

SUPPLEMENTAL INFORMATION:

Bile acid analogues are activators of Pyrin inflammasome

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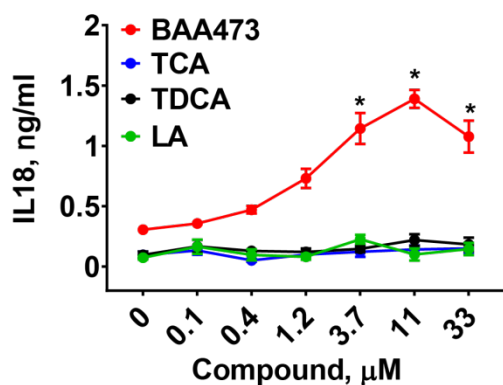
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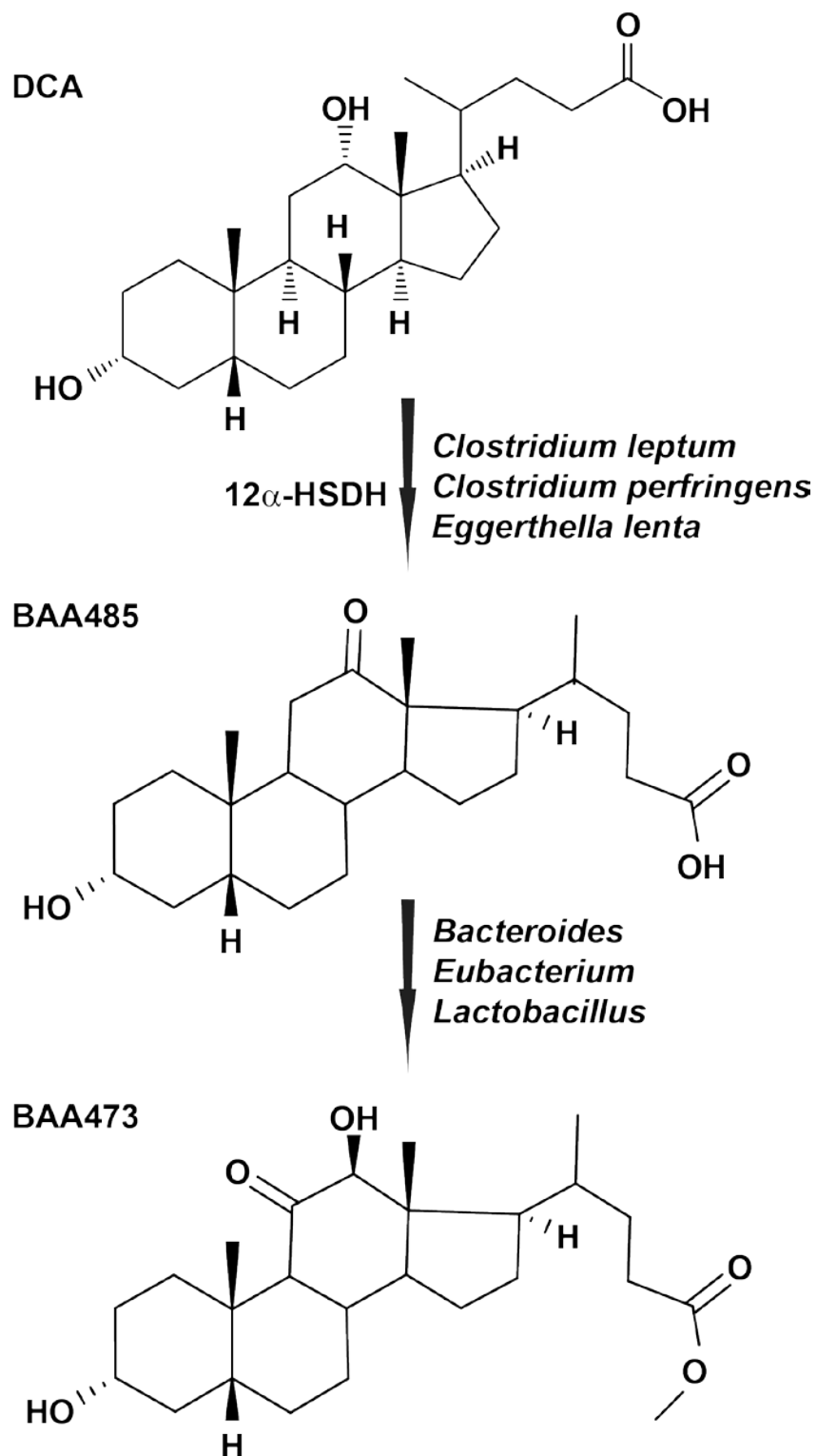
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Supplemental Figure 1. Primary bile acids such as Taurocholic acid (TCA), Taurochenodeoxycholic acid (TCDA), and Lithocholic acid (LA) do not activate the inflammasome pathway in PBMCs. PBMCs were primed with 0.1 ng/ml LPS overnight followed by compound treatment for 16 h and AlphaLISA detection of secreted levels of IL18 (A). Data presented is mean \pm SEM (n=3), *p value <0.0001 (one-way ANOVA).

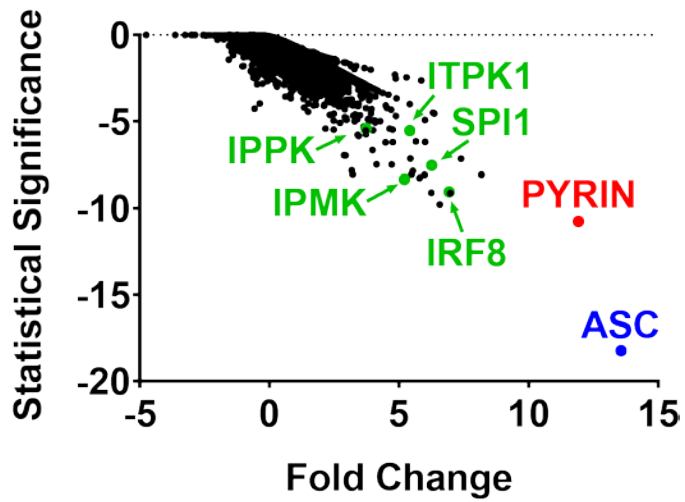
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Supplemental Figure 2. Hypothetical two-step transformation synthesis of BAA485 and BAA473 from DCA by gut bacteria.



Supplemental Figure 3. More detailed annotation of the BAA473 CRISPR screen which also appears as Figure 4D. Gene-centric visualization is shown of average log₂ fold change versus significance score. Inositol phosphate signaling pathway and IFN transcription factor complex hits are highlighted in green.



Supplemental Table 1. Enriched gene sets identified by MSigDB analysis in the BAA473 CRISPR screen.

GO Biological Process	Genes	Number of Genes in Gene Set	P-value (-Log 10)
Inositol Trisphosphate Kinase Activity	<i>IPMK, ITPK1</i>	11	4.1
Inositol Phosphate Metabolic Process	<i>IPPK, IPMK, ITPK1</i>	56	4.4
Alcohol Metabolic Process	<i>CEBPA, IPPK, CLN3, IPMK, ITPK1</i>	348	4.2
Defense Response To Other Organism	<i>PYCARD, IRF8, SPN, EIF2S1, SLC35C1, REP15</i>	505	4.5
Immune System Development	<i>SPI1, SIN3A, NCOA6, CEBPA, IRF8, CDC42, SPN</i>	582	5.2
Regulatory Region Nucleic Acid Binding	<i>CXXC1, SPI1, RBBP5, SIN3A, CEBPA, IRF8, MEF2D</i>	818	4.3
Regulation of Immune System Process	<i>PAXIP1, SPI1, SIN3A, PHB2, PYCARD, CDC42, SPN, EIF2S1, SLC35C1, REP15</i>	1403	5.2

Supplemental Table 2. Individual sgRNAs sequences selected for validation experiments according to sgRNA sequences used in the study.

Gene	gRNA	sgRNA sequence
<i>MEFV</i>	Pyrin-1	GCAGCTGGACCTGCTTCAGG
<i>MEFV</i>	Pyrin-2	GGTGCCAGAACTGCCTCG
<i>PYCARD</i>	ASC-1	GAACTTCTTGAGCTCCTCGG
<i>PYCARD</i>	ASC-2	GTTCAAGCTGAAGCTGCTGT
CTRL0002	Control-1	GACCGGAACGATCTCGCGTA
EGFP	Control-2	GGGCGAGGAGCTGTTACCG