

**S1 Table. Peptide mass finger print analysis of host cell proteins coeluting with YopM-SBP-CBP**

protein	Mascot score	Expectation value	Peptide
DDX3X	99	1.7e-6	YIPPHLR; SDEDDWSKPLPPSER; LEQELFSGGNTGINFE; YTRPTPVQK; QYPISLVLAPTR; ELAVQIYEEAR; VRPCVVYGGAEIGQQI; GCHLLVATPGR; MLDMGFEPQIR; HTMMFSATFPK; EEALHQFR; SPILVATAVAAR; VGNLGLATSFNER
DDX3Y	119	1.6e-8	YIPPHLR; GKPNYFSDR; SDEDDWSKPLPPSER; LEQELFSGGNTGINFE; YTRPTPVQK; QYPISLVLAPTR; ELAVQIYEEAR; VRPCVVYGGADTVQQI; GCHLLVATPGR; MLDMGFEPQIR; HTMMFSATFPK; YACTSIHGDR; EEALHQFR; KPILVATAVAAR

**S1 Table:** The final elution fraction of YopM-SBP-CBP tandem affinity purified from infected murine J774A.1 macrophages [1] was subjected to SDS-PAGE. Visible Coomassie-stained bands were excised, enzymatically digested and masses of the resulting peptides were subsequently determined on MALDI-TOF mass spectrometer. Data analysis of the obtained peptide mass fingerprints was performed with Mascot (<http://www.matrixscience.com>) obtaining indicated scores. The peptide-mass-fingerprint analysis significantly identified DDX3X or -Y ( $p < 0.05$  at a score threshold of 55). Using this methodology, RSK 1, 2 and PKN 1, 2 were confirmed as binding partners of YopM-SBP-CBP in previous work [1]. In the same experiments peptides matching the protein sequence of DEAD box RNA helicase 3 (DDX3) were identified. DDX3X and DDX3Y are encoded by the X- and Y-chromosome, respectively [2]. The peptide mass fingerprint analysis revealed indicated expectation values matching the DDX3X- or DDX3Y protein sequence. The lower the expectation value, the more significant the score.

**References Supporting Information**

1. Hentschke M, Berneking L, Belmar Campos C, Buck F, Ruckdeschel K, Aepfelbacher M. Yersinia virulence factor YopM induces sustained RSK activation by interfering with dephosphorylation. *PloS one*. 2010;5(10).
2. Sekiguchi T, Iida H, Fukumura J, Nishimoto T. Human DDX3Y, the Y-encoded isoform of RNA helicase DDX3, rescues a hamster temperature-sensitive ET24 mutant cell line with a DDX3X mutation. *Experimental cell research*. 2004;300(1):213-22.