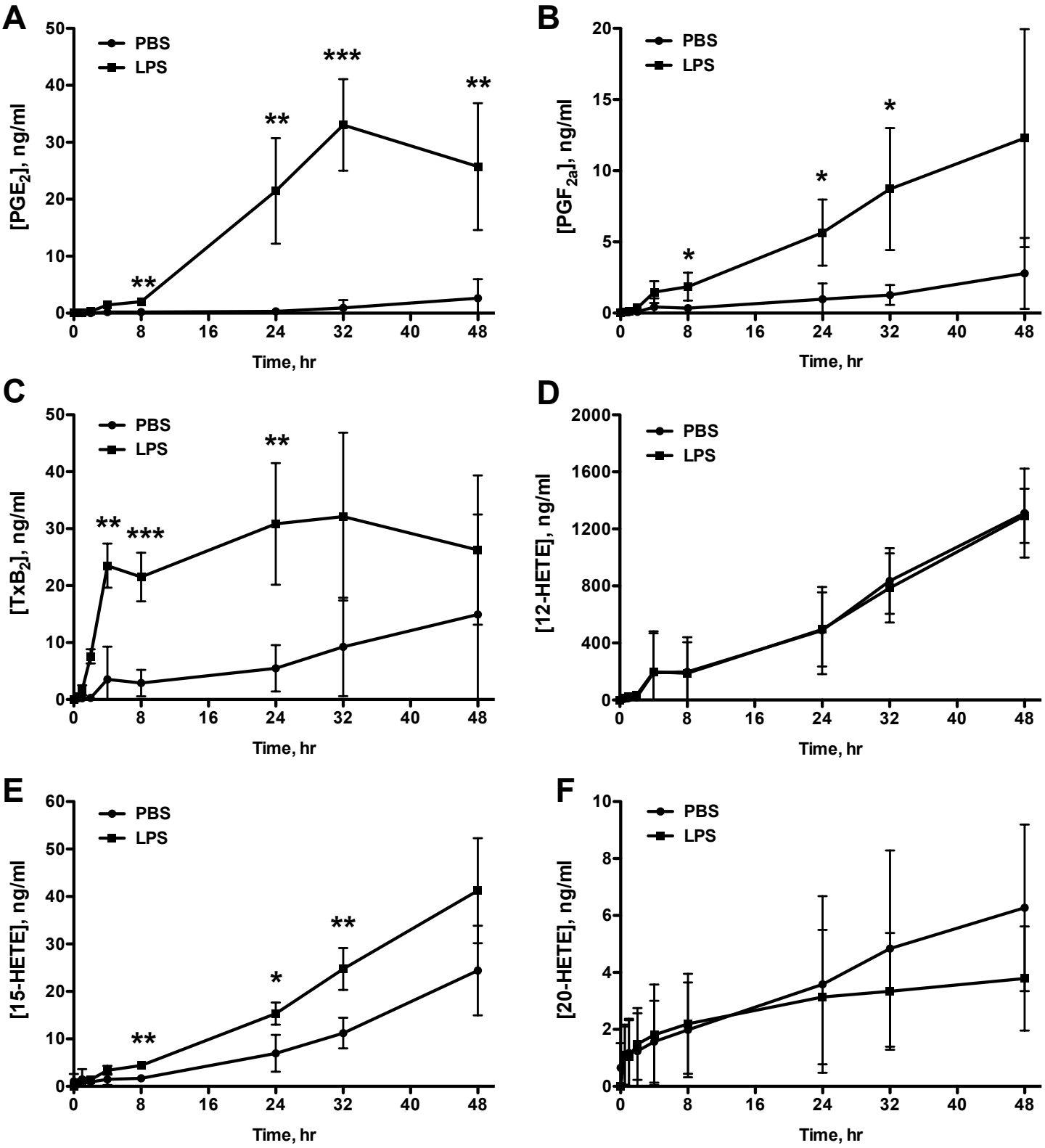
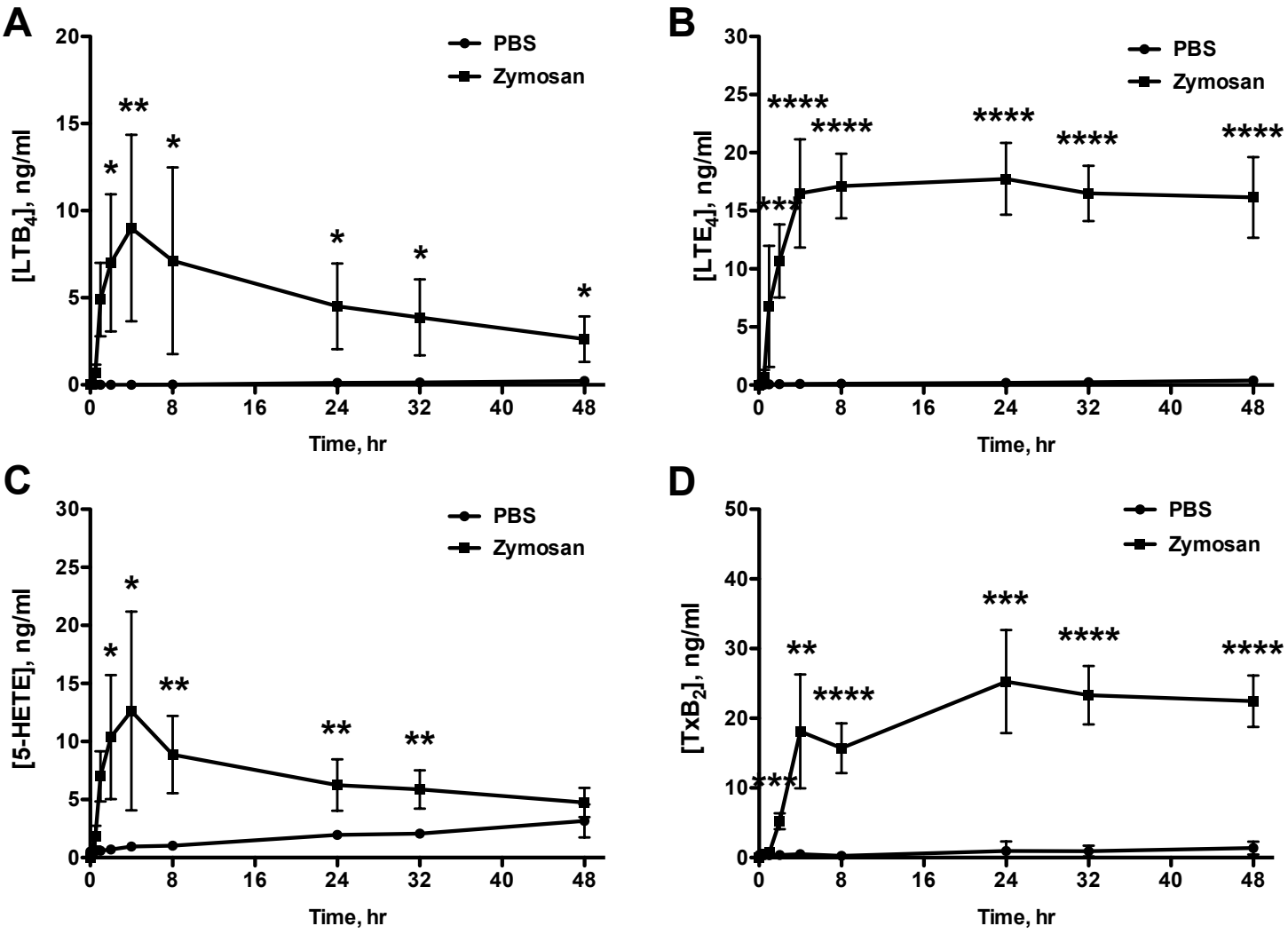
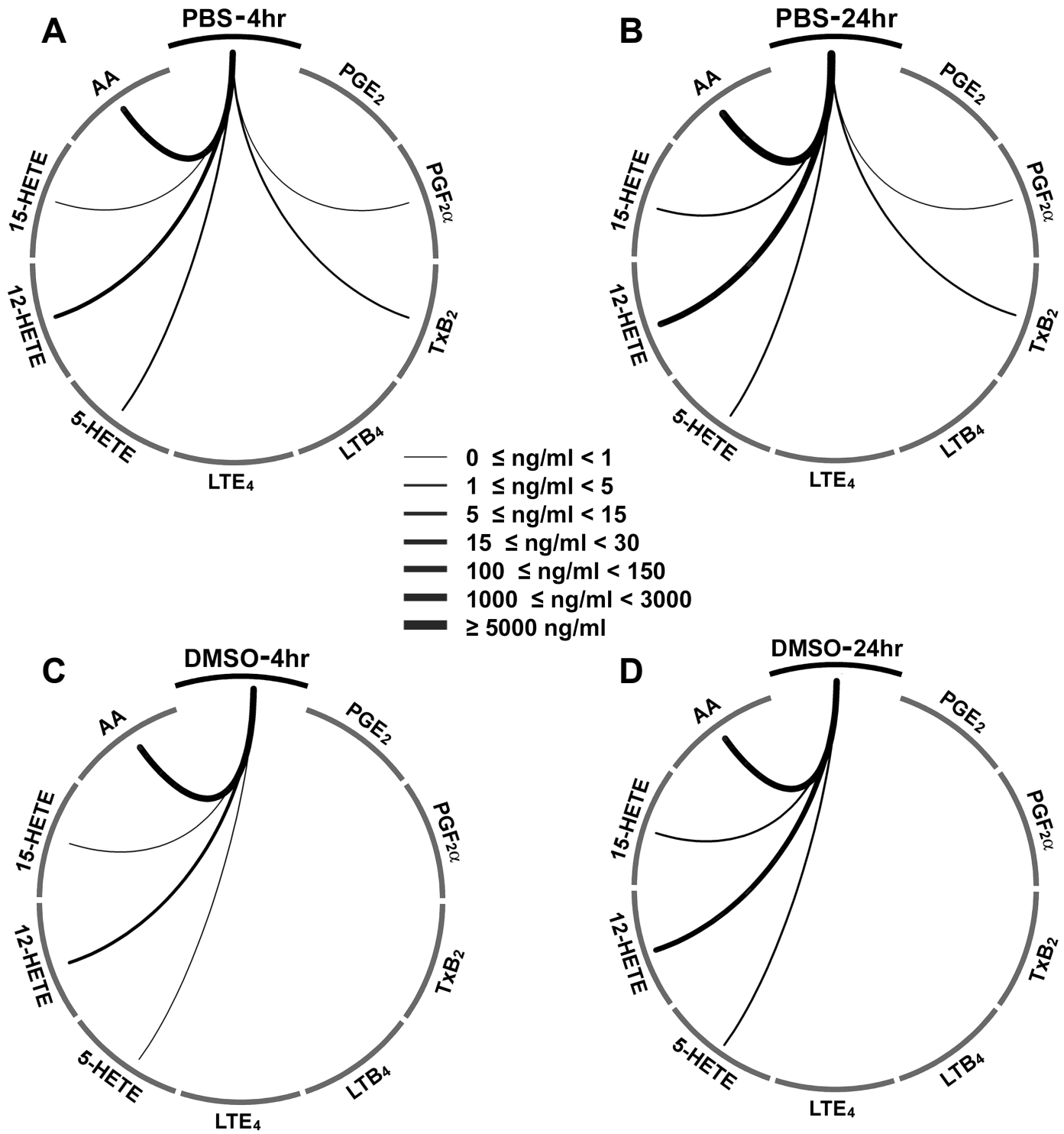


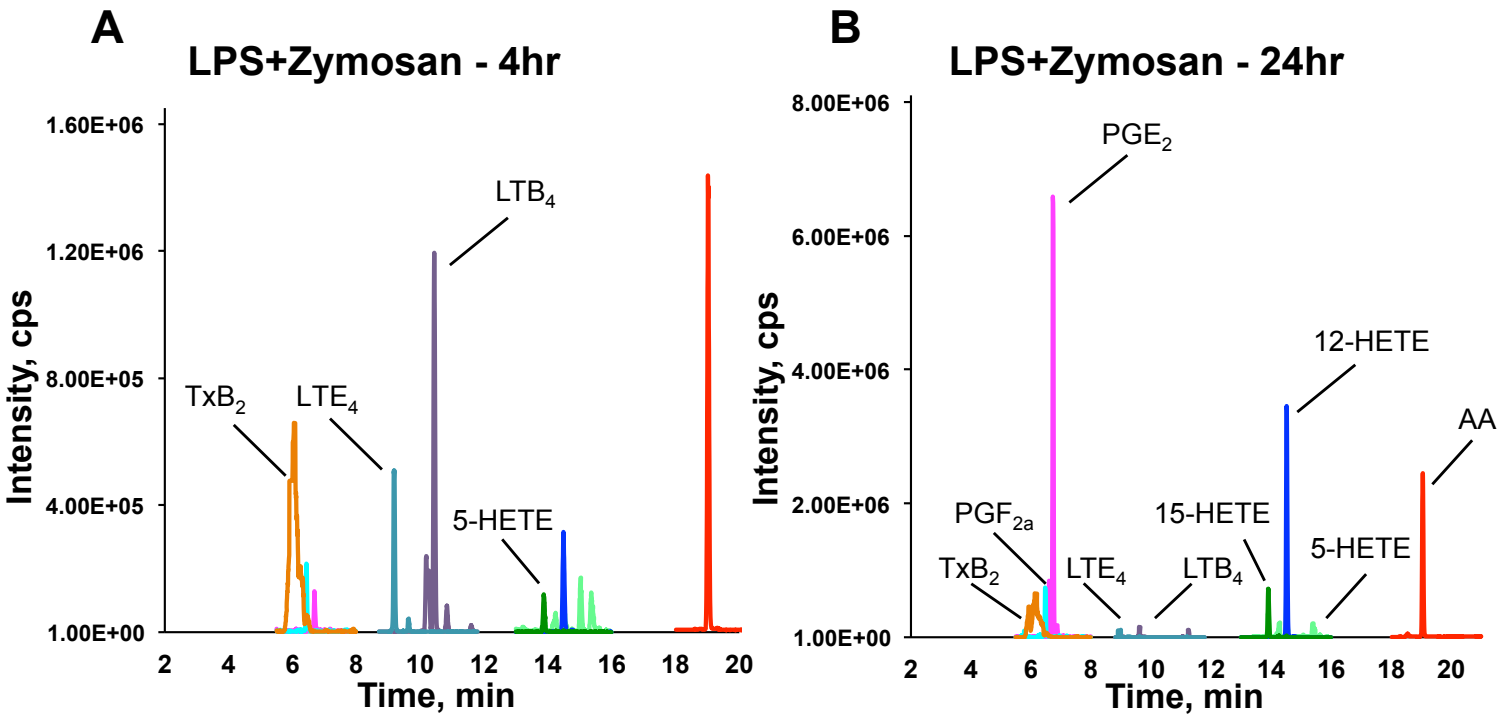
Supplementary Figure 1. Kinetics of LPS-induced eicosanoid production in human whole blood *in vitro*. Heparinized whole blood was incubated with 100 µg/ml of LPS for 0, 0.5, 1, 2, 4, 8, 24, 32 and 48 hr at 37 °C. PGE_2 (**A**), $PGF_{2\alpha}$ (**B**), TxB_2 (**C**), 12-HETE (**D**), 15-HETE (**E**) and 20-HETE (**F**) were measured in plasma samples by UPLC-MS/MS as described in Methods. Data represent means ± S.D. from 4 donors. Unpaired t test, two-tailed *P < 0.05, **P ≤ 0.01, ***P ≤ 0.001 for values significantly greater than PBS control at the corresponding time point.



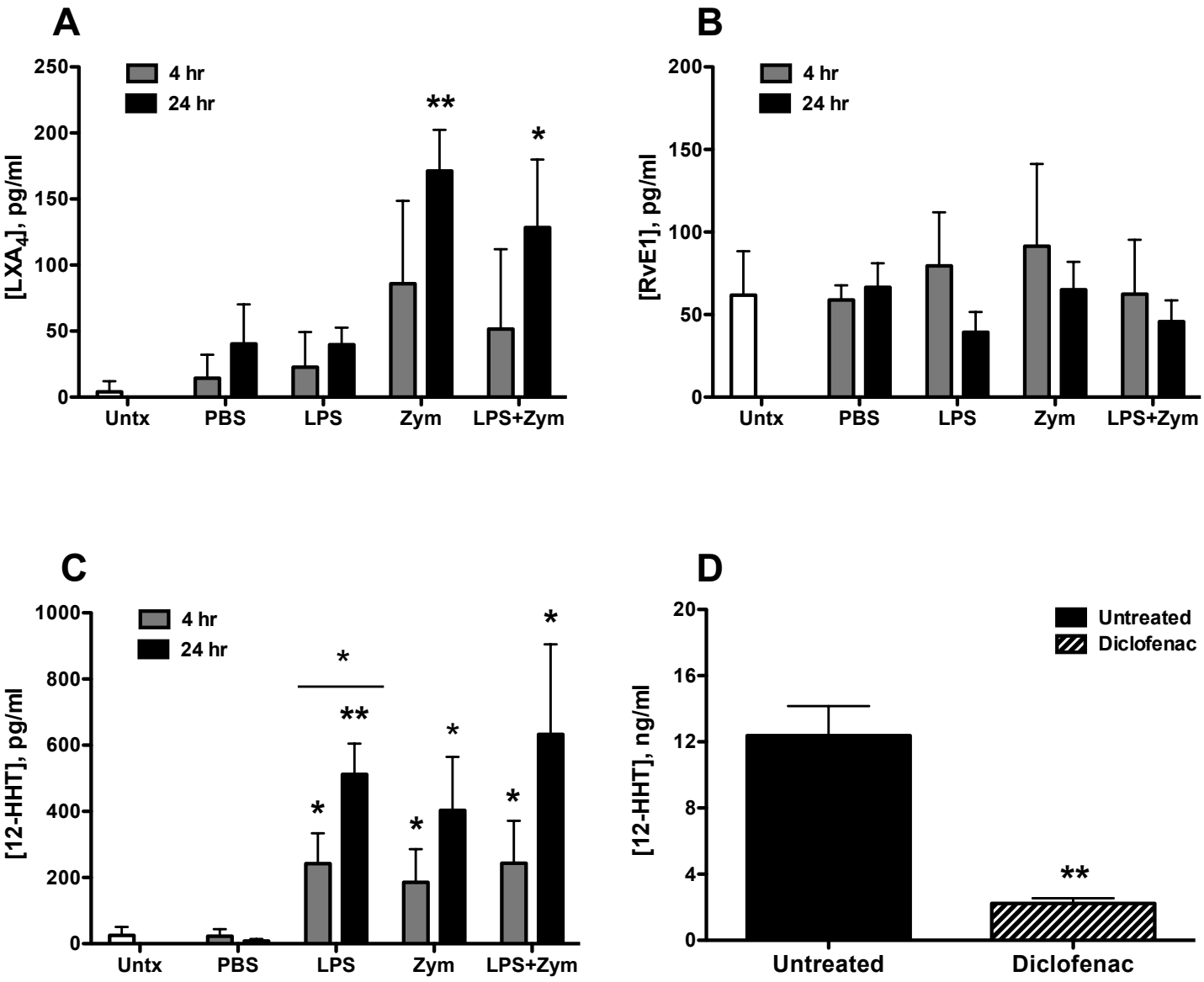
Supplementary Figure 2. Kinetics of zymosan-induced eicosanoid production in human whole blood *in vitro*. Heparinized whole blood was incubated with 125 µg/ml of zymosan for 0, 0.5, 1, 2, 4, 8, 24, 32 and 48 hr at 37 °C. LTB_4 (**A**), LTE_4 (**B**), 5-HETE (**C**) andTxB_2 (**D**) were measured in plasma samples by UPLC-MS/MS as described in Methods. Data represent means ± S.D. from 4 donors. Unpaired t test, two-tailed *P < 0.05, **P ≤ 0.01, ***P ≤ 0.001, ****P ≤ 0.0001 for values significantly greater than PBS control at the corresponding time point.



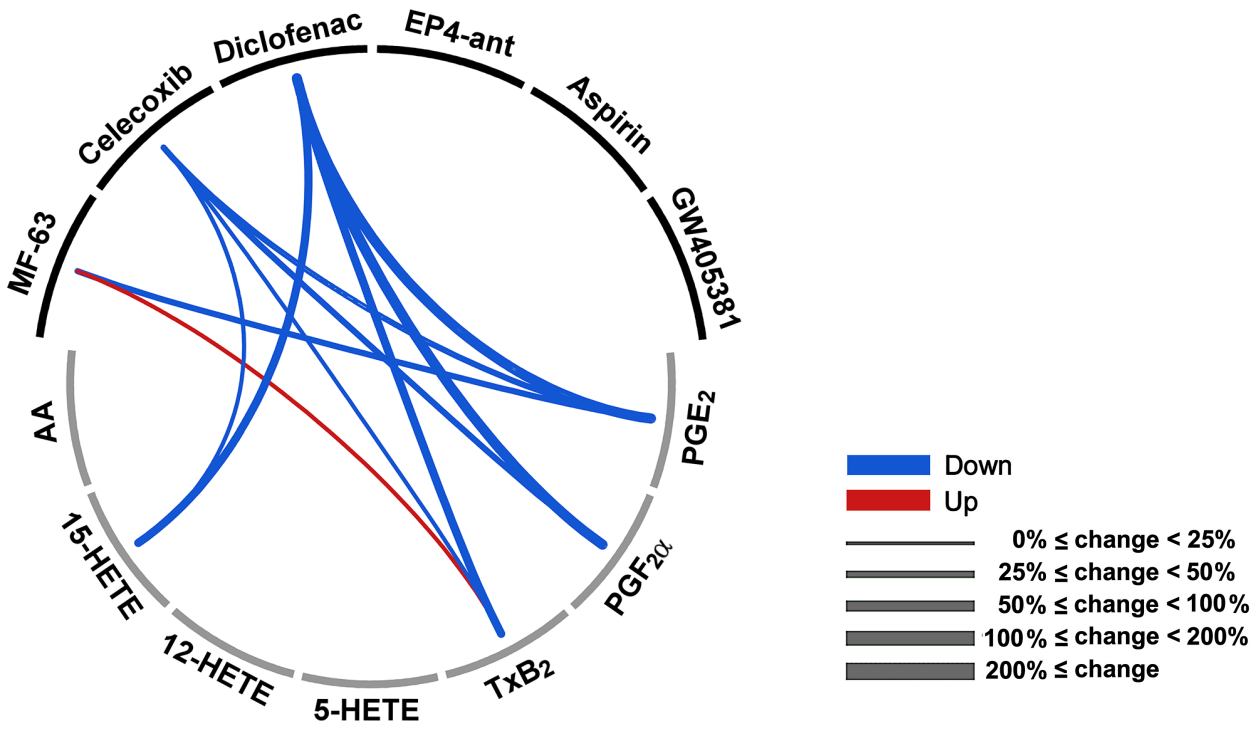
Supplementary Figure 3. Human plasma eicosanoid production in PBS or DMSO vehicle controls in whole blood *in vitro*. Circos plots comparing plasma eicosanoids of human whole blood incubated with PBS or DMSO vehicles for 4 or 24 hours (n=22). *In vitro* human whole-blood assay and UPLC-MS/MS analysis were performed as described in Methods.



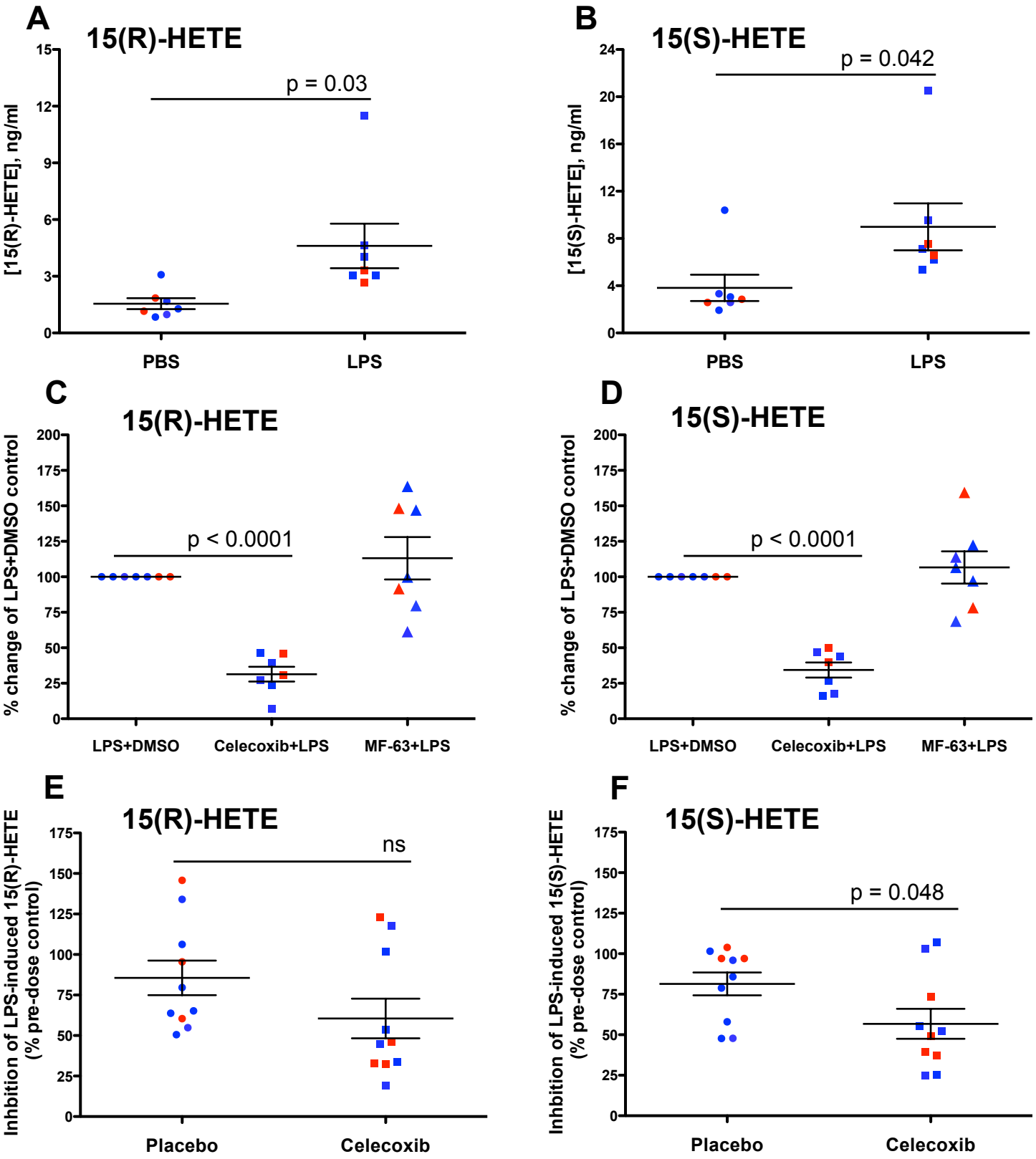
Supplementary Figure 4. UPLC-MS/MS profiling of bioactive eicosanoids in *in vitro* stimulated human whole blood. Representative chromatograms of eicosanoids in whole blood co-stimulated with 100 μg/ml of LPS and 125 μg/ml of zymosan for 4 hr (**A**) or 24 hr (**B**). Selected peaks were identified by comparison with isotope-labeled standards.



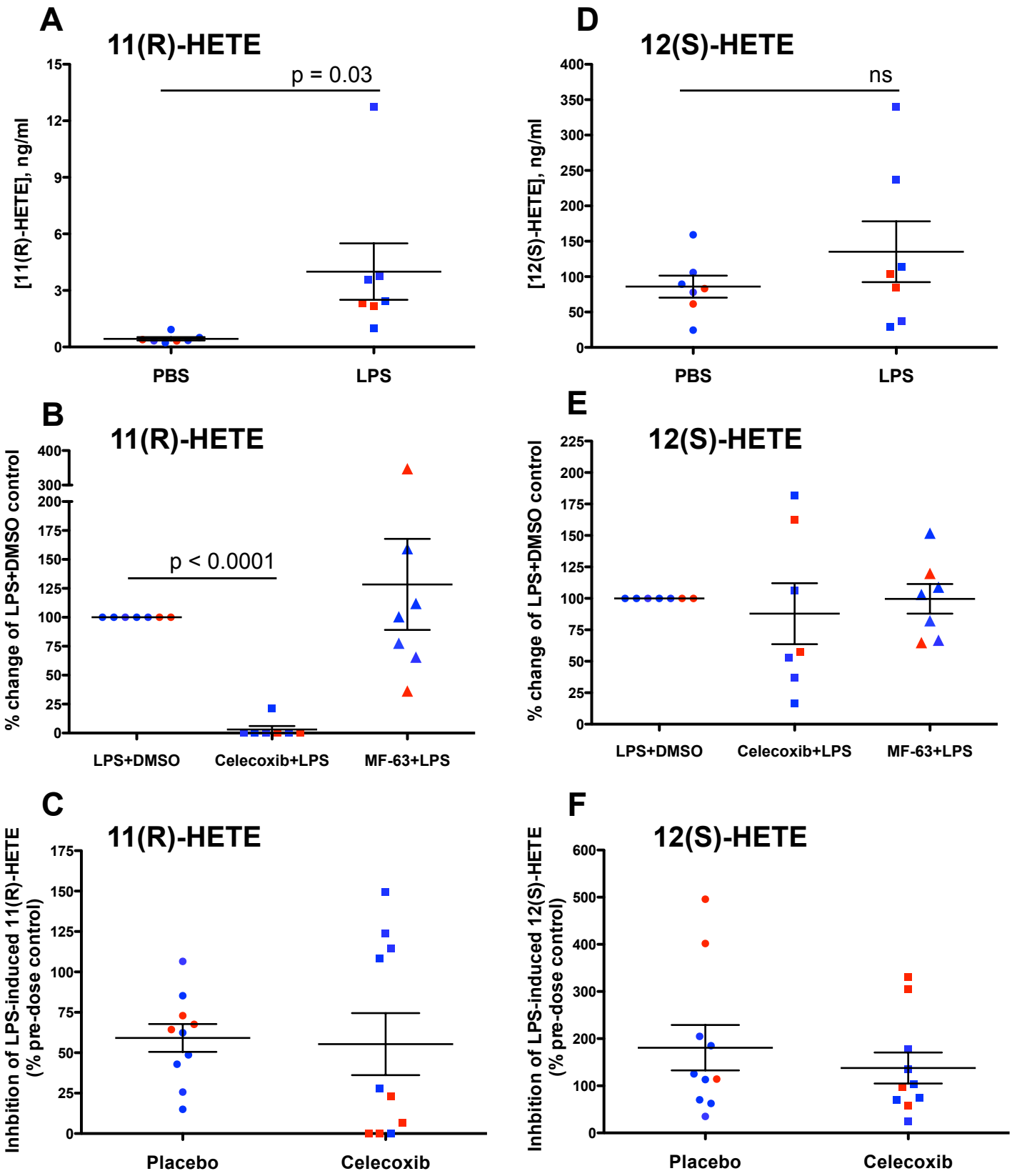
Supplementary Figure 5. Production of lipoxin A_4 (LXA_4), resolvin E1 (RvE1) and 12-HTT by human whole blood *in vitro*. Heparinized whole blood was incubated with 100 µg/ml of LPS and/or with 125 µg/ml of zymosan for 4 and 24 hr at 37 °C. LXA_4 (**A**), RvE1 (**B**) and 12-HHT (**C**) were measured in plasma samples by UPLC-MS/MS. Human whole blood was incubated for 1 hr at 37 °C to allow coagulation. 12-HHT (**D**) was measured in sera from untreated and diclofenac-inhibited (10 µM) whole blood by UPLC-MS/MS. Data represent means ± S.D. from 4 donors. Paired t test, two-tailed *$P < 0.05$, **$P \leq 0.01$ for values significantly greater than PBS control at the corresponding time point for plasma and greater than untreated control for serum.



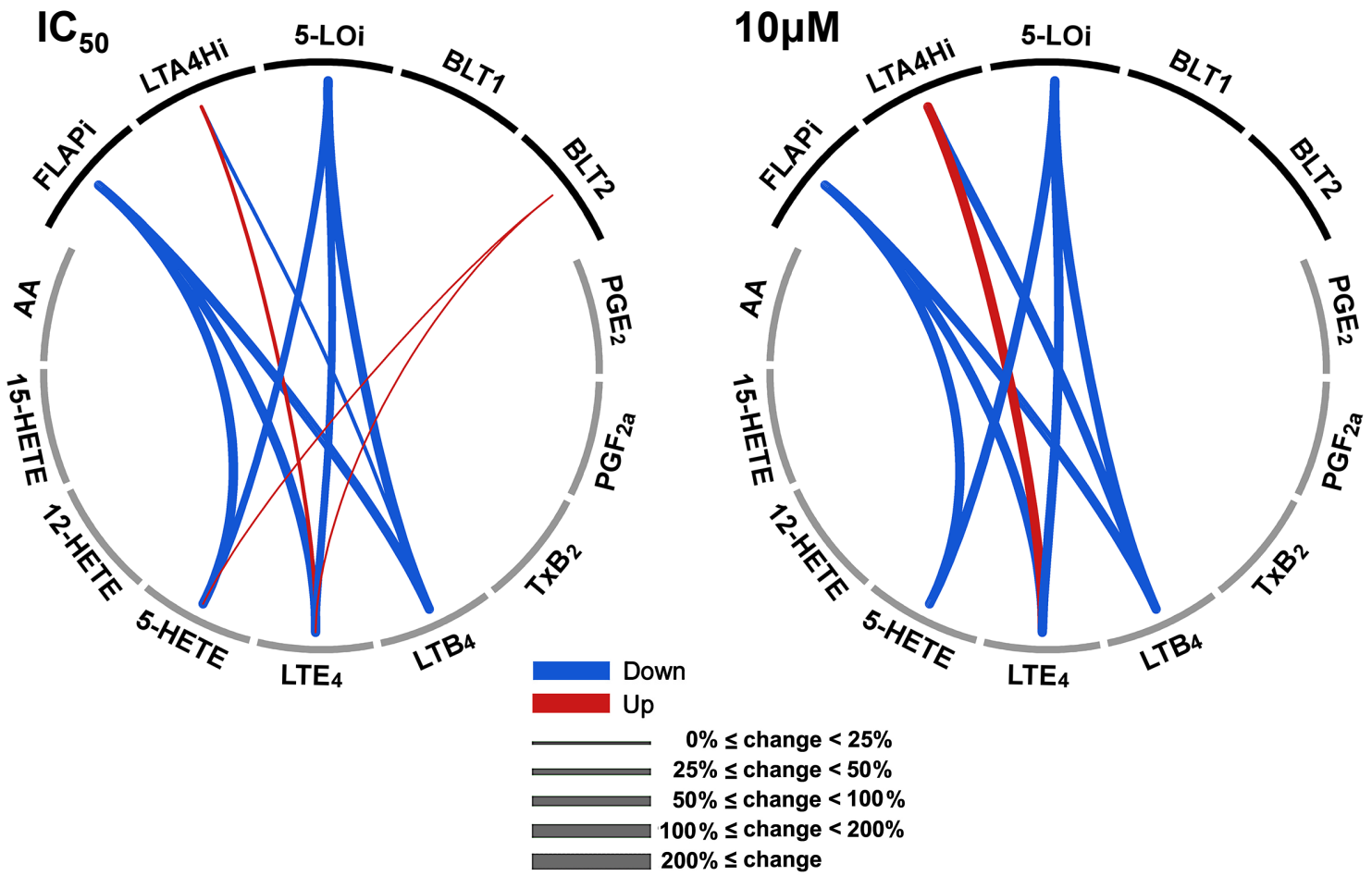
Supplementary Figure 6. Effect of drugs targeting the COX pathway and used at the corresponding IC_{50} on human plasma lipidome *in vitro*. Circos plot comparing plasma eicosanoid profiles of human whole blood stimulated with 100 µg/ml LPS and treated with the mPGES-1 inhibitor, MF-63 (n=5), COX-2 inhibitor celecoxib (n=5), COX-1/2 inhibitor diclofenac (n=5), COX-2 inhibitor GW406381 (n=5), COX-1/-2 inhibitor aspirin (n=4) or EP4 antagonist CJ-042794 (n=5). All compounds, except aspirin (15 µM), were used at the corresponding IC_{50}. *In vitro* human whole-blood assay and UPLC-MS/MS analysis were performed as described in Methods. Data expressed as percent of LPS +DMSO control. Red lines indicate significantly elevated levels of the corresponding lipid, while blue lines indicate significant reductions; thickness of lines represents degree of change. One-sample, two-tailed *t* test.



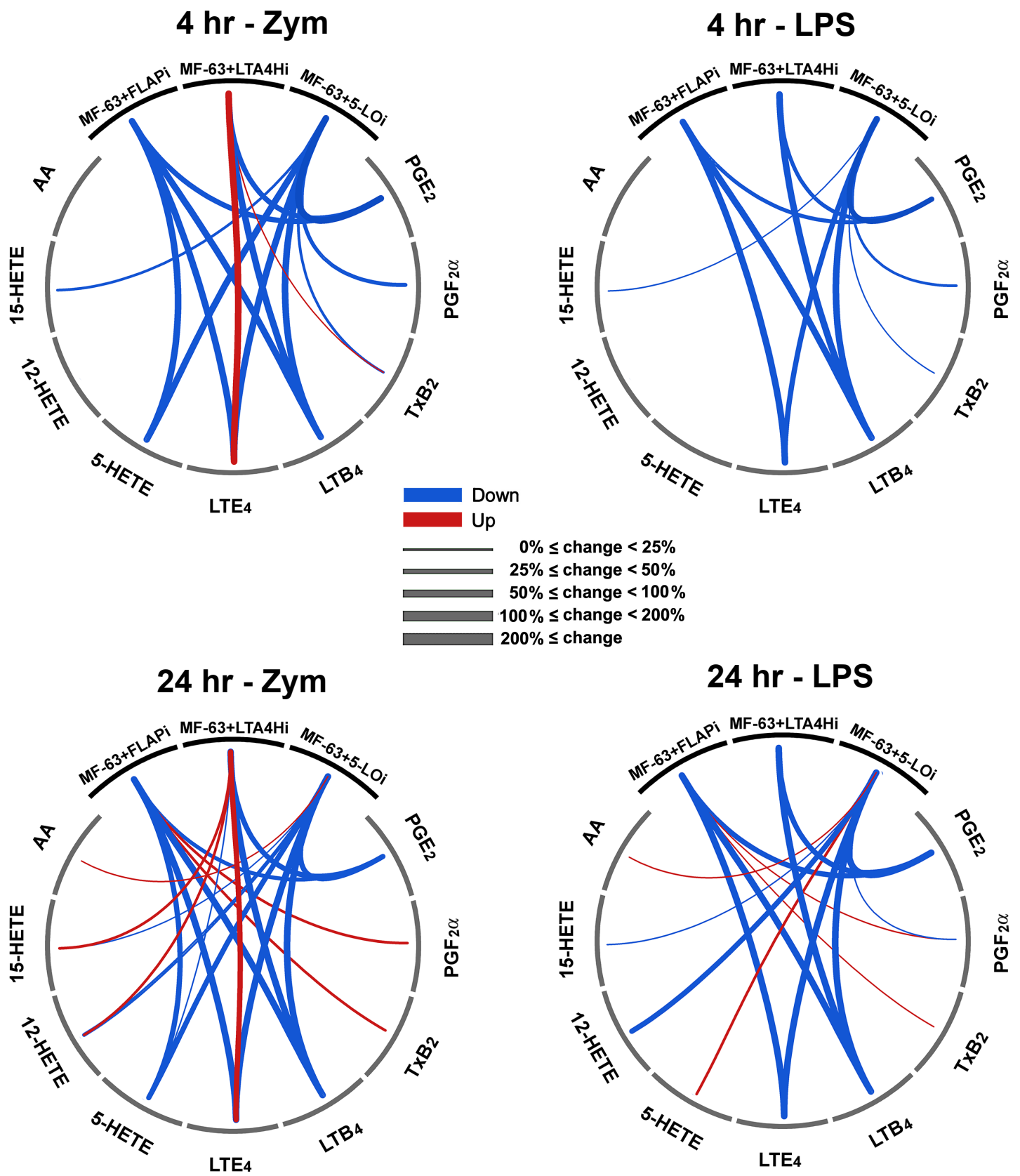
Supplementary Figure 7. Effects of COX-2 vs mPGES-1 inhibition on chiral products of 15-HETE in human plasma. For *in vitro* assay, whole blood was stimulated with 100 µg/ml LPS and treated with the mPGES-1 inhibitor, MF-63, or with a COX-2 inhibitor, celecoxib, at 10 µM each, for 24 hr. *In vitro* human whole-blood assay and chiral LC-ECAPCI/MS analysis were performed as described in Methods. LPS-triggered 15(R)-HETE (**A**) and 15(S)-HETE (**B**) represent means ± S.D. expressed in ng/ml. Unpaired, two-tailed *t* test, n=7. Drug effects on plasma levels of 15(R)-HETE (**C**) and 15(S)-HETE (**D**) expressed as percent of LPS+DMSO control. One-sample, two-tailed t test, n=7. For *ex vivo* assay, whole blood was collected before (pre-dose) and 3 hr after (post-dose) celecoxib or placebo administration. Plasma levels of 15(R)-HETE (**E**) and 15(S)-HETE (**F**) expressed as percent of pre-dose control. *Ex vivo* human whole blood assay and chiral LC-ECAPCI/MS analysis were performed as described in Methods. Red and blue dots represent female and male subjects, respectively. Unpaired, two-tailed t test, n=10/group; ns: non significant.



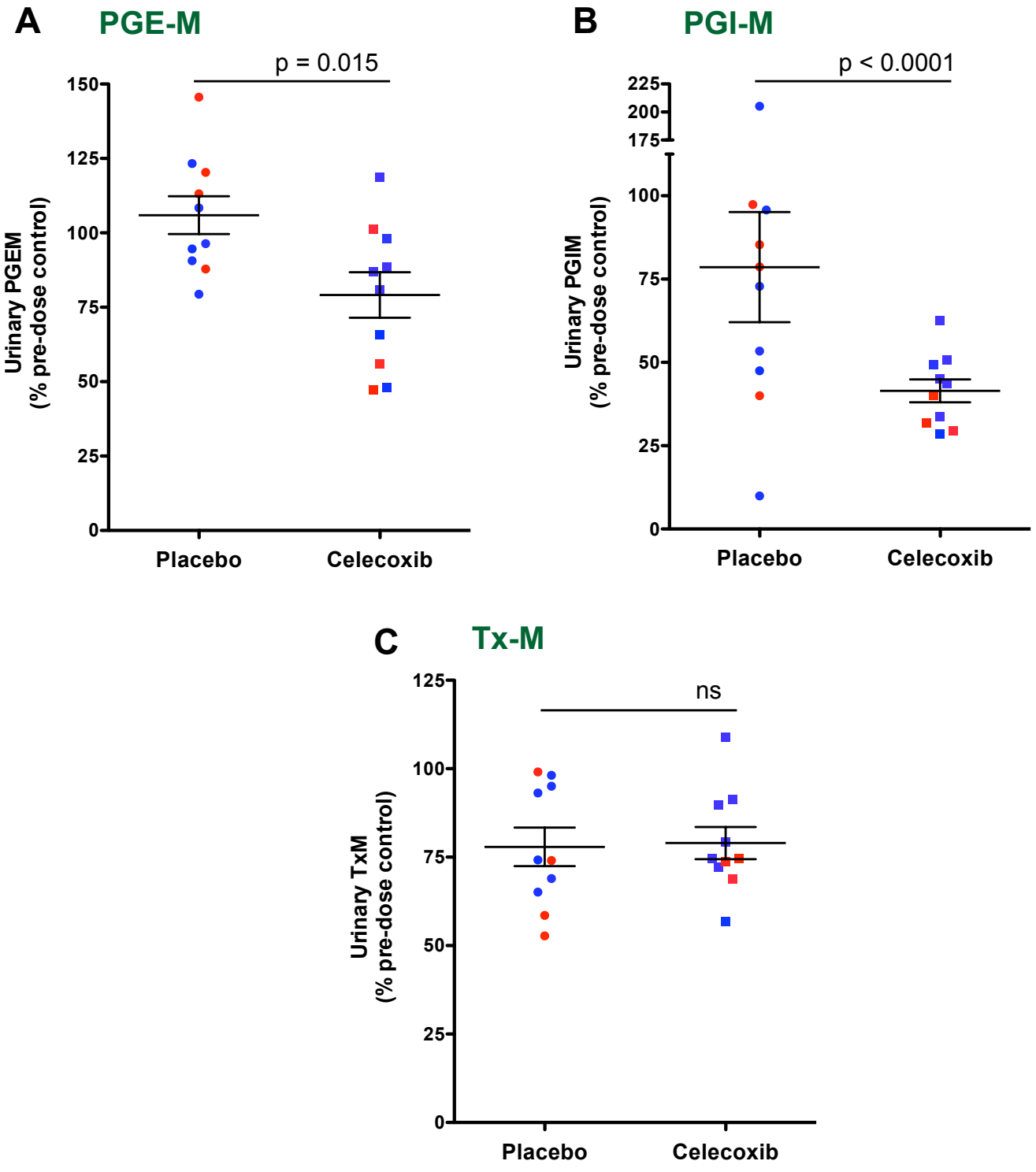
Supplementary Figure 8. Effects of COX-2 vs mPGES-1 inhibition on chiral products of 11(R)-HETE and 12(S)-HETE in human plasma. For *in vitro* assay, whole blood was stimulated with 100 µg/ml LPS and treated with the mPGES-1 inhibitor, MF-63, or with a COX-2 inhibitor, celecoxib, at 10 µM each, for 24 hr. *In vitro* human whole-blood assay and chiral LC-ECAPCI/MS analysis was performed as described in Methods. LPS-triggered 11(R)-HETE (**A**) and 12(S)-HETE (**D**) represent means ± SEM expressed in ng/ml. Unpaired, two-tailed *t* test, n=7. Drug effects on plasma levels of 11(R)-HETE (**B**) and 12(S)-HETE (**E**) expressed as percent of LPS+DMSO control. One-sample, two-tailed t test, n=7. For *ex vivo* assay, whole blood was collected before (pre-dose) and 3 hr after (post-dose) celecoxib or placebo administration. Plasma levels of 11(R)-HETE (**C**) and 12(S)-HETE (**F**) expressed as percent of pre-dose control. *Ex vivo* human whole blood assay and chiral LC-ECAPCI/MS analysis was performed as described in Methods. Red and blue dots represent female and male subjects, respectively. Unpaired, two-tailed t test, n=10/group. ns: non significant.



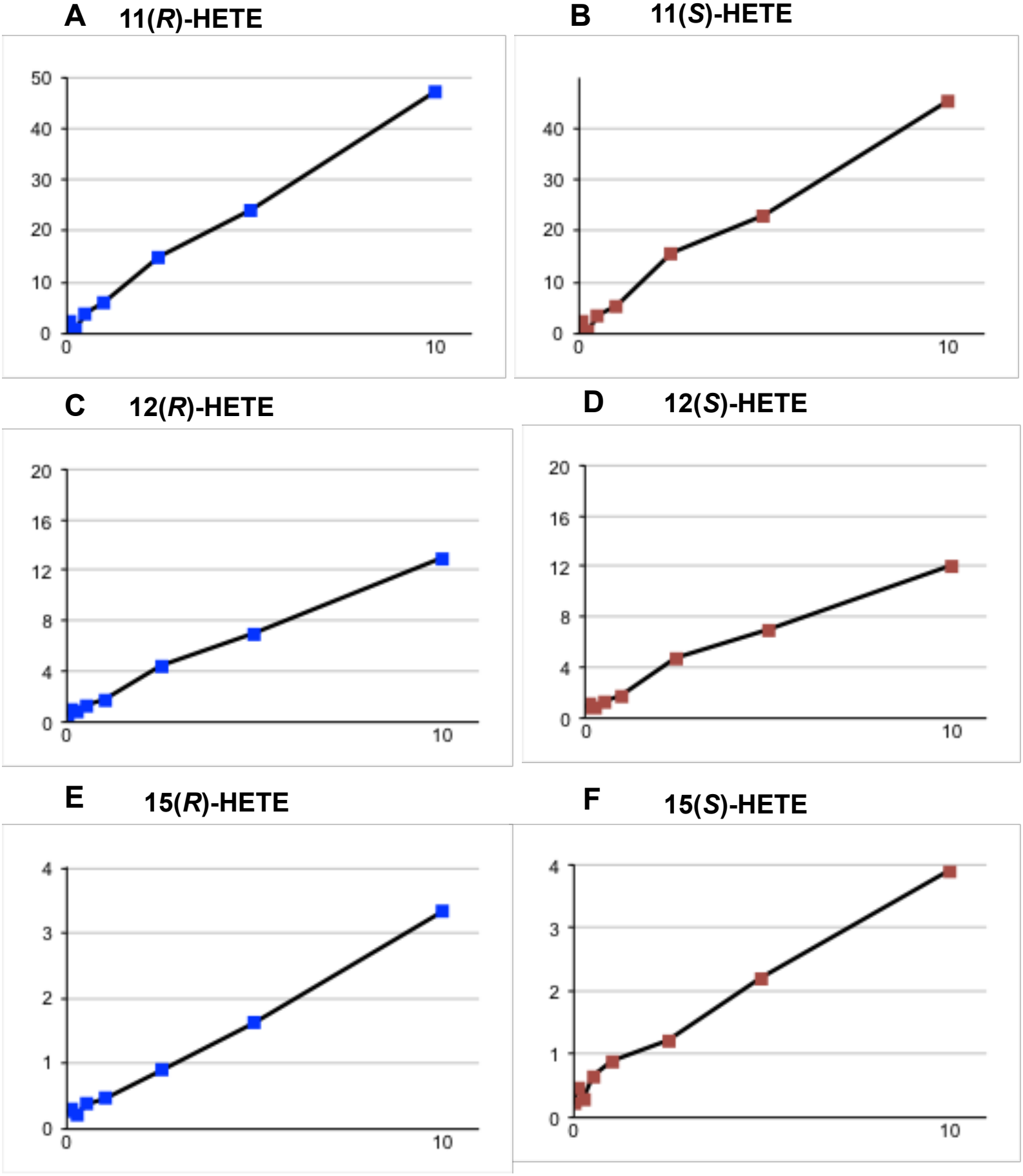
Supplementary Figure 9. Differential effects of drugs targeting the 5-LOX pathway on human plasma lipidome *in vitro*. Circos plots comparing plasma eicosanoid profiles of human whole blood stimulated with 125 μg/ml zymosan and treated with the FLAP inhibitor MK-0591 (n=5), LTA_4H inhibitor SC-57461A (n=5), 5-LOX inhibitor ABT-761 (n=5), BLT1-type LTB_4 antagonist LY293111 (n=5) or BLT2-type LTB_4 antagonist LY255283 (n=5). All compounds were used at the corresponding IC_{50} or 10 μM concentration. *In vitro* human whole-blood assay and UPLC-MS/MS analysis were performed as described in Methods. Data expressed as percent of zymosan+DMSO control. Red lines indicate significantly elevated levels of the corresponding lipid, while blue lines indicate significant reductions; thickness of lines represents degree of change. One-sample, two-tailed *t* test.



Supplementary Figure 10. Stimulus- and time-dependent effects of drug combinations on human plasma lipidome *in vitro*. Human whole blood was stimulated with 100 µg/ml of LPS or 125 µg/ml of zymosan and treated with the mPGES-1i MF-63, in combination with the FLAPi MK-0591, or with the LTA_4Hi SC-57461A, or with the 5-LOXi ABT-761, for 4 or 24 hr. All compounds were used at 10 µM. *In vitro* human whole-blood assay and UPLC-MS/MS analysis were performed as described in Methods. Data expressed as percent of LPS+DMSO or zymosan+DMSO controls. Red lines indicate significantly elevated levels of the corresponding lipid, while blue lines indicate significant reductions; thickness of lines represents degree of change. One-sample, two-tailed *t* test, n=5.



Supplementary Figure 11. Effect of celecoxib on urinary eicosanoid metabolites at T_{max}. Urine samples were collected before (pre-dose) and 3 hr after (post-dose) celecoxib or placebo administration. Urinary metabolites of PGE_2, PGE-M (**A**), of PGI_2, PGI-M (**B**), and TxB_2, Tx-M (**C**) were analyzed by UPLC-MS/MS and expressed as percent of pre-dose control. Red color – female subjects, blue color – male subjects. Unpaired, two-tailed *t* test, n=10/group. PGE-M – tetranor-PGEM, PGI-M – 2,3-dinor-6-keto-$PGF_{1\alpha}$, Tx-M – 11-dehydro-TxB_2, ns: non significant.



Supplementary Figure 12. Calibration curves for chiral 11-hydroxyeicosatetraenoic acid (11-HETE), 12-hydroxyeicosatetraenoic acid (12-HETE) and 15-hydroxyeicosatetraenoic acid (15-HETE). Calibration samples were spiked with authentic standards of 11(*S*)-HETE, 11(*R*)-HETE, (±)12-HETE and (±)15-HETE in the amounts of 0, 0.1, 0.25, 0.5, 1, 2.5, 5 ng, and 1 ng of the internal standard [d8]-15-(*S*)-HETE. Calibration curves for 11(*R*)-HETE (**A**), 11(*S*)-HETE (**B**), 12(*R*)-HETE (**C**), 12(*S*)-HETE (**D**), 15(*R*)-HETE (**E**) and 15(*S*)-HETE (**F**) were plotted using a linear regression of peak area ration of analytes against the internal standard.

Drug/ compound	Target	IC_{50}, μM	Assay	Method of detection	Reference
ABT-761	5-LOX	0.15	hWBA, A23187-trigg LTB_4	EIA for LTB_4	**53**
MK-0591	FLAP	0.51	hWBA, A23187-trigg LTB_4	RIA for LTB_4	**54**
SC-57461A	LTA_4H	0.05	hWBA, A23187-trigg LTB_4	ELISA for LTB_4	**55**
LY-293111	BLT1-type LTB_4 receptor	1.1	Plasma-depleted human blood, LTB_4 triggered by fMLP and thrombin	EIA for LTB_4	**56**
LY-255283[A]	BLT2-type LTB_4 receptor	~1	Competitive inhibition of LTB_4 for binding to human recombinant BLT2	Binding assay	**58**
MF-63	mPGES-1	0.8	hWBA, LPS-triggered PGE_2	EIA for PGE_2	**51**
Aspirin	COX-1/-2	15[B]	NA	NA	**49**
Celecoxib	COX-2	0.3	hWBA, LPS-triggered PGE_2	EIA for PGE_2	**48**
Diclofenac	COX-1/-2	0.01	hWBA, LPS-triggered PGE_2	RIA for PGE_2	**49**
GW406381	COX-2	0.042	hWBA, LPS-triggered PGE_2	EIA for PGE_2	**50**
CJ-042794	EP_4 receptor	0.4	hWBA, inhibition of LPS-triggered TNFα in competition with exogenous PGE_2	ELISA for TNFα	**52**

[A] – LY-255283 was tested in human whole blood for LTB_4 inhibition (57) but no IC_{50} was provided.

[B] – 15 μM corresponds to peak systemic plasma concentration of low-dose aspirin (59).

Supplementary Table 1. Concentrations of the tested drugs that inhibited 50% of response (IC_{50}) in a human whole-blood assay or the corresponding assay.

Compounds	Parent ion (*m/z*)	Product ion (*m/z*)	Collision energy (eV)	Retention time (min)
PGE_2	351.2	315.2	**10**	6.69
$PGF_{2\alpha}$	353.2	193.1	**24**	6.44
TxB_2	369.2	169.1	**17**	6.07
LTB_4	335.2	195.1	**16**	10.44
LTE_4	438.2	333.2	**16**	9.66
5-HETE	319.2	115.0	**15**	15.03
12-HETE	319.2	179.1	**7**	14.49
15-HETE	319.2	219.2	**13**	13.87
AA	303.2	259.2	**12**	19.01
*12-HHT	279.2	179.15	**12**	11.46
RvD1	375.2	141.1	**13**	7.76
RvD2	375.2	141.1	**13**	7.09
LXA_4	351.2	115.1	**13**	7.83
*RvE1	349.2	161.1	**15**	4.61
*MaR1	359.2	177.1	**14**	10.15
*PD	359.2	153.1	**15**	10.02
8(R)-HETE-PFB	319.23	155.06	**16**	9.18
8(S)-HETE-PFB	319.23	155.06	**16**	9.76
11(R)-HETE-PFB	319.23	167.1	**16**	8.9
11(S)-HETE-PFB	319.23	167.1	**16**	9.73
12(R)-HETE-PFB	319.23	179.1	**14**	9.37
12(S)-HETE-PFB	319.23	179.1	**14**	9.64
15(R)-HETE-PFB	319.23	219.1	**13**	9.57
15(S)-HETE-PFB	319.23	219.1	**13**	11.1
PGE_2-d_4	355.2	319.2	**10**	6.66
$PGF_{2\alpha}$-d_4	357.2	197.1	**24**	6.43
TxB_2-d_4	373.2	173.1	**17**	6.09
LTB_4-d_4	339.2	197.1	**16**	10.41
LTE_4-d_5	443.2	338.2	**16**	9.62
5-HETE-d_8	327.2	116.0	**15**	14.93
12-HETE-d_8	327.2	184.2	**7**	14.38
15-HETE-d_8	327.2	226.2	**13**	13.76
AA-d_8	311.2	267.2	**12**	18.91
RvD1-d_5	380.1	141.1	**13**	7.71
RvD2-d_5	380.1	141.1	**13**	7.05
LXA_4-d_5	356.2	115.1	**13**	7.75
15(S)-HETE-d_8-PFB	327.28	226.18	**13**	11.23

* 15-HETE-d_8- I.S. for 12-HHT; RvD2-d_5- I.S. for RvE1; LXA_4-d_5- I.S. for MaR1; LXA_4-d_5- I.S. for PD

I.S. – Internal Standard, PFB - 2,3,4,5,6-pentafluorobenzyl.

Supplementary Table 2. Multiple reaction monitoring (MRM) conditions for UPLC-MS/MS and parallel reaction monitoring (PRM) conditions for UPLC-ECAPCI/HRMS methods for eicosanoid identification and quantitation.

Treatment	Eicosanoid	Males, n=11 (ng/ml, ± S.D.)	Females, n=11 (ng/ml, ± S.D.)	All subjects, n=22 (ng/ml, ± S.D.)
LPS (100 µg/ml), 24hr	**PGE_2**	**28.37 ± 17**	**22.98 ± 11**	**25.68 ± 14.3**
	(PBS control)	*0.04 ± 0.07*	*0.05 ± 0.06*	*0.04 ± 0.06*
	PGF_{2a}	**5.95 ± 2.84**	**6.1 ± 2.1**	**6 ± 2.43**
	(PBS control)	*0.31 ± 0.48*	*0.37 ± 0.2*	*0.32 ± 0.34*
	TxB_2	**25.62 ± 8.75**	**27.72 ± 9**	**26.67 ± 8.73**
	(PBS control)	*1.64 ± 1.33*	*1.74 ± 1.63*	*1.52 ± 1.48*
	15-HETE	**4.68 ± 2.47**	**4.73 ± 1.82**	**4.7 ± 2.1**
	(PBS control)	*1.15 ± 0.42*	*1.23 ± 0.42*	*1.2 ± 0.42*
Zymosan (125 µg/ml), 4hr	**LTB_4**	**8.67 ± 3.87**	**8.2 ± 4.53**	**8.43 ± 4.12**
	(PBS control)	*0.02 ± 0.08*	*0.08 ± 0.25*	*0.05 ± 0.2*
	LTE_4	**12.9 ± 9.68**	**6.34 ± 3.61**	**9.62 ± 7.88**
	(PBS control)	*0.1 ± 0.3*	*0.04 ± 0.06*	*0.07 ± 0.2*
	5-HETE	**15.78 ± 6.2**	**14.4 ± 6.77**	**15.1 ± 6.37**
	(PBS control)	*1.42 ± 0.5*	*1.45 ± 0.7*	*1.43 ± 0.6*

Supplementary Table 3. Production of eicosanoids in stimulated human whole blood *in vitro*. Heparinized whole blood was incubated with 100 µg/ml of LPS for 24 hr or with 125 µg/ml of zymosan for 4 hr at 37 °C. Eicosanoids were measured in plasma samples by UPLC-MS/MS as described in Methods. Data represent means ± S.D. of values significantly greater than PBS control. Unpaired, two-tailed *t* test.

Analyte	Drug response, ng/ml ± S.D.											
	MF-63 (n=5)		Aspirin (n=4)		Celecoxib (n=5)		Diclofenac (n=5)		GW406381 (n=5)		CJ-042794 (n=5)	
	LPS +DMSO	LPS+ drug	LPS +DMSO	LPS+ drug	LPS +DMSO	LPS+ drug	LPS +DMSO	LPS+ drug	LPS +DMSO	LPS+ drug	LPS +DMSO	LPS+ drug
PGE_2	26.3±14	12±12.2	16.7±7	15.8±5	31.8±19	2.3±2.2	24.5±18	0.1±0.1	16.7±6	0.5±0.4	28±17.5	30.7±18
$PGF_{2\alpha}$	9.3±4.2	14.1±7	9.2±2.4	3.1±1.1	10.6±4	1.1±0.4	11.4±6	0.3±0.1	9.2±2.4	0.5±0.3	8.4±3.7	10.7±6
TxB_2	34.2±13	46.8±17	44.4±6	15.3±4	33.8±12	6.6±2.5	35.1±16	0.9±0.2	44.4±6	6.3±2	35.3±11	47.6±20
15-HETE	8.9±2.8	10.2±4	6±0.85	3.9±0.5	9.8±3.2	2.5±1	8.2±3.3	1.7±0.5	6±0.8	1.5±0.5	7.5±2.1	9±3
12-HETE	221±150	244±149	86±28.8	117±30	204±138	221±130	145±80	176±56	86±29	95±23	144±98	190±125
5-HETE	5.8±6.7	5.7±6.7	1.6±1.3	1.8±0.5	2.8±0.8	3±0.9	2.7±0.8	3±0.9	1.6±1.3	4.1±4.9	3.1±1	3.2±1.1

Supplementary Table 4. Interindividual variability in drug response to high doses of the COX pathway inhibitors. Plasma eicosanoid profiles of human whole blood stimulated with 100 µg/ml LPS and treated MF-63, celecoxib, diclofenac, GW406381, CJ-042794 or aspirin for 24 hr. All compounds, except aspirin (1.5 mM), were used at 10 µM concentration. *In vitro* human whole-blood assay and UPLC-MS/MS analysis were performed as described in Methods. Data expressed in ng/ml and compared to LPS+DMSO control.

Analyte	Drug response, ng/ml ± S.D.											
	MF-63 (n=5)		Aspirin (n=4)		Celecoxib (n=5)		Diclofenac (n=5)		GW406381 (n=5)		CJ-042794 (n=5)	
	LPS +DMSO	LPS+ drug	LPS +DMSO	LPS+ drug	LPS +DMSO	LPS+ drug	LPS +DMSO	LPS+ drug	LPS +DMSO	LPS+ drug	LPS +DMSO	LPS+ drug
PGE_2	26.3±14	15.3±8	16.7±7	17.8±6	31.8±19	18.8±11	18±12	4.8±5	16.7±6	18.1±6	28±17.5	28.6±18
$PGF_{2\alpha}$	9.3±4.2	13.5±9	9.2±2.4	9.8±3.5	10.6±4	6.2±2.5	6±3.8	1.5±0.9	9.2±2.4	10±3	8.4±3.7	9.4±5
TxB_2	34.2±13	44±19.4	44.4±6	44.5±10	33.8±12	24±7.6	28.2±11	12±5.7	44.4±6	47.2±12	35.3±11	42.5±18
15-HETE	8.9±2.8	9.5±3.4	6±0.85	6.2±1.2	9.8±3.2	6.2±1	3.6±1.4	1.5±0.6	6±0.8	6.2±1	7.5±2.1	9.2±4.6
12-HETE	221±150	232±133	86±28.8	85±18	204±138	191±108	52±23	65±24	86±29	94±27	144±98	173±117
5-HETE	5.8±6.7	6.4±7.6	1.6±1.3	1.4±1.2	2.8±0.8	3.1±0.8	1.2±0.3	1.3±0.4	1.6±1.3	4±5.6	3.1±1	3.2±1.1

Supplementary Table 5. Interindividual variability in drug response to the COX pathway inhibitors at IC_{50} doses. Plasma eicosanoid profiles of human whole blood stimulated with 100 µg/ml LPS and treated MF-63, celecoxib, diclofenac, GW406381, CJ-042794 or aspirin for 24 hr. All compounds, except aspirin (15 µM), were used at the corresponding IC_{50}. *In vitro* human whole-blood assay and UPLC-MS/MS analysis were performed as described in Methods. Data expressed in ng/ml and compared to LPS+DMSO control.