*Mycobacterium tuberculosis* binds human serum amyloid A, and the interaction modulates the colonization of human macrophages and the transcriptional response of the pathogen.

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Supplementary data:

**Figure S1.** SDS-PAGE analysis of the quality of *Mycobacterium tuberculosis* whole-cell protein extract used for isolation of the ligands binding human SAA1.

**Figure S2.** Western blot analysis of recombinant *Mycobacterium tuberculosis* rAtpA, rABC, rThiC, rEspB, rPpiA and rTB18.6 proteins developed in *E. coli* with anti-His Tag mouse monoclonal IgG1 antibodies.

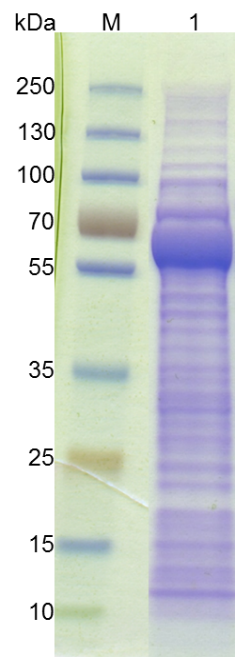
**Figure S3**. Intensity of human SAA1 binding by live *Mycobacterium tuberculosis* (*Mtb*) and *Mycobacterium smegmatis* (*Msmeg*) cells. BI-binding intensity index; C-control *Mtb* and *Msmeg* cells incubated in culture medium alone.

**Figure S4.** Inhibition of biotin-labeled human SAA1 binding by live *Mycobacterium smegmatis* cells in the presence of 1-fold (5 µg/ml) and 3-fold (15 µg/ml) excess of unlabeled homologous protein. C-control bacilli incubated with the addition of culture medium instead of unlabeled SAA1; the percent of inhibition is marked in red circle.

**Table S1.** Primer sequences used for PCR amplification of the gene sequences.

**Table S2.** *Mycobacterium tuberculosis* SAA1 binding protein and peptide list identified by LC/ESI-MS/MS.

**Table S3.** RNAseq data base (see Excel file)



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**Table S1.** Primer sequences used for PCR amplification of the gene sequences.

|  |  |  |  |
| --- | --- | --- | --- |
| Amplified region | Primer | Sequence (5’→3’)a | Product size [bp] |
| *atpA* sense | *rv1308*/BamHI | cggatccgATGGCTGAGTTGACAATCCCCGC | 1650 |
| *atpA* reverse | *rv1308*/BamHI | cggatccCTATTTCTTCTTCTTCGGCGCCGG |
| *abc* sense | *rv2477c*/EcoRI | cgaattcATGGCTGAGTTCATCTACACGATGAAGAAG | 1677 |
| *abc* reverse | *rv2477c*/HindIII | caagcTTAGCCGCGCGTCAGCTTGC |
| *tb18.6* sense | *rv2140c*/BamHI | cggatccgATGACAACTTCACCCGACCCG | 531 |
| *tb18.6* reverse | *rv2140c*/HindIII | caagcttCTAACGCTGCTCGTAGGTGCC |
| *espB* sense | *rv3881c*/EcoRI | cgaattcATGACGCAGTCGCAGACCGTG | 1383 |
| *espB* reverse | *rv3881c*/HindIII | caagctTCACTTCGACTCCTTACTGTC |
| *thiC* sense | *rv0423c*/HindIII | caagcttATGACCATCACCGTTGAACCG | 1644 |
| *thiC* reverse | *rv0423c*/HindIII | caagctTCACTGGGTTATCGGGAGATAC |
| *ppiA* sense | *rv0009*/BamHI | cggatccgATGGCAGACTGTGATTCCGTG | 549 |
| *ppiA* reverse | *rv0009*/ HindIII | caagctTCAGGAGATGGTGATCGAC |

**a** the restriction enzyme recognition sites are underlined

**Table S2.** *Mycobacterium tuberculosis* SAA1 binding protein and peptide list identified by LC/ESI-MS/MS.

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Fasta Headers** | **Proteins** | **Peptides fa** | **Peptides fb** | **Peptides mtb** | **Mol. Weight [kDa]** | **Sequence Length** | **Sequence Coverage fa [%]** | **Sequence Coverage fb [%]** | **Sequence Coverage mtb [%]** | **Intensity** | **Intensity fa** | **Intensity fb** | **Intensity mtb** |
| >Rv0384c\_clpB | 1 |  |  | 1 | 92.535 | 848 |  |  | 0.9 | 1.29E+08 |  |  | 1.29E+08 |
| >Rv1308\_atpA | 1 | 18 |  | 3 | 59.288 | 549 | 39 |  | 6.2 | 3.60E+08 | 3.35E+08 |  | 2.51E+07 |
| >Rv1310\_atpD | 1 |  |  | 1 | 53.094 | 486 |  |  | 2.5 | 3.55E+06 |  |  | 3.55E+06 |
| >Rv1650\_pheT | 1 |  |  | 1 | 88.373 | 831 |  |  | 1.2 | 1.04E+05 |  |  | 1.04E+05 |
| >Rv0440\_groEL2 | 1 | 13 | 1 |  | 56.726 | 540 | 27.4 | 3,3 |  | 1.18E+08 | 1.18E+08 | 1.50E+05 |  |
| >Rv1133c\_metE | 1 | 4 | 1 |  | 81.548 | 759 | 6.6 | 1,3 |  | 5.19E+06 | 5.06E+06 | 1.31E+05 |  |
| >Rv0350\_dnaK | 1 | 22 |  |  | 66.83 | 625 | 39 |  |  | 3.69E+08 | 3.69E+08 |  |  |
| >Rv2477c\_Rv2477c ABCtransport EttA/ChvD | 1 | 16 |  |  | 61.892 | 558 | 32.6 |  |  | 1.72E+08 | 1.72E+08 |  |  |
| >Rv2455c\_Rv2455c | 1 | 15 |  |  | 69.15 | 653 | 31.2 |  |  | 1.48E+08 | 1.48E+08 |  |  |

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Fasta headers** | **Proteins** | **Peptides** | **Mol. weight [kDa]** | **Sequence length** | **PEP** | **Sequence coverage SAA1 [%]** | **Sequence coverage SAA2 [%]** | **Sequence coverage SAA3 [%]** | **Intensity SAA1** | **Intensity SAA2** | **Intensity SAA3** |
| >Rv0440\_groEL2 | 1 | 17 | 56.726 | 540 | 5.99E-214 | 44,4 | 20,9 |  | 3,95E+08 | 1,04E+07 |  |
| >Rv3881c\_espB | 1 | 10 | 47.593 | 460 | 1.19E-55 |  | 33 |  |  | **2,50E+07** |  |
| >Rv0009\_ppiA | 1 | 7 | 19.239 | 182 | 6.15E-94 |  | 12.6 | 54.4 |  | 3,92E+05 | **1,96E+09** |
| >Rv2140c\_TB18.6 | 1 | 6 | 18.634 | 176 | 6.21E-54 |  | 9.1 | 63.6 |  | 2,02E+05 | **2,01E+08** |
| >Rv1617\_pykA | 1 | 6 | 50.667 | 472 | 3.92E-35 |  | 16.1 |  |  | 9,12E+06 |  |
| >Rv0423c\_thiC | 1 | 6 | 59.897 | 547 | 7.93E-85 | 15.2 | 13 |  | **6,36E+06** | 4,27E+06 |  |
| >Rv0957\_purH | 1 | 5 | 55.026 | 523 | 2.74E-46 |  | 20.1 |  |  | **3,01E+07** |  |
| >Rv2858c\_aldC | 1 | 5 | 48.258 | 455 | 9.60E-30 |  | 11 |  |  | 2,98E+06 |  |
| >Rv3417c\_groEL1 | 1 | 5 | 55.877 | 539 | 5.44E-17 | 15.4 | 2.2 |  | 4,37E+06 | 3,23E+05 |  |
| >Rv3841\_bfrB | 1 | 4 | 20.442 | 181 | 3.37E-47 |  |  | 28.2 |  |  | 2,77E+07 |
| >Rv3442c\_rpsI | 1 | 3 | 16.436 | 151 | 1.21E-05 |  |  | 19.2 |  |  | 4,14E+06 |
| >Rv3418c\_groES | 1 | 3 | 10.772 | 100 | 3.05E-05 | 9 | 15 | 32 | 3,60E+05 | 9,30E+05 | 3,83E+06 |
| >Rv2831\_echA16 | 1 | 3 | 26.63 | 249 | 6.54E-08 |  |  | 18.5 |  |  | 2,64E+06 |
| >Rv3583c\_Rv3583c | 1 | 3 | 17.907 | 162 | 9.21E-15 |  |  | 20.4 |  |  | 2,28E+06 |
| >Rv1829\_Rv1829 | 1 | 2 | 18.114 | 164 | 0.000194 |  |  | 14.6 |  |  | 5,71E+06 |
| >Rv0580c\_Rv0580c | 1 | 2 | 18.035 | 163 | 1.87E-05 |  |  | 18.4 |  |  | 3,17E+06 |
| >Rv0054\_ssb | 1 | 2 | 17.321 | 164 | 1.14E-05 |  |  | 12.8 |  |  | 2,08E+06 |
| >Rv0020c\_fhaA | 1 | 2 | 56.88 | 527 | 4.69E-07 |  |  | 4.7 |  |  | 1,32E+06 |
| >Rv1484\_inhA | 1 | 2 | 28.528 | 269 | 7.24E-06 | 3,7 | 3.7 | 3 |  | 4,02E+05 | 5,55E+05 |
| >Rv0379\_secE2 | 1 | 2 | 7.9661 | 71 | 1.88E-22 |  | 49.3 | 18.3 |  | 9,06E+06 | 2,94E+05 |
| >Rv0685\_tuf | 1 | 2 | 43.561 | 396 | 6.32E-28 |  | 8.1 |  |  | 3,35E+06 |  |
| >Rv2213\_pepB | 1 | 2 | 53.449 | 515 | 2.17E-08 |  | 4.7 |  |  | 1,85E+06 |  |
| >Rv0753c\_mmsA | 1 | 2 | 54.453 | 510 | 1.00E-13 |  | 6.5 |  |  | 1,27E+06 |  |
| >Rv2251\_Rv2251 | 1 | 2 | 55.77 | 529 | 2.88E-06 | 8,5 |  |  | **5,62E+06** |  |  |
| >Rv1284\_canA | 1 | 1 | 18.157 | 163 | 1.58E-17 |  |  | 15.3 |  |  | 1,45E+06 |

|  |
| --- |
| common background |
| identified by => 2 peptides |
| best candidates |