

Supplementary Materials:

The Novel Small-molecule Annexin-A1 Mimetic, Compound 17b, Elicits Vasoprotective Actions in Streptozotocin-induced Diabetic Mice

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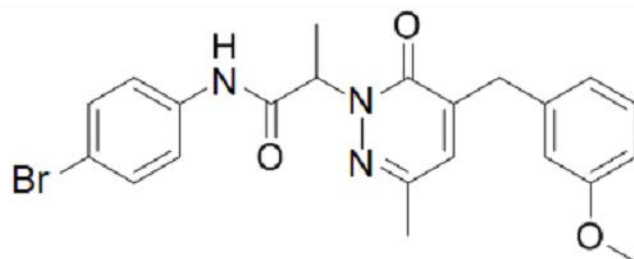
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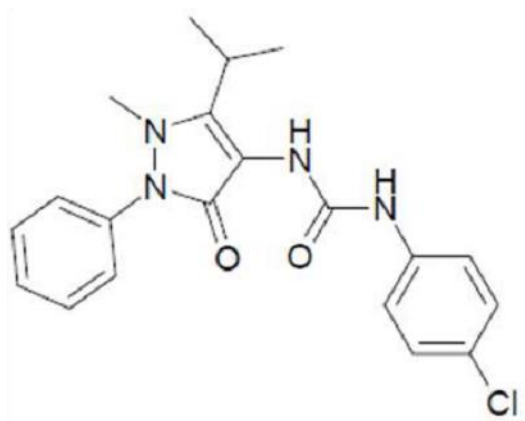
Supplementary Figure

A



Compound 17b (Cmpd17b)

B



Compound 43 (Cmpd43)

Supplementary Figure S1: Crystalline structure of (A) Cmpd17b and (B) Cmpd43.

Supplementary Table S1: Primer sequences for the quantitative amplification of murine genes using SYBR green.

Gene	Accession Number	Oligo Type	Primer Sequence 5'–3'	Amplicon Length
<i>Rn18S</i>	NR_003278	Forward	TG TTCACCATGAGGCTGAGATC	67 bp
		Reverse	TGGTTGCCTGGGAAAATCC	
<i>Fpr1</i>	NM_013521	Forward	CCTTGGCTTTCTTCAACAGC	247 bp
		Reverse	GCACAGTGGA ACTCAAAGCA	
<i>Fpr2</i>	NM_008039	Forward	ACAGCAGTTGTGGCTTCCTT	192 bp
		Reverse	CCTGGCCCATGAAAACATAG	
<i>Fpr3</i>	NM_008042	Forward	GGGCAACTCTGTTGAGGAAA	201 bp
		Reverse	GAAGGAAGCCACA ACTCCTG	

Supplementary Table S2: Primer and probe sequences for the quantitative amplification of murine genes using TaqMan.

Gene	Accession Number	Oligo Type	Primer Sequence 5'–3'	Amplicon Length
<i>18S</i>	NR_003278	Forward	GCATGGCCGTTCTTAGTTGG	67 bp
		Reverse	TGCCAGAGTCTCGTTCGTTA	
		Probe	TGGAGCGATTTGTCTGGTTATTCCGA	
<i>Fpr1</i>	NM_013521	Forward	TCTTCCTGATTGCCCTCATT	121 bp
		Reverse	GCACAAATCCAGGGTACGAT	
		Probe	GTGAGCCTAGCCAAGAAGGTAA	
<i>Fpr2</i>	NM_008039	Forward	TGATTGCTCTCATTGCCTTG	62 bp
		Reverse	GTTCTGAGCCCAGACTGGAT	
		Probe	GACCGCTGCATTTGTGTTCT	
<i>Ptgs1</i>	NM_008969	Forward	TCGGTCCTGCTCGCAGAT	95 bp
		Reverse	AGGCCAAAGCGGACACAGA	
		Probe	CACCAGTCAATCCCTGTTGTTA	
<i>Ptgs2</i>	NM_011198	Forward	TCCTCCCGTAGCAGATGAC	158 bp
		Reverse	TGCTGGGCAAAGAATGCAAAC	
		Probe	GGGAAATAAGGAGCTTCCTGAT	
<i>Ptgis</i>	NM_008968	Forward	GGGGCAGATACGTCACTGTT	78 bp

		Reverse	GAAGATGGCATAGGGATGGA	
		Probe	TTACGACACAGTGGTGTGGG	
<i>Ptgir</i>	NM_008967	Forward	GCTCGTTTGTACCGACCTCT	121 bp
		Reverse	TGAGTGAAGCCTCGGATCAT	
		Probe	GGCACGAGAGGATGAAGTTT	
<i>Tnf</i>	NM_001278601	Forward	GGCTGCCCCGACTACGT	66 bp
		Reverse	TTTCTCCTGGTATGAGATAGCAAATC	
		Probe	TCACCCACACCGTCAG	
<i>Mcp-1 (Ccp1)</i>	NM_011333	Forward	GTCTGTGCTGACCCCAAGAAG	62 bp
		Reverse	TGGTTCCGATCCAGGTTTTTA	
		Probe	AATGGGTCCAGACATAC	
<i>Icam-1</i>	NM_010493	Forward	GGAGGTGGCGGGAAAGTT	72 bp
		Reverse	TCCAGCCGAGGACCATACAG	
		Probe	CTGAAAGCTCTCCACCTC	
<i>IL1β</i>	NM_008361	Forward	TCGTGCTGTCGGACCCATA	93 bp
		Reverse	CTTGTACAAAGCTCATGGAGAATATCAC	
		Probe	CTGAAAGCTCTCCACCTC	

Supplementary Table S3: Vascular reactivity of small molecule FPR agonists, Cmpd17 and Cmpd43 in the mouse. Values are expressed as mean \pm S.E.M; *n*, aorta from individual mice. ACh, acetylcholine; Cmpd17b, compound 17b; Cmpd43, compound 43; DMSO, placebo control; E_{max} , maximum constriction relative to %U46619; pEC_{50} , sensitivity; R_{max} , maximum relaxation (negative values represent the reversal of relaxation). ND = not determined. *Significantly different from DMSO control (by one-way ANOVA, Tukeys *post hoc* test).

	DMSO			Cmpd17b			Cmpd43		
	<i>n</i>	pEC_{50}	R_{max}	<i>n</i>	pEC_{50}	R_{max}	<i>n</i>	pEC_{50}	R_{max}
<i>Direct vasodilation</i>	5	ND	-6 \pm 7	5	5.09 \pm 0.03	62 \pm 5*	5	ND	-2 \pm 9
<i>(FPR agonists)</i>									
<i>ACh</i>	5	6.82 \pm 0.15	93 \pm 2	5	6.95 \pm 0.19	96 \pm 1	5	7.04 \pm 0.09	93 \pm 3
	<i>n</i>	pEC_{50}	E_{max}	<i>n</i>	pEC_{50}	E_{max}	<i>n</i>	pEC_{50}	E_{max}
<i>U46619</i>	5	7.65 \pm 0.06	112 \pm 1	5	7.22 \pm 0.06*	96 \pm 4*	5	7.51 \pm 0.11	108 \pm 3