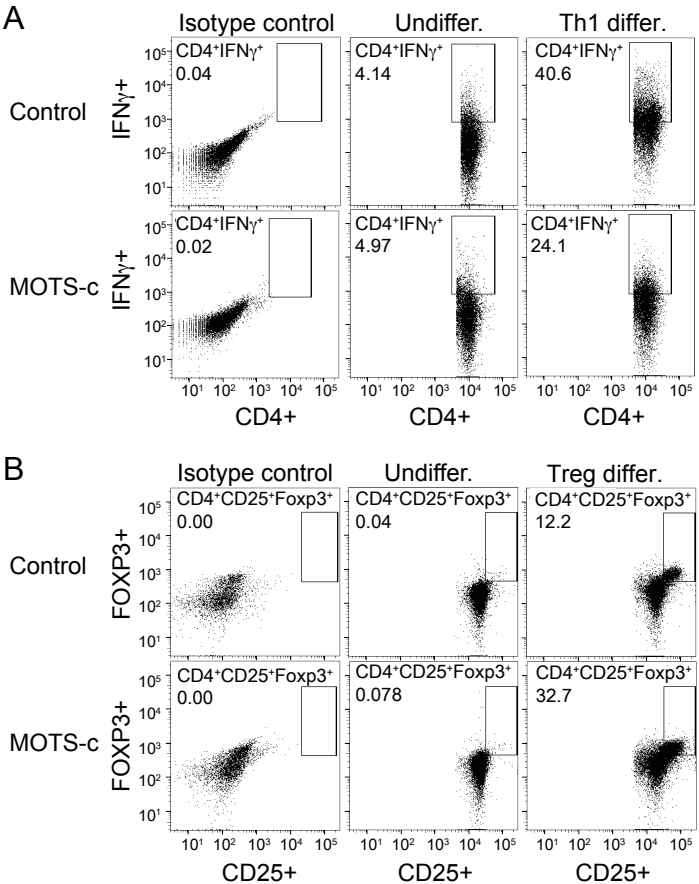
B. KEGG analysis on Th1 and Treg related genes.

C, D. The full Raptor structure (PDB ID:5WBI) was used as template for MOTS-c homology modeling. Ramachandran plot statistics for (**C**) MOTS-c and (**D**) Raptor are shown.



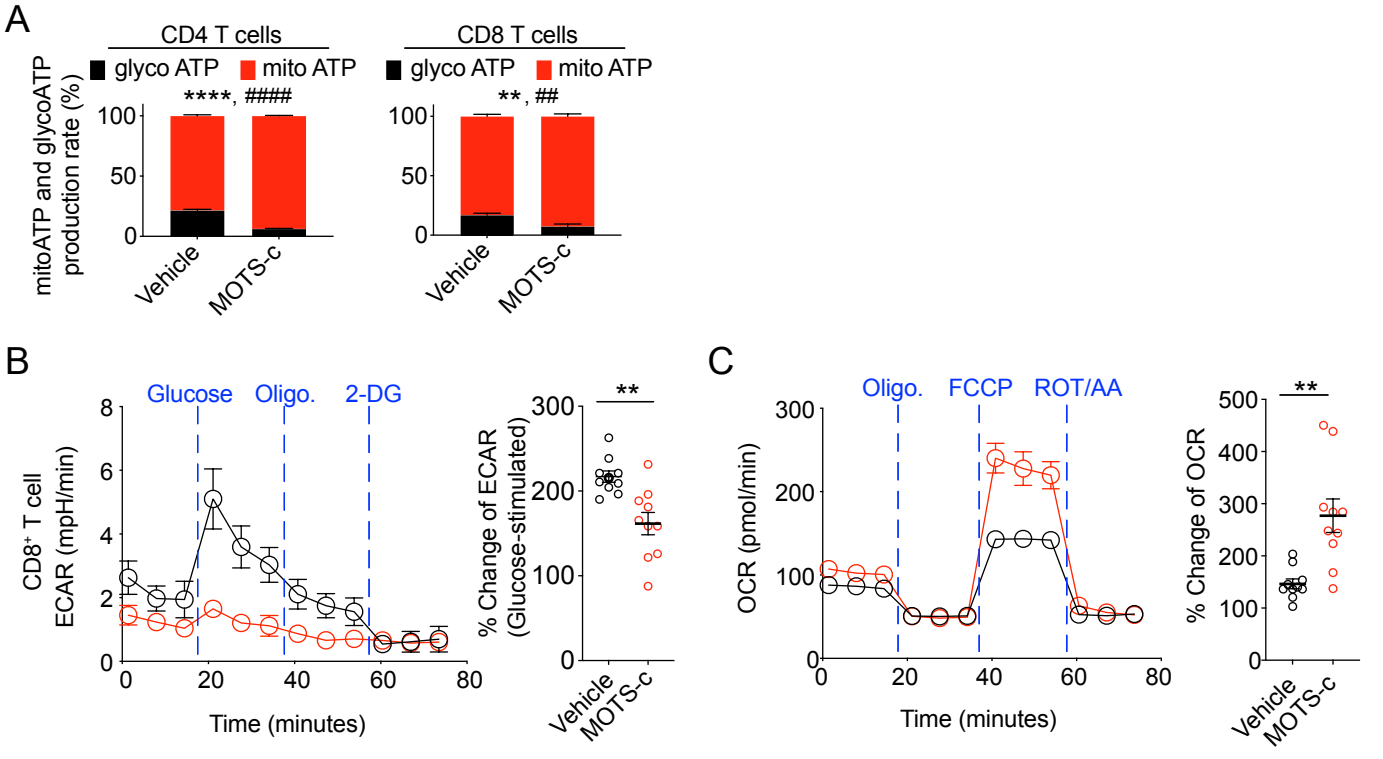
Supplemental Figure 5, related to Fig. 3, 4.
CD3+, CD4+, CD8+ T cells were negatively isolated from C57BL/6J mice spleen. T cells were activated with either soluble mouse α CD3 (4 μg/ml) and αCD28 (10 μg/ml), IGF-1 (1 μg/ml), or glutamine (5 mM), as previously reported for mTORC1 induction, for following conditions: (**A**) CD3+ T cells (30 min) and (**B**) $CD4^+$ and $CD8^+$ T cells (30 min).
Jurkat cells were treated with (**C**) human αCD3 (4 μg/ml) and αCD28 (10 μg/ml) in time-dependent manner and with (**D**) rapamycin in concentration-dependent manner. MOTS-c tagged with EGFP Jurkat cells were activated with human αCD3 (4 μg/ml) and αCD28 (10 μg/ml) (3 hours). Then, cells were fixed to analyze cells with time-lapse live imaging with (**E**) Nanolive and (**F**) confocal microscopy. Data are from one representative of five independent experiments.

Supplemental Figures 6



Supplemental Figure 6, related to Fig. 4.
$CD4^+$ T cells were negatively isolated from C57BL/6J mice spleen. (**A**) T_H1 or (**B**) T_{reg} polarization were performed, as described in the method, in the presence or absence of MOTS-c (10 µM). Then, cells were stained with adequate antibodies for FACS analysis (n=5).

Supplemental Figures 7



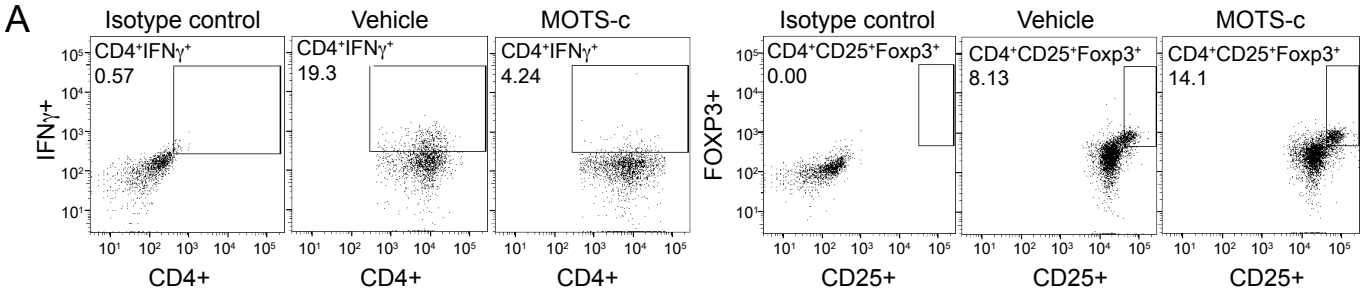
Supplemental Figure 7, related to Fig. 5

A. mitoATP/glycoATP production rate on NOD CD4⁺ and CD8⁺ T cells (n=5), two-way ANOVA, **p<0.01, ****p<0.0001 between group, #p<0.01, ####p<0.0001 between glycoATP and mitoATP.

B. Glycolysis assessment after glucose treatment in NOD CD8⁺ T cells (n=10), Student's t-test, **p<0.01.

C. Assessment of respiratory capacity in NOD CD8⁺ T cells (n=10), Student's t-test, **p<0.01.

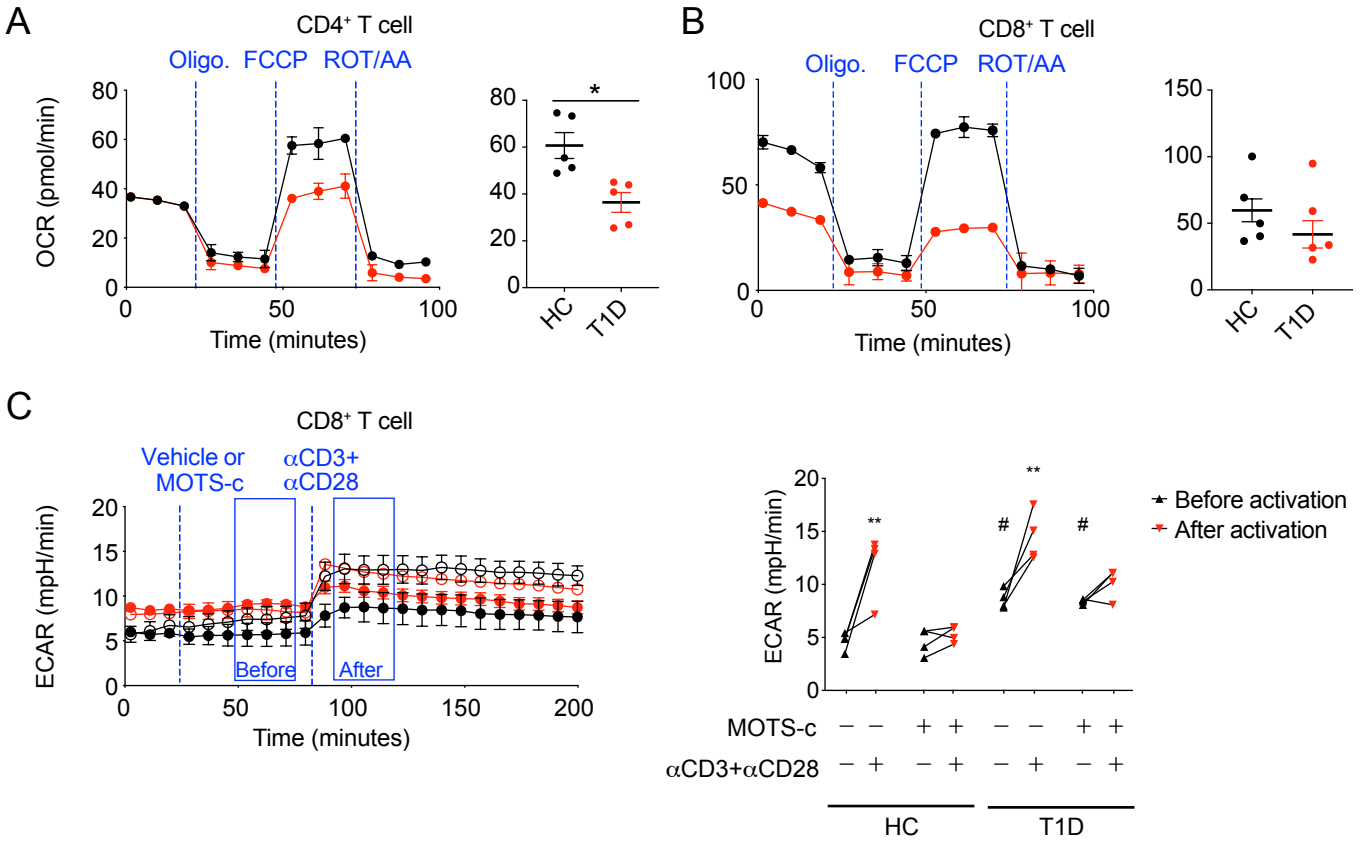
Supplemental Figures 8



Supplemental Figure 8, related to Fig. 6

A. 18-week-old NOD splenocytes (1 x 10^7 cells, donor) from each group were i.v. injected to 6-week-old NOD/SCID mice (n=4, recipient). NOD-SCID mice were sacrificed after 18-weeks post-transfer. Then, the recipient NOD-SCID spleens were isolated to analyze (**A**) $CD4^+IFN\gamma^+$ and $CD4^+CD25^+FOXP3^+$ cells.

Supplemental Figures 9



Supplemental Figure. 9, related to Fig. 7

A-C. Human PBMC-derived T cells were isolated from healthy controls and T1D patients to assess respiratory capacity of (**A**) CD4⁺ T cells (n=5) and (**B**) CD8⁺ T cells (n=5); error bars are S.E.M, *p<0.05, **p<0.01, two-tailed t-test.

C. For the real-time human T cell activation, T1D patients and healthy controls (HC) CD8+ T cells were analyzed by Xfe96 Seahorse analyzer (n=4). T cells were activated with human αCD3 (4 μg/ml) and αCD28 (20 μg/ml) in the presence or absence of MOTS-c (10 μM); error bars are S.E.M, two-way ANOVA, #p<0.05 indicates significance between HC and T1D **p<0.01, #p<0.05,