Supplemental Data 3

A. Description of transgenic TCRs

	P25	P25-related	C7 TCR	C7-related
Specificity	Ag85b	Ag85b	ESAT6	ESAT6
$\text{CDR3}\beta \text{ sequence}$	CASSFESSAETLYF	CASSLESSAETLYF	CASSYWGGGQNTLYF	CASSSWGGGQNTLYF
TRBV	TCRBV16-01	TCRBV16-01	TCRBV04-01	TCRBV04-01
TRBJ	TCRBJ02-03	TCRBJ02-03	TCRBJ02-04	TCRBJ02-04
CDR3β length	14	14	15	15

B. Detection of P25 TCR and related sequence in polyclonal response to Ag85b.

Sample	P25	P25-related	TRBV16	TRBJ2-3	TRBJ2-7
Ag85b_10w1	None	0.12%	31.7%	3.0%	30.7%
Ag85b_10w2	None	None	34.7%	3.6%	29.3%
Ag85b_10w3	None	None	39.0%	4.6%	15.9%
Ag85b_w4L1	None	None	36.6%	11.6%	11.5%
Ag85b_w4L2	None	None	49.1%	27.4%	29.9%
Ag85b_w4L3	None	None	35.2%	None	17.9%
Ag85b_w4L4	None	None	59.8%	9.4%	30.1%
Ag85b_w4L5	None	4.73%	58.2%	7.9%	28.8%

C. Detection of C7 TCR and related sequence in polyclonal response to ESAT6.

	C7 TCR		C7-related	
Sample	frequency	Count	Row Labels	Count
ESAT6_10w1	0.0368%	1	None	0
ESAT6_10w2	None	0	None	0
ESAT6_10w3	0.0687%	1	None	0
ESAT6_10w4	0.0119%	1	0.229%	1
ESAT6_10w5	None	0	None	0
ESAT6_w4L1	None	0	None	0
ESAT6_w4L2	None	0	None	0
ESAT6_w4L3	None	0	None	0
ESAT6_w4L4	None	0	3.665%	1
ESAT6_w4L5	None	0	None	0

A. The transgenic TCRs used in this study. The P25-related and C7-related TCRs were closely related sequences detected in Mtb-infected mice, which had similar gene segment usage as P25 and C7, and closely related CDR3 β sequences. P25 does not contain the 'LEG' motif that we frequently detected in Ag85b-specific CD4 T cells. C7 contains the 'GGG' motif that we observed in ESAT6-specific CD4 T cells.

B. The frequency of the P25 and P25-related CDR3β sequence in the tetramer ⁺Ag8b-specific CD4 T cells. Also listed are the frequency of TRBV16, TRBJ2-3, and TRBJ2-7, which are frequently used by Ag85b-specific CD4 T cells. Note that the P25 CDR3β amino acid sequence was not detected in any of our samples.

C. The frequency of the C7 and C7-related CDR3β amino acid sequence and number of unique clonotypes (based on DNA sequence) among the tetramer ESAT6-specific CD4 T cells sequenced.