Supplementary figure legends

Figure S1. Functional validation of the KBM7 reporter cells for probing the TLR3 pathway. A, KBM7 cells are not responsive to poly(I:C). Cells were transduced with a cassette encoding RFP under a NF-κB dependent promotor and stimulated with poly(I:C) (25 μg/ml) or TNF-α (10 ng/ml) for 16 h. NF-κB activity of KBM7 cells was monitored by FACS. B, KBM7 cells response to poly(I:C) exposure is dependent on TLR3 expression and endosomal acidification for poly(I:C) detection. IL-8 and IP-10 production were measured in the supernatant from KBM7 complemented or not with TLR3 cDNA, stimulated or not with poly(I:C) (25 μg/ml) for 16 h, with or without 45 min of pre-treatment with 20 nM ConB. C, NF-κB activity of KBM7 cells complemented with TLR3 was monitored by FACS. Cells were transduced with a cassette encoding dscGFP under a NF-κB dependent promotor and stimulated with poly(I:C) (25 μg/ml) or TNF-α (10 ng/ml) for 16 h, with or without 45 min of pre-treatment by 20 nM ConB.

Figure S2. Results of the genetic screen. -log(*p* Value) of the results of sgRNA enrichments identified by sequencing, corresponding to 19 genes. After 1, 2 or 3 rounds of enrichment, the DNA from the enriched population was harvested, and enriched sgRNAs were identified by sequencing and comparison to an unsorted library. This determination was carried out for the two independent screens performed in parallel, generating in total 12 cell populations enriched. -Log(p-value) are presented when > 3 for at least 5 of the 12 comparisons. Each dot represents the gene enrichments calculated from one condition. The RSA algorithm was used to identify the significantly enriched genes targeted in the selected cells. Red: known key members of TLR3 pathway, blue: genes further studied.