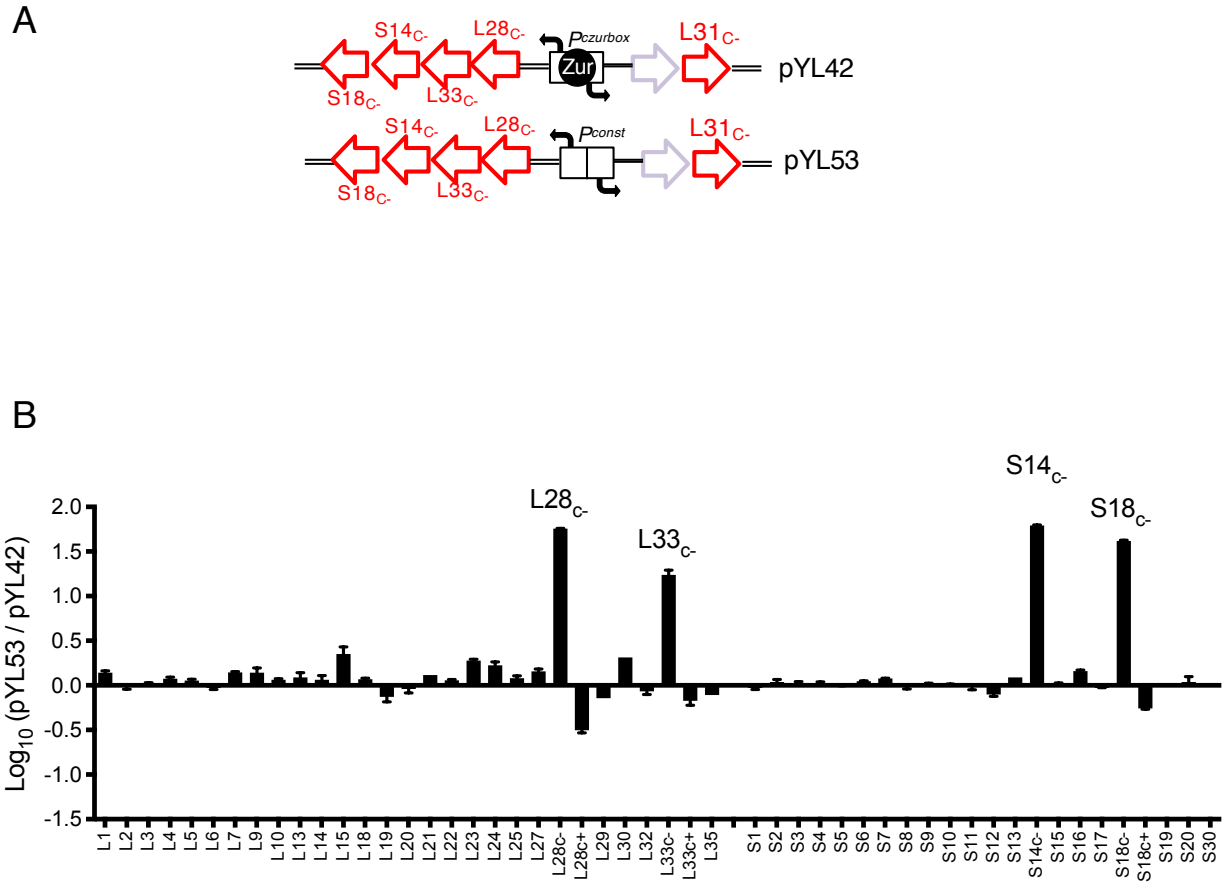

Figure S1. Incorporation of C- r-proteins in 70S ribosomes purified from high-zinc cultures of recombinant *M. smegmatis* cells constitutively expressing *c-* operon. **A.** Schematic representation of wild-type *c-* operon expressed by native (P^{zurbox}) promoter on pYL42 plasmid or an engineered constitutive promoter P^{const} on pYL53 plasmid. **B.** Relative abundance of each of the ribosomal protein in 70S ribosomes purified from high-zinc culture of mc^{2}155:*Δc-* strains transformed with either pYL42 or pYL53 plasmid.

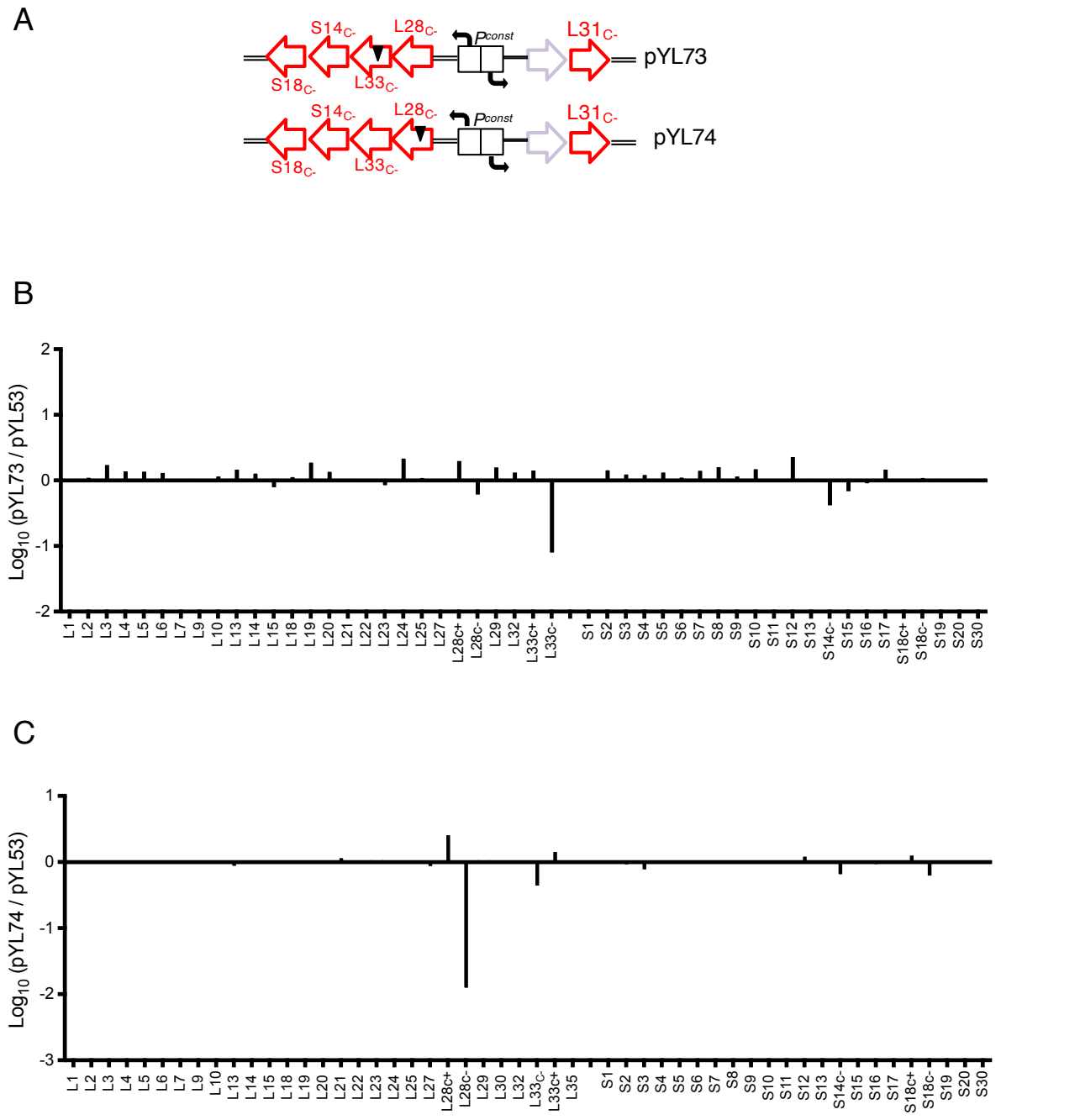

Figure S2. iTRAQ-MS based confirmation of protein composition of ribosomes from mutant strains (described in figure 3A) carrying in-frame deletion in L33c- (on pYL73 plasmid) and L28c- (on pYL74 plasmid) of the *c-* operon of *M. smegmatis.* It is to be noted that ribosomes from S14c- and S18c- deletion strains were confirmed previously (8). **A.** Schematic representation of the *c-* operon carrying in-frame deletion in L28c- (pYL74) and L33c- (pYL73) expressed from the P^{const} promoter. **B-C.** Relative abundance of each of the ribosomal protein in 70S ribosomes purified from high-zinc culture of mc^{2}155:*Δc-* strain transformed with either pYL73 or pYL74 plasmids. Ribosomes purified from high-zinc culture of mc^{2}155:*Δc-* strain transformed with pYL53 were used as reference.

Table S1: List of plasmids and strains used in this study

Plasmid	Remarks	Reference
pMH94	L5-attp-based integrative vector for mycobacteria; *kan*^{r}	(1)
pYUB854	Cosmid vector, *hyg*^{r}	(1)
pJV53-SacB	Sucrose-sensitive marker SacB cloned in pJV53 @ SpeI site; *kan*^{r}	(1)
pYL3	*kan*^{r} cassette in pMH94 @EcoRV & NotI sites is replaced by *hyg*^{r} cassette from pYUB854; *hyg*^{r}	(1)
pYL42	*MSMEG_6065-6070* including P^{zur}-box cloned in pYL3 @ SacI & XbaI; *hyg*^{r}	(1)
pYL53	*MSMEG_6065-6070* with zur box mutation in P^{zur}-box region (P^{const}) cloned in pYL3 @ SacI+XbaI, *hyg*^{r}	(1)
pYL60	P^{const} fused to *Rv2058-2055* ORF in pYL3 backbone @ SacI & XbaI; *hyg*^{r}	This study
pYL71	pYL53 with *Msmeg_6065* internal deletion (ΔS18c-) @ SacI & XbaI, *hyg*^{r}	(1)
pYL72	pYL53 with *Msmeg_6066* internal deletion (ΔS14c-) @ SacI & XbaI, *hyg*^{r}	(1)
pYL73	pYL53 with *Msmeg_6067* internal deletion (ΔL33c-) @ SacI & XbaI, *hyg*^{r}	This study
pYL74	pYL53 with *Msmeg_6068* internal deletion (ΔL28c-) @ SacI & XbaI, *hyg*^{r}	This study
pYL76	pYL53 with *Msmeg_6070* internal deletion (ΔL31c-) @ SacI & XbaI, *hyg*^{r}	This study
pYL97	*Rv2055c-2058c* with *Rv2056c* internal deletion (ΔS14c-) including P^{zur}-box cloned in pYL3 @ EcoRI & XbaI; *hyg*^{r}	This study
pYL230	pYL53 with S14c- (D67A), *hyg*^{r}	This study
pYL232	pYL53 with S14c-$_{(\Delta ins)}$, *hyg*^{r}	This study
pYL233	pYL53 with S14c- (R83A), *hyg*^{r}	This study
Strain	**Remarks**	**Reference**
mc^{2}155	high-frequency transformation strain of *M. smegmatis as wild-type*	(1)
mc^{2}155:Δmpy	*ΔMsmeg_1878* in mc^{2}155, *zeo*^{r}	(1)
mc^{2}155:Δc-	Unmarked Δ*Msmeg_6065-6070* in mc^{2}155	(1)
mc^{2}7000	*M. tuberculosis* H37Rv:*ΔRD1*:*ΔpanCD* as wild-type	(2)
mc^{2}7000:Δc-	Unmarked *ΔRv_2055c-2058c* in mc^{2}7000	This study

Table S2: List of primers used in this study.

Primer Name	Sequence	Used in:
Pmsc--SacIF	CTGCTCCAGCCAGAGCTCGGTGTCCACGCATGTCAC	pYL60
Pmsc--R	TGACTCTCCTTCGTCGTGCGGTACCG	
Rv2058F	CACGACGAAGGAGAGTCATTGTCCGCCCACTGCCAA	
Rv2055XR	CGGCTTGCCGGATCTAGATCATGGGCACAGTGCAGC	
6065Up200SF	AACCACCGACCA GAGCTCGAT CGAGCGCCGCGCCGC	pYL73, pYL74, pYL76, pYL230, pYL232, pYL233
6070Dn200XR	ATCGTCACGCACGGTGCACGAGACGCATCCGTGCAC	
6067-delete-R	CGCGAGGAACGCTGATGGCGAAGAAGTCGAAGATTG	pYL73
6067-delete-F	CCATCAGCGTTCCTCGCGCTCGTTGCGAGCCATCAG	
6068-delete-R	CGGGGGGAGAAGGTCTGATGGCTCGCAACGAGATCC	pYL74
6068-delete-F	TCAGACCTTCTCCCCCCGGCAGTGGGCCGACATTGA	
6070-delete-R	GATGCCGGGTTTCACGATTGCTCTCCTGTCTGGGGG	pYL76
6070-delete-F	ATCGTGAAACCCGGCATCTGACGGTCCACCTTAGGG	
Pmtbc--EF	CATTGGGGTCGTGAATTCCGTGTTCGTTGGGCGCGT	pYL97
pRv2055-2058XR	TCGCCTGGTGAGTCTAGAGTACGACTTCGACTCCCC	
Rv2056-F	GACGTG GCCAAGAAGTCCGTCCGGAAGGCCAGCTGG	
Rv2056-R	GGACTTCTTGGCCACGTCAGCGTTCCTCGCGAAAGT	
p6066-D67A-R	AACCGCGATGTCGTCGCCGGCCGACCACGCGGG	pYL230
p6066-D67A-F	CCCGCGTGGTCGGCCGGCGACGACATCGCGGTT	
p6066DelR14toR53F	CCGCTGCTCGTTCTTGACAATCTTC	pYL232
p6066DelR14toR53R	AAGAACGAGCAGCGGGACTCCAGTCCCGTG	
p6066-R83A-R	TTCGGGTTGTCCCGTGTGGCCGTGCGCGAGATGGCGCAT	pYL233
p6066-R83A-F	ATGCGCCATCTCGCGCACGGCCACACGGGACAACCCGAA	

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2. **Sambandamurthy VK, Derrick SC, Hsu T, Chen B, Larsen MH, Jalapathy KV, Chen M, Kim J, Porcelli SA, Chan J, Morris SL, Jacobs WR, Jr.** 2006. Mycobacterium tuberculosis DeltaRD1 DeltapanCD: a safe and limited replicating mutant strain that protects immunocompetent and immunocompromised mice against experimental tuberculosis. Vaccine **24:**6309-6320.