

## **Supplementary Notes**

### **Direct binding of *M. leprae* to recombinant extracellular domain of ErbB2**

A close association of *M. leprae* with ErbB2 receptor clustering on Schwann cell membrane raised the possibility that *M. leprae* may directly interact with ErbB2 and induce receptor phosphorylation. To examine this, we prepared recombinant extracellular domain of ErbB2 and performed an *in vitro* binding assay in ELISA. The extracellular domain comprises of a signal peptide (amino acids 1-21) within the region of amino acids 1-652. We amplified this domain by RT-PCR using human Schwann cell total RNA as a template and the resulting PCR fragment was cloned into the Eam1104 I site of the pDUAL expression vector to obtain the pDUAL<sup>ErbB2ex</sup> construct. The expression, purity and the activity of the His-tagged recombinant (r) extracellular ErbB2 domain (rErbB2ex) after transient transfection of COS-7 cells with pDUAL<sup>ErbB2ex</sup> was confirmed by total protein staining, immunoblotting with antibodies to His-Tag and the N-terminal/extracellular domain of ErbB2, and by ELISA using anti-ErbB2 antibody Herceptin<sup>TM</sup> (Trastuzumab; 4D5) respectively <sup>1, 2</sup> (**Suppl. Figs. 2a, b**). Dose-dependent binding of *M. leprae* to rErbB2ex (1µg) was determined by ELISA. Mean values of nonlinear regression analysis from four independent experiments revealed that  $3.75 \pm 1.066 \times 10^6$  *M. leprae* were sufficient to produce specific binding to rErbBex (**Suppl. Fig. 2c**). Alternatively, various concentrations of rErbB2ex were incubated with *M. leprae* ( $3 \times 10^6$ ), and non-linear regression analysis from four separate experiments showed the high affinity binding of soluble rErbB2ex to *M. leprae* (KD:  $4.16 \pm 0.8$  nM) (**Suppl. Fig. 2d**). To further confirm the specificity of direct *M. leprae* binding to rErbB2ex, we blocked ErbB2 activity with the anti-ErbB2 monoclonal antibody Herceptin<sup>TM</sup> (Trastuzumab) (4D5) that specifically abolish ErbB2 biologic activity and is being used in treatment of breast cancers overexpressing ErbB2 <sup>1, 2</sup>. The binding of *M. leprae* ( $1 \times 10^8$ /ml) was significantly inhibited when rErbB2ex was preincubated with 1µg of Herceptin<sup>TM</sup> antibody (**Fig. 2e**), and thus confirming the binding specificity of *M. leprae* to rErbB2ex.

### **References:**

1. Yarden, Y. & Sliwkowski, M.X. Untangling the ErbB signalling network. *Nat Rev Mol Cell Biol* **2**, 127-37 (2001).
2. Hynes, N.E. & Lane, H.A. ERBB receptors and cancer: the complexity of targeted inhibitors. *Nat Rev Cancer* **5**, 341-54 (2005).