

Title: TNF- α and Poly(I:C) induction of A20 and activation of NF- κ B signaling are independent of ABCF1 in human airway epithelial cells

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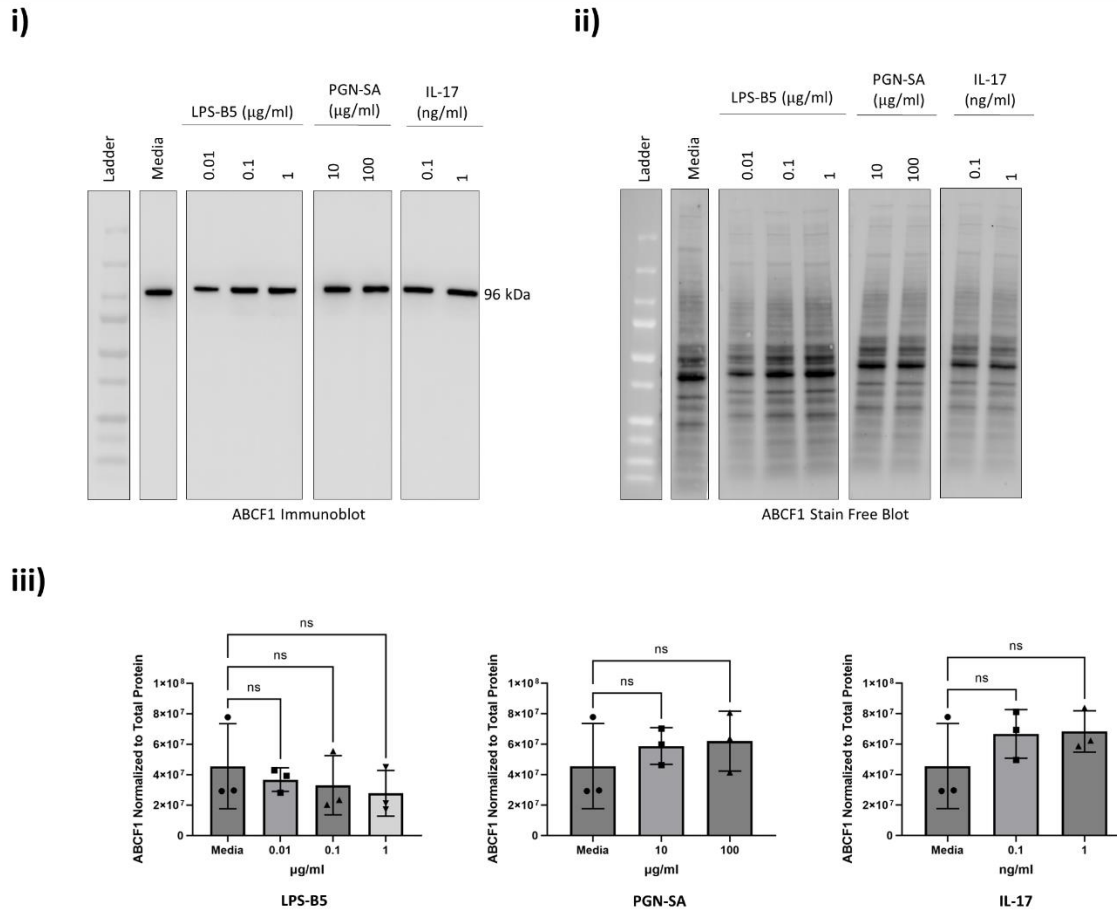
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Supplementary Figures

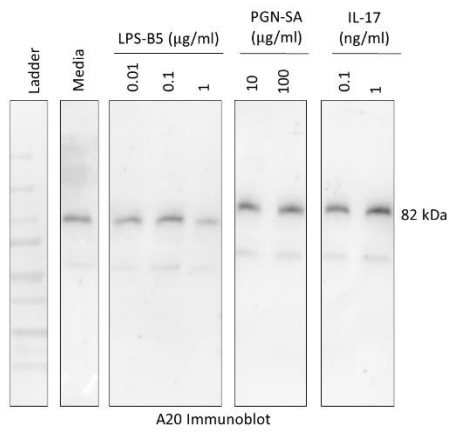


Supplementary Figure 1 – Immunostimulatory challenges on ABCF1 expression in HBEC-6KT *in vitro*.

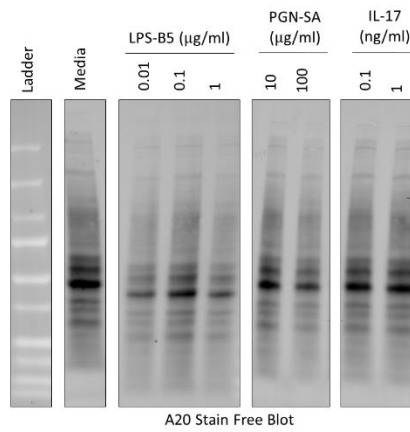
i) Immunoblot of ABCF1 protein expression with IL-17, LPS-B5, and PGN-SA dosage-based stimulation in HBEC-6KT cells. **ii)** Stain-free blot was used to **iii)** quantify ABCF1 protein expression.

All studies n = 3; ns > 0.05

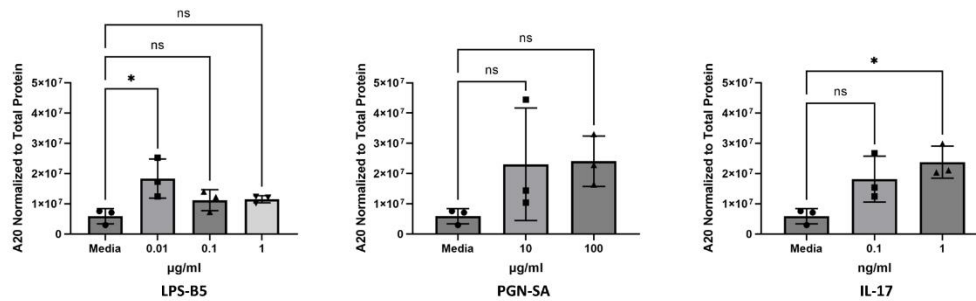
i)



ii)



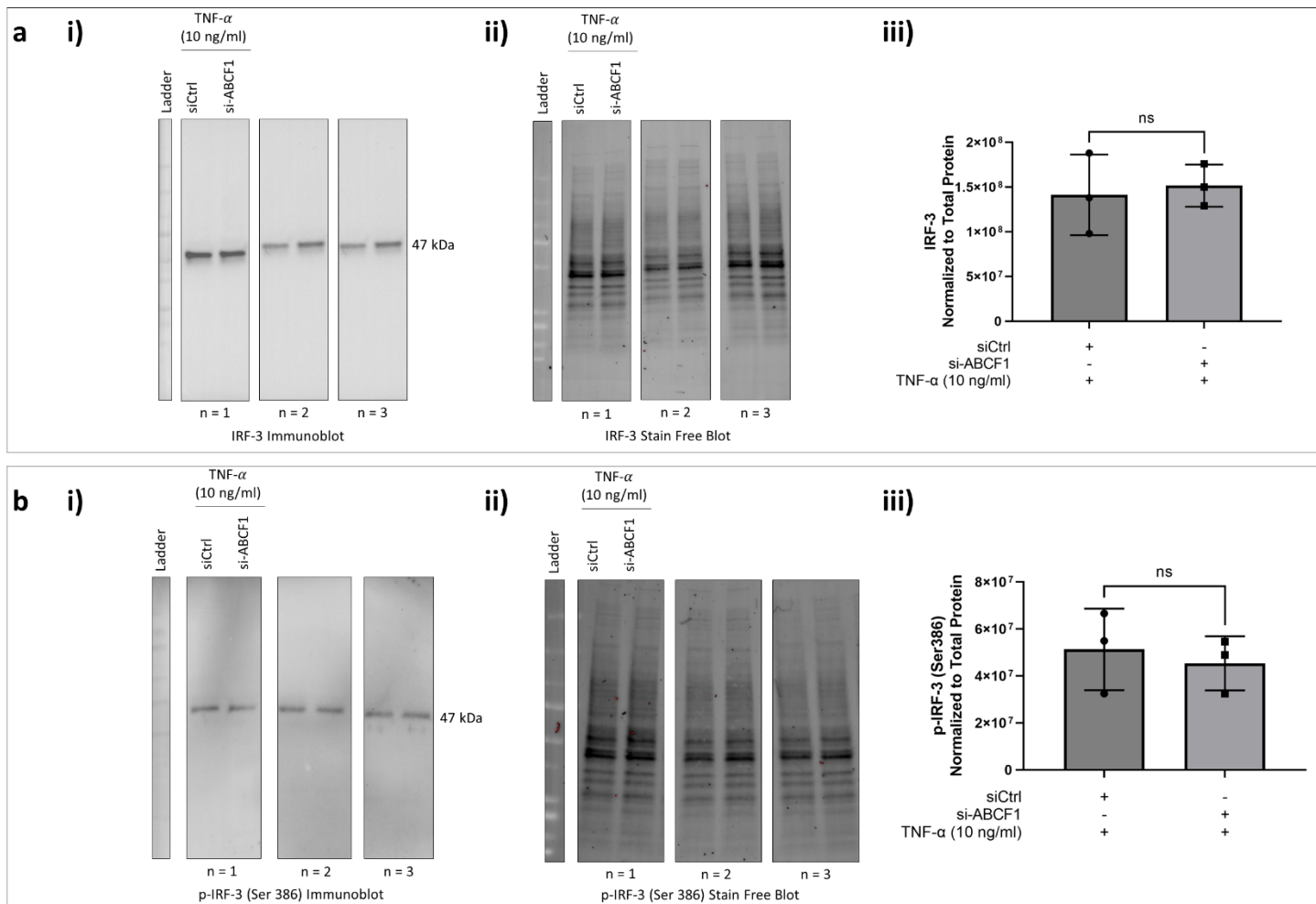
iii)



Supplementary Figure 2 – Immunostimulatory challenges on A20 expression in HBEC-6KT *in vitro*.

i) Immunoblot of A20 protein expression with IL-17, LPS-B5, and PGN-SA dosage-based stimulation in HBEC-6KT cells. ii) Stain-free blot was used to

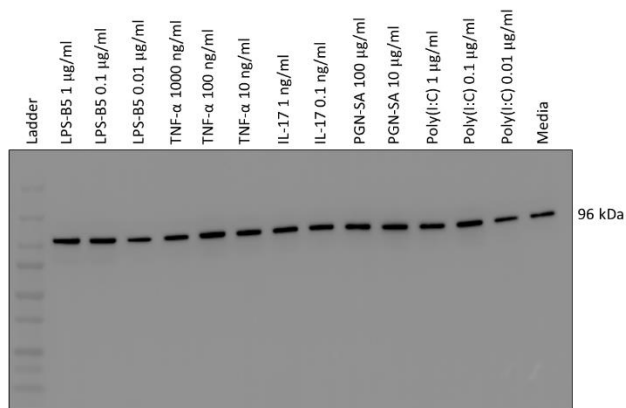
iii) quantify A20 protein expression.
All studies n = 3; ns > 0.05



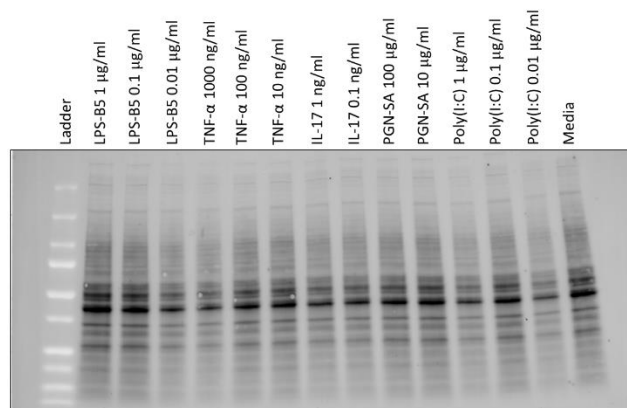
Supplementary Figure 3 – ABCF1 silencing on IRF-3 protein expression and activation under TNF- α challenge in HBEC-6KT *in vitro*.

a) Immunoblot with quantification of total IRF-3, and **b)** phosphorylated IRF-3 (Ser 386) protein expression in HBEC-6KT cells under TNF- α (10 ng/ml) challenge with and without si-ABCF1 treatment. All studies $n = 3$; * $p \leq 0.05$

a



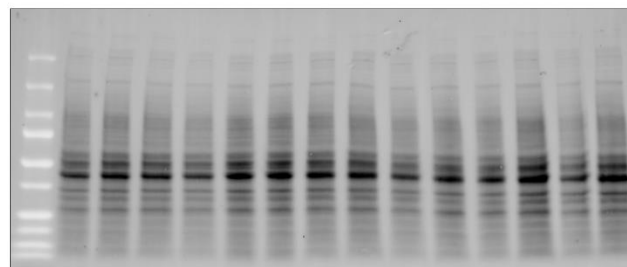
n=1
ABCf1 Immunoblot



n = 1
ABCf1 Stain Free Blot



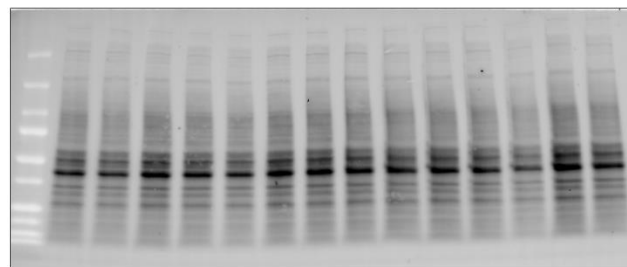
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ABCf1 Immunoblot



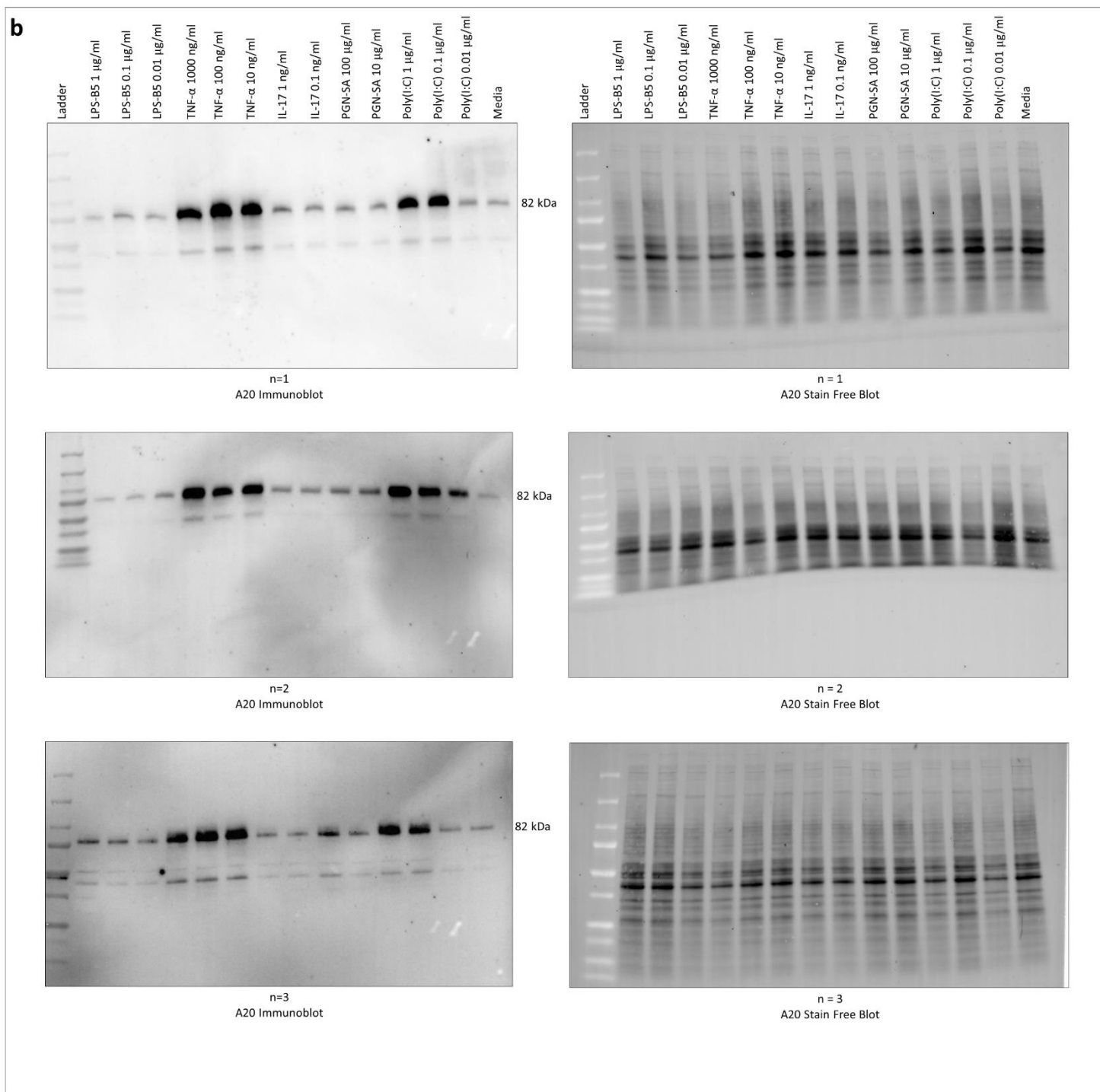
n = 2
ABCf1 Stain Free Blot



n=3
ABCf1 Immunoblot

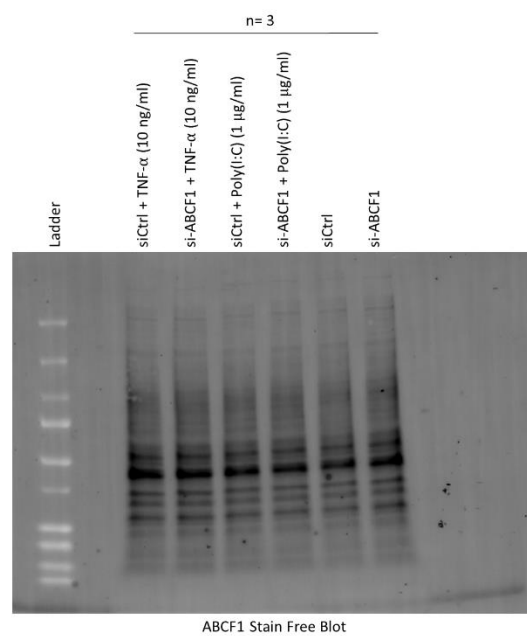
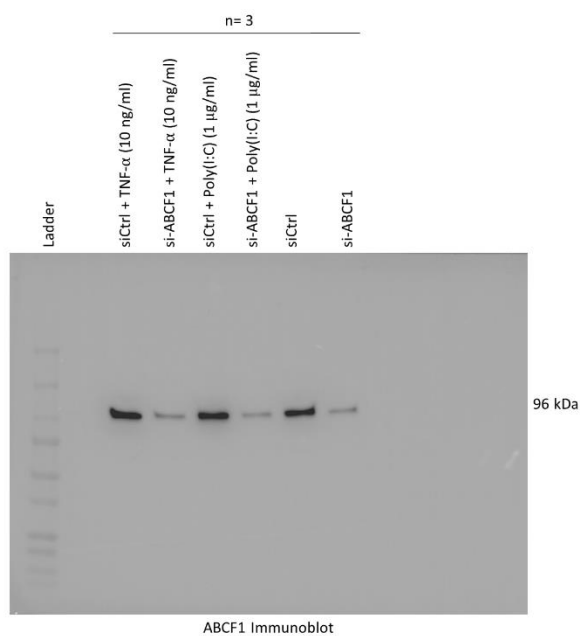
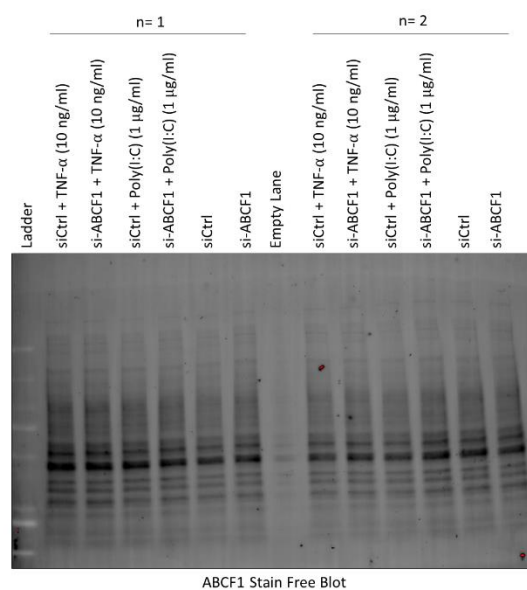
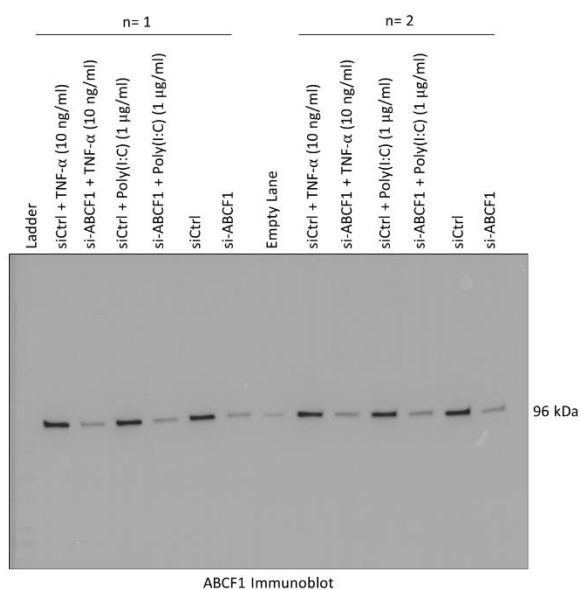


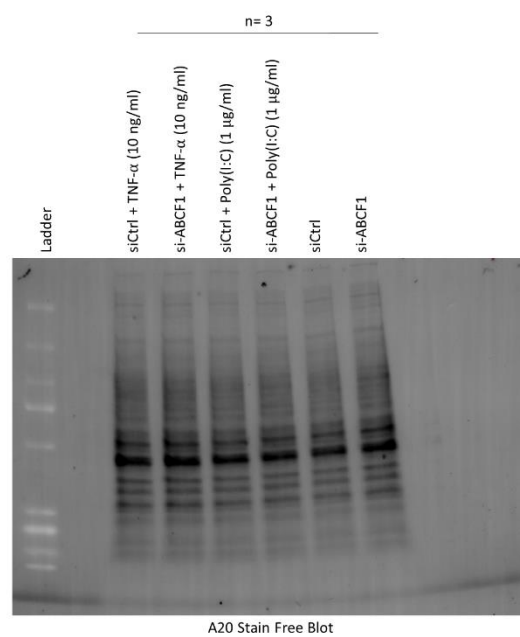
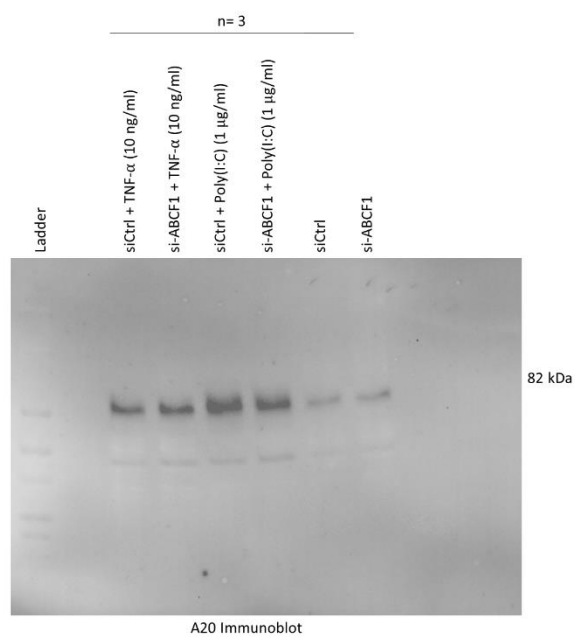
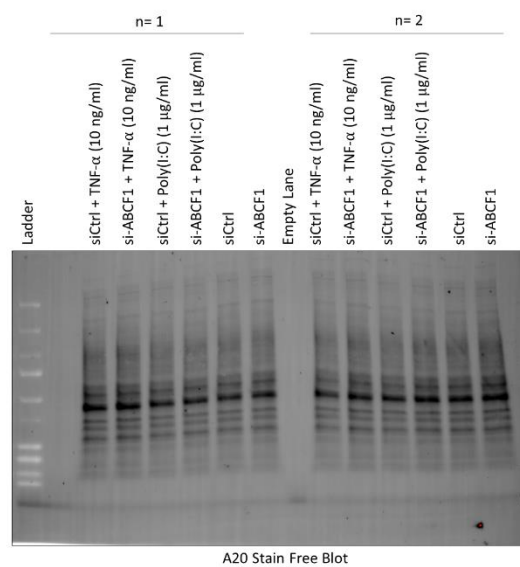
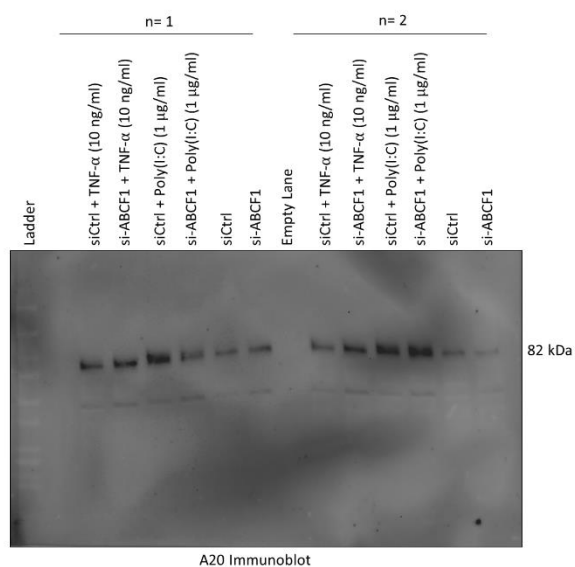
n = 3
ABCf1 Stain Free Blot



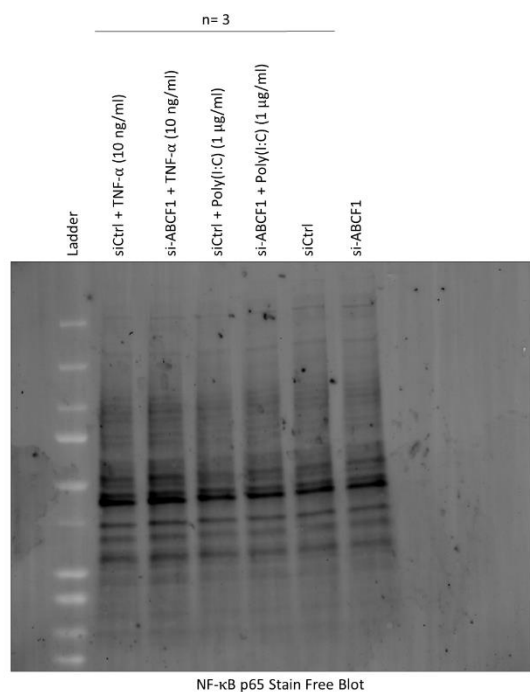
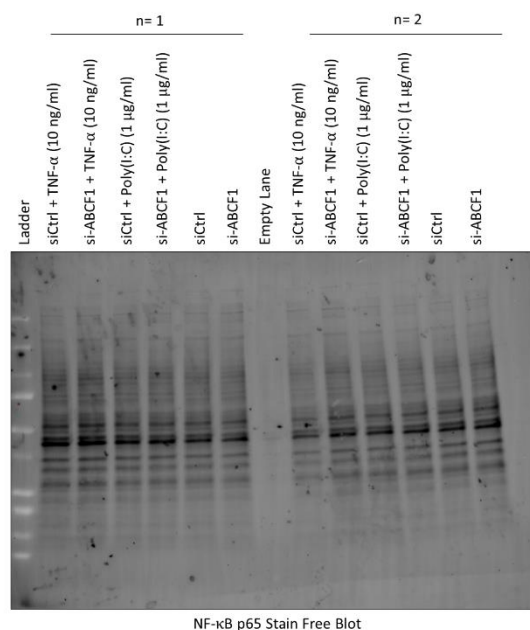
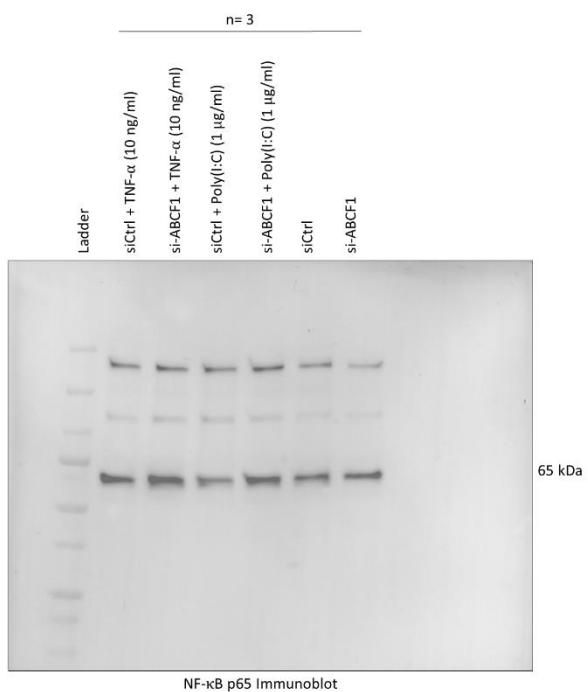
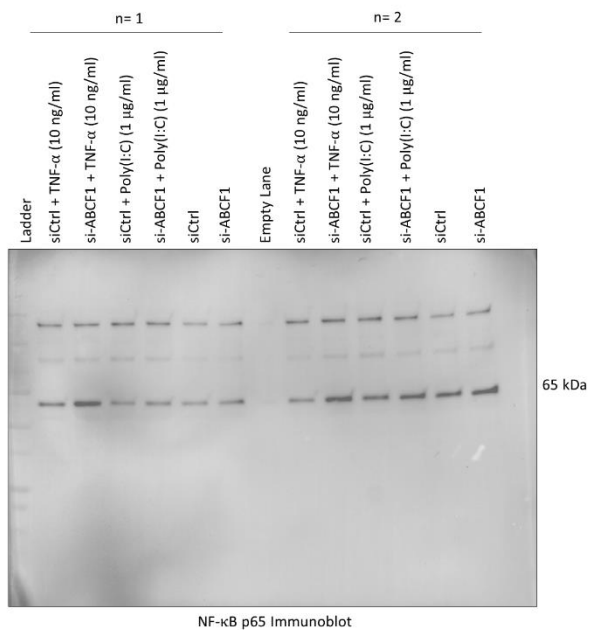
Supplementary Figure 4 – Full length immunoblots of **a)** ABCF1 and **b)** A20 in HBEC-6KT under immunostimulatory challenges *in vitro*.

a

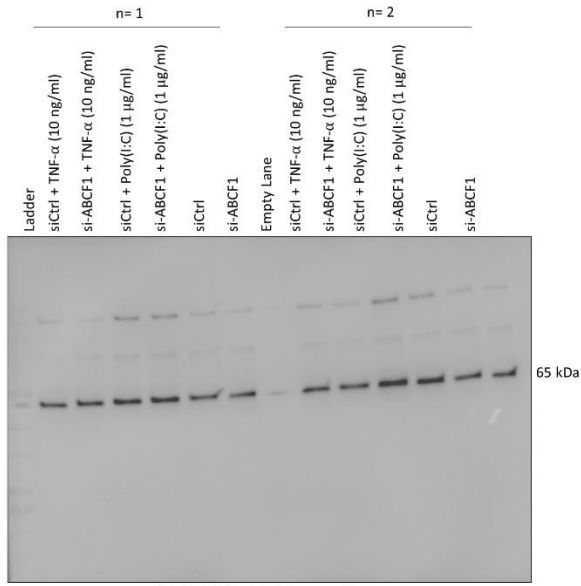


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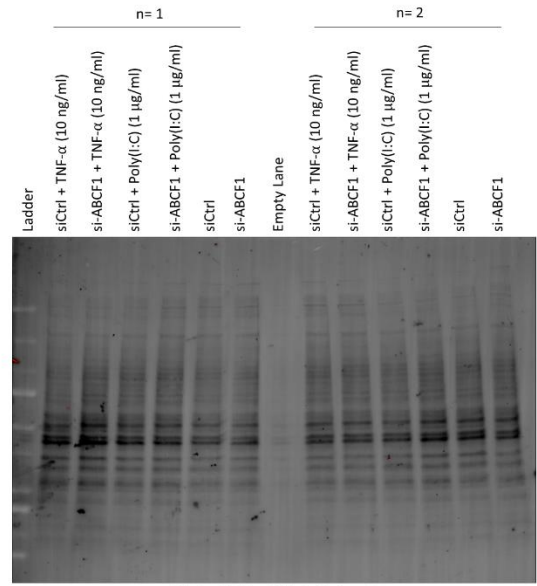
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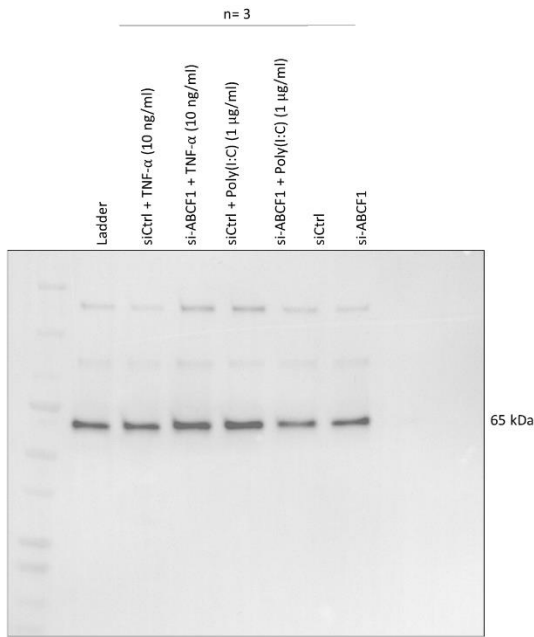
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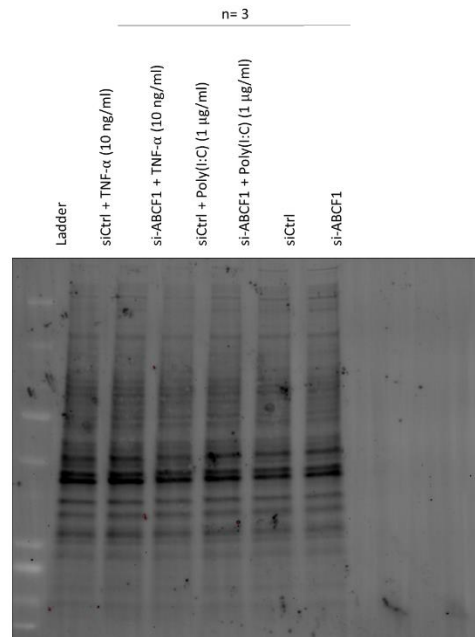
p-NF- κ B p65 (Ser 536) Immunoblot



p-NF- κ B p65 (Ser 536) Stain Free Blot

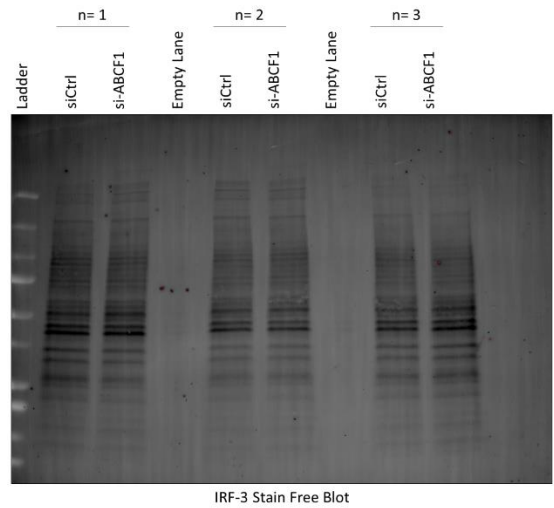
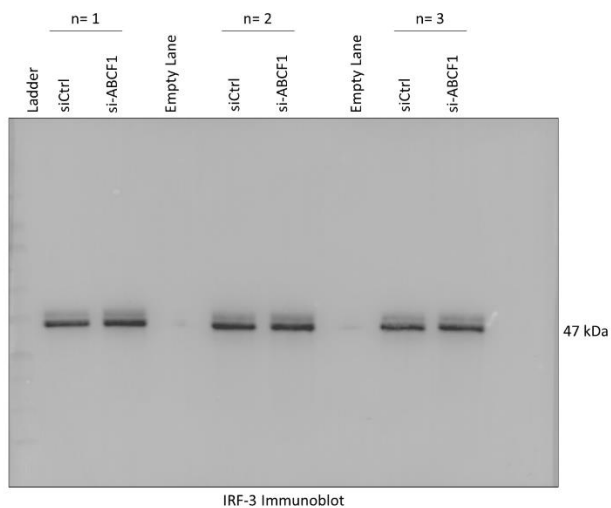
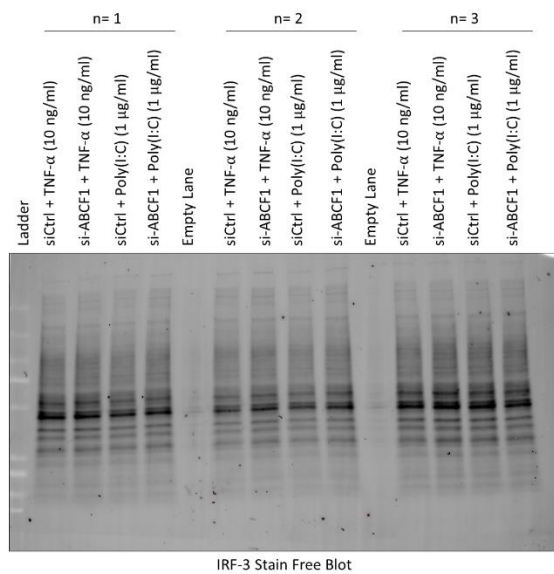
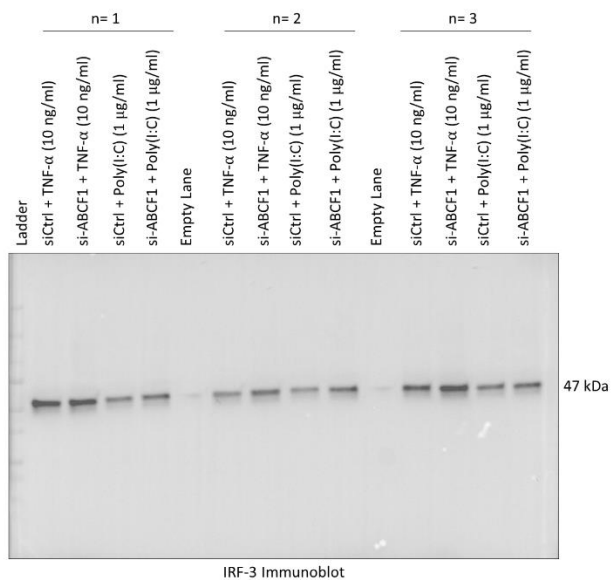


p-NF- κ B p65 (Ser 536) Immunoblot

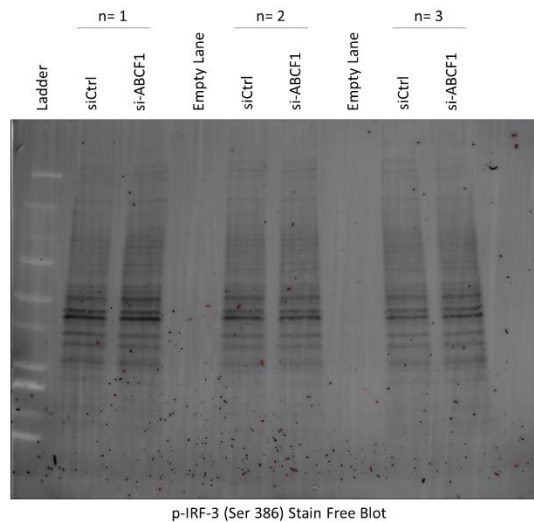
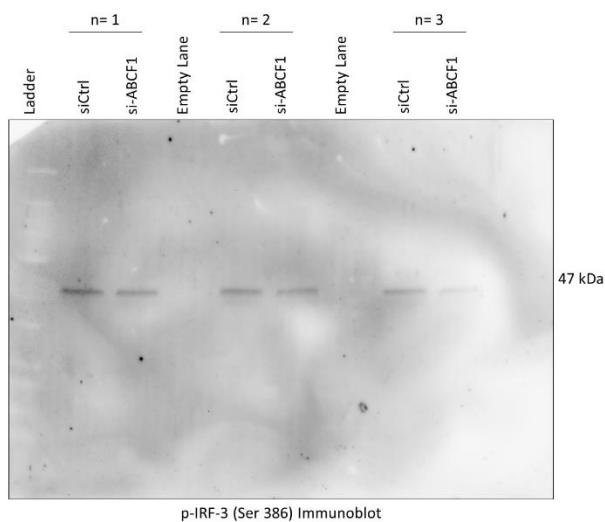
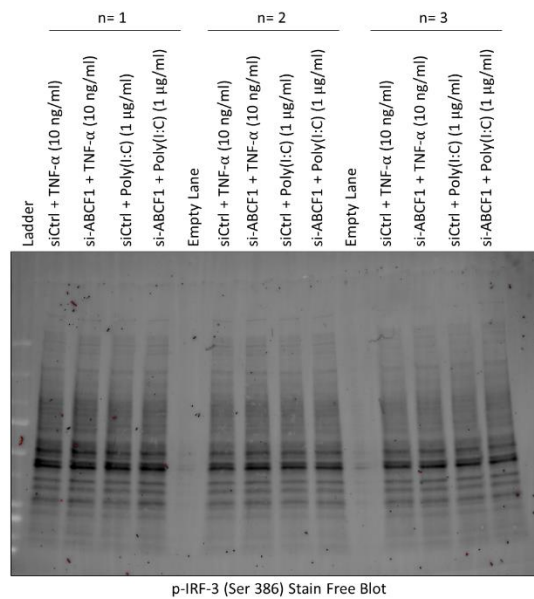
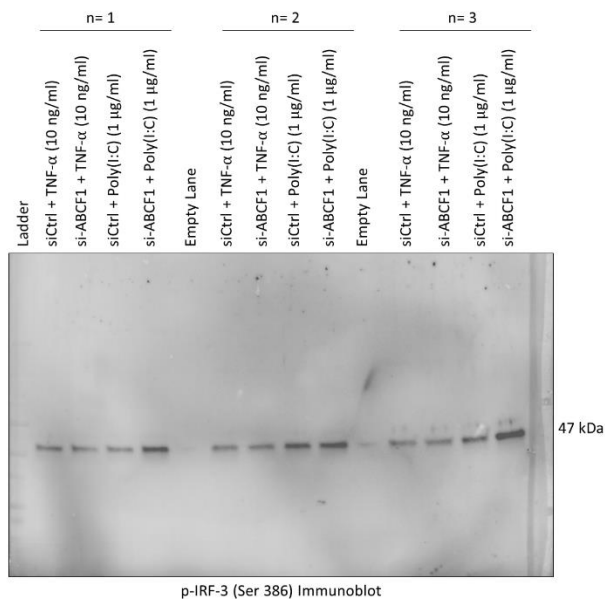


p-NF- κ B p65 (Ser 536) Stain Free Blot

e



f



Supplementary Figure 5 – Full length immunoblots of **a)** ABCF1, **b)** A20, **c)** NF- κ B p65, **d)** Phosphorylated NF- κ B p65 (Ser 536), **e)** IRF-3, and **f)** Phosphorylated IRF-3 (Ser 386) in HBEC-6KT under immunostimulatory challenges with siCtrl and si-ABCF1 treatment *in vitro*.