

Figure S1

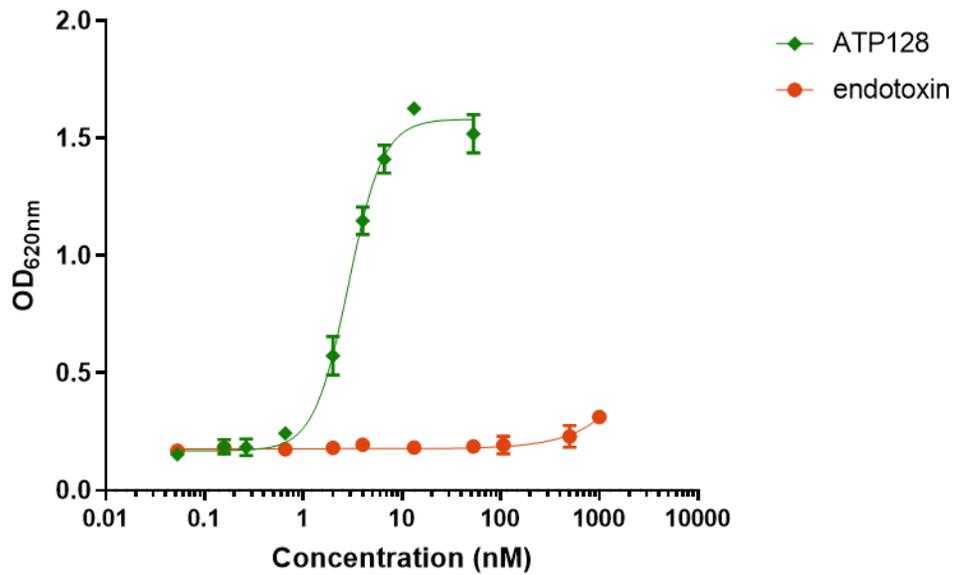


Figure S1. Endotoxin level comparable to the one present in the constructs do not activate NF- κ B pathway. THP-1 MD2-CD14 cells were incubated with increasing concentrations of endotoxin. ATP128 vaccine was used as a positive control. After 18 hours, supernatant was recovered, and SEAP activity was measured by QUANTI-Blue assay.

Table S1

Table S1 **Endotoxin levels in ATP128 variants**

Protein	Endotoxin level
ATP128 FL	<0.3 EU/mg
ATP128 w/o Anaxa	1.15 EU/mg
ATP128 w/o CPP	2.49 EU/mg

FL, full length

Endotoxin levels of ATP128, ATP128 w/o Anaxa and ATP128 w/o CPP proteins

Figure S2

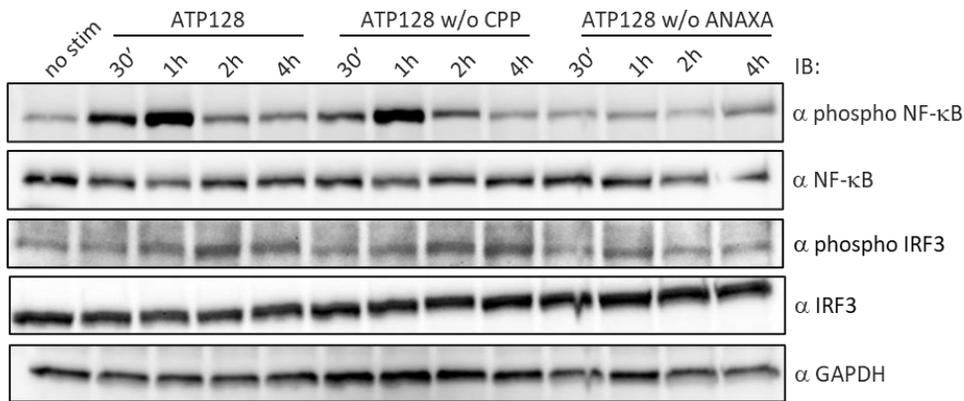


Figure S2. NF- κ B and IRF3 phosphorylation induced by ATP128 constructs. THP-1 Dual cells were stimulated with 300 nM of various ATP128 constructs at different time points and phosphorylation of NF- κ B and IRF3 was assessed through Western Blot using specific phospho-antibodies. Anti-GAPDH was used as normalizer.

Figure S3

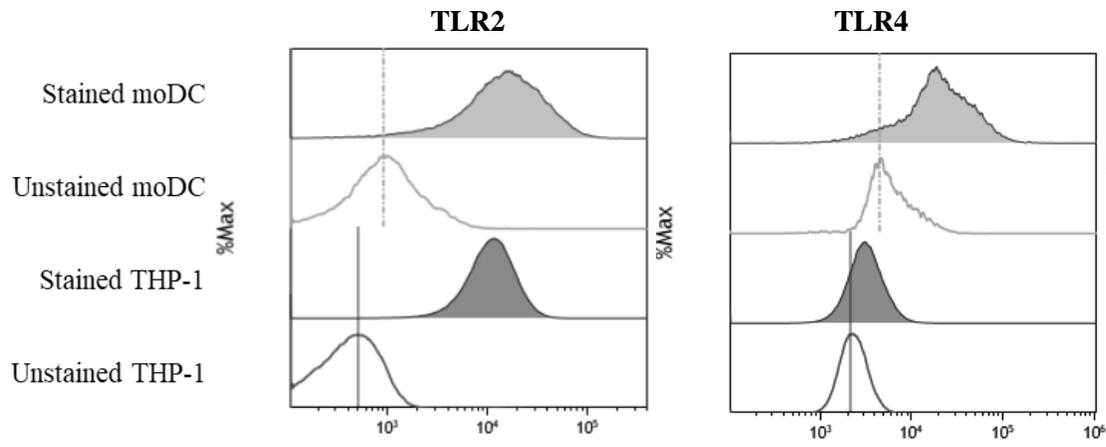


Figure S3. THP-1 cell line and human moDCs express different levels of TLR2 and TLR4. THP-1 cell line and human moDCs were assessed for the surface expression of TLR2 and TLR4.

Figure S4

