

Supplement

Figure S1. Pseudopterostin inhibits activation of NF- κ B after two different stimuli. Cells are stably transfected with a NF- κ B-Luc reporter gene. PsA-D treatment was performed for 20 minutes following 1 μ g/ml LPS or 6 ng TNF α incubation for 1 hour. Error bars were calculated using +SEM; n=3. Three stars show a significance of p<0.001.

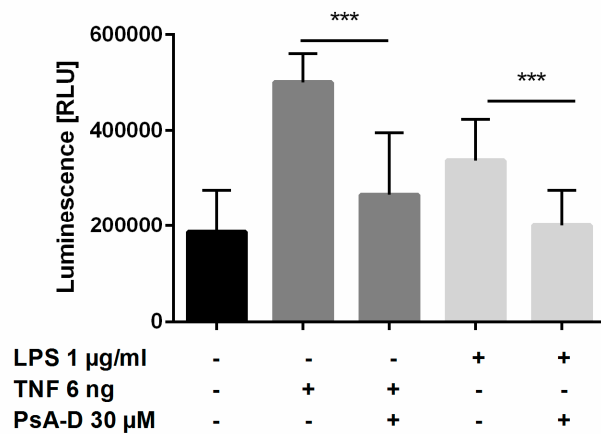


Figure S2. PsA-D blocked phosphorylation of NF- κ B/p65 and I κ B α . MDA-MB-231 cells were seeded in a 10 cm dish. Treatment with 30 μ M PsA-D for 15 minutes was followed with treatment of 6 ng TNF α for 15 minutes. Cells were lysed and protein concentration was measured with Bradford Reagent. A total protein amount of 0.8 mg/ml was used. Control cells were treated with DMSO in the same amounts as PsA-D. Error bars were calculated using +SEM; n=3. P-values of three stars show a significance of p<0,001.

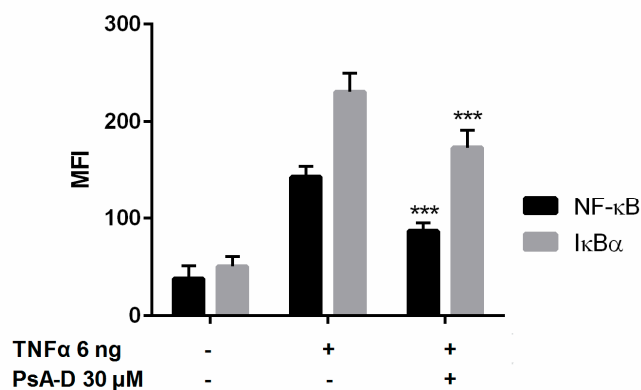


Table S1. Inhibition of cytokine release in MDA-MB-453 triple negative breast cancer cells. MDA-MB-453 cells were seeded at a density of 6×10^6 cells per ml. 30 μ M PsA-D was incubated for 20 minutes followed by 20 ng/ml TNF α . Cytokine amounts were analyzed in supernatants after 24 hours incubation time. No treatment serves as a control. Values were normalized to TNF α treatment and set to 100 % \pm SD. % inhibition reflects the percentage of the amount of cytokines reduced by PsA-D treatment compared to TNF α . P-values were analyzed according to student's t-test; n=3.

MDA-MB-453	Control %	+TNF α %	+PsA-D %	p-value	% Inhibition
IL-8	4,7 (\pm 5,5)	100,5 (\pm 24,9)	51,7 (\pm 14,0)	0,012	48,9
MCP-1	17,3 (\pm 11,6)	97,4 (\pm 7,9)	87,2 (\pm 14,1)	0,43	10,2