

SUPPLEMENTAL DIGITAL CONTENT

Prenatal exposure to per- and polyfluoroalkyl substances and childhood autism-related outcomes

Table of Contents

eTable 1. Final subsamples available for analysis, by cohort.	2
eTable 2. Descriptive Characteristics of Participants Contributing to Analyses, by NIH ECHO Cohort	6
eTable 3. Distributions of PFAS (untransformed values, $\mu\text{g/L}$) and % below the limit of detection (LOD).	9
eTable 4. Associations of PFAS and SRS Raw Scores from Single-Pollutant Models	10
eTable 5. Associations between PFAS and SRS total T-score stratified by SRS-2 form completed	11
eTable 6. Sensitivity analysis dropping samples below the LOD for the specific analytes with >20% values below the LOD.	12
eTable 7. Associations of PFAS and T-scores on SRS subscales from single-pollutant models	13
eTable 8. Associations of PFAS and ASD diagnoses from single-pollutant models.....	14
eTable 9. Associations of PFAS and ASD diagnosis in mixture models using Bayesian Weighted Sums (n=809).....	15
eTable 10. Sex-stratified associations between PFAS and SRS total T-score	16
eTable 11. Sex-stratified associations between PFAS and SRS using Bayesian Weighted Sums.....	17
eFigure 1. Directed acyclic graph ^a	18
eFigure 2. Boxplots of PFAS analytes (log-transformed) in the pooled ECHO sample	19
eFigure 3. Boxplots of PFAS analyte distributions (log-transformed) by ECHO cohort	20
eFigure 4. Correlation plots of PFAS analytes	21
eFigure 5. Coefficient plots of leave-one-out results for SRS total T-score.....	22

eTable 1. Final subsamples available for analysis, by cohort.

Cohort	Eligibility criteria	Total enrolled	Measured PFAS		Among those with PFAS, # with available outcome data		Among those with PFAS, # missing outcome data		Among those with PFAS and outcome data, # with singleton births
			n	% ^a	n	% ^b	n	% ^b	
		N	n	%^a	n	%^b	n	%^b	n
Project Viva¹	Eligible women were fluent in English, had singleton pregnancies, had pregnancies of <22 weeks' gestation, and had no plans to move away from the study area	1224	850	69%	467	55%	383	45%	467
New Hampshire Birth Cohort Study (NHBCS)²	Eligible women were 18-45 years old, fluent in English, had singleton pregnancies, and used a private, unregulated water system (e.g., private well) at home.	2238	325	15%	318	98%	7	2%	318
Healthy Start³	Eligible women were ≥16 years old, had singleton pregnancies, and had no history of previous stillbirth or extremely preterm birth, and no self-reported history of diabetes, cancer, asthma treated with steroids, or psychiatric conditions	1379	651	47%	184	28%	467	72%	184
University of California-Markers of Autism Risk in Babies (MARBLER)⁴	Eligible women had one or more biological child(ren) or first degree relatives with ASD or the gestating child had an older half-sibling with ASD b) Mother was ≥18 years old; d) Mother was proficient in English and the gestating child would be taught to speak English; e) Mother lived within 2.5 hrs of the Davis/Sacramento region	381	44	12%	31	70%	13	30%	31

Emory University Atlanta⁵	Eligible women were self-reported Black/African American race/ethnicity, had singleton pregnancies, self-reported that they were born in the United States, had English fluency, were aged 18–40 years old, and did not have IVF or pre-existing chronic medical conditions	268	245	91%	144	59%	101	41%	144
Pregnancy Environment and Lifestyle Study (PETALS)⁶	Eligible women were aged 18–45 years, able to provide informed consent in English, had singleton pregnancies, and did not have a pre-conception history of diabetes, cancer, hepatitis C, or liver cirrhosis.	1428	124	9%	78	63%	46	37%	78
Rochester⁷	Eligible women were ≥18 years old, had singleton pregnancies, and had no known substance abuse issues or history of psychotic illness, no major endocrine disorder, high-risk health condition, or significant obstetric concern at baseline.	296	273	92%	102	37%	171	63%	102
Kaiser Permanente Research Bank (KPRB)⁸	Eligible women were members of KPNC healthcare delivery system during their pregnancy, were ≥18 years old, and initiated prenatal care at a KPNC medical facility where cohort recruitment was taking place.	1250	13	1%	12	92%	1	8%	12
Chemicals in our Bodies (CIOB)⁹	Eligible mothers were fluent in English or Spanish, had singleton births, were ≥18 years of age, and had no major pregnancy complications	500	441	88%	67	15%	374	85%	67

Illinois Kids Development Study (IKIDS)¹⁰	Eligible mothers were 18–40 years old, had singleton pregnancies, were not in a high-risk pregnancy, were fluent in English, and were not planning to leave the area before the child’s first birthday	588	225	38%	26	12%	199	88%	26
^a Proportion calculated using total enrolled N as denominator ^b Proportion calculated using n with PFAS measured as denominator									

Reference:

1. Oken E, Baccarelli AA, Gold DR, et al. Cohort profile: project viva. *Int J Epidemiol*. Feb 2015;44(1):37-48. doi:10.1093/ije/dyu008
2. Gilbert-Diamond D, Emond JA, Baker ER, Korrick SA, Karagas MR. Relation between in Utero Arsenic Exposure and Birth Outcomes in a Cohort of Mothers and Their Newborns from New Hampshire. *Environ Health Perspect*. Aug 2016;124(8):1299-307. doi:10.1289/ehp.1510065
3. Starling AP, Adgate JL, Hamman RF, et al. Perfluoroalkyl Substances during Pregnancy and Offspring Weight and Adiposity at Birth: Examining Mediation by Maternal Fasting Glucose in the Healthy Start Study. *Environ Health Perspect*. Jun 26 2017;125(6):067016. doi:10.1289/EHP641
4. Hertz-Picciotto I, Schmidt RJ, Walker CK, et al. A Prospective Study of Environmental Exposures and Early Biomarkers in Autism Spectrum Disorder: Design, Protocols, and Preliminary Data from the MARBLES Study. *Environ Health Perspect*. Nov 2018;126(11):117004. doi:10.1289/EHP535
5. Brennan PA, Dunlop AL, Smith AK, Kramer M, Mulle J, Corwin EJ. Protocol for the Emory University African American maternal stress and infant gut microbiome cohort study. *BMC Pediatr*. Jul 22 2019;19(1):246. doi:10.1186/s12887-019-1630-4
6. Zhu Y, Hedderson MM, Feng J, Mevi AA, Ferrara A. The Pregnancy Environment and Lifestyle Study (PETALS): a population-based longitudinal multi-racial birth cohort. *BMC Pregnancy Childbirth*. Apr 17 2017;17(1):122. doi:10.1186/s12884-017-1301-0
7. O'Connor T, Best M, Brunner J, et al. Cohort profile: Understanding Pregnancy Signals and Infant Development (UPSIDE): a pregnancy cohort study on prenatal exposure mechanisms for child health. *BMJ Open*. Apr 1 2021;11(4):e044798. doi:10.1136/bmjopen-2020-044798
8. Hedderson MM, Ferrara A, Avalos LA, et al. The Kaiser Permanente Northern California research program on genes, environment, and health (RPGEH) pregnancy cohort: study design, methodology and baseline characteristics. *BMC Pregnancy Childbirth*. Nov 29 2016;16(1):381. doi:10.1186/s12884-016-1150-2

9. Morello-Frosch R, Cushing LJ, Jesdale BM, et al. Environmental Chemicals in an Urban Population of Pregnant Women and Their Newborns from San Francisco. *Environ Sci Technol*. Nov 15 2016;50(22):12464-12472. doi:10.1021/acs.est.6b03492
10. Eick SM, Enright EA, Geiger SD, et al. Associations of Maternal Stress, Prenatal Exposure to Per- and Polyfluoroalkyl Substances (PFAS), and Demographic Risk Factors with Birth Outcomes and Offspring Neurodevelopment: An Overview of the ECHO.CA.IL Prospective Birth Cohorts. *Int J Environ Res Public Health*. Jan 16 2021;18(2)doi:10.3390/ijerph18020742

eTable 2. Descriptive Characteristics of Participants Contributing to Analyses, by NIH ECHO Cohort

	Healthy Start (N=184)	Emory (N=144)	KP PETALS (N=78)	KPRB (N=12)	NHBCS (N=318)	MARBLES (N=31)	Rochester (N=102)	Project Viva (N=467)	IKIDS (N=26)	CIOB (N=67)
Maternal Age										
<25	36 (19.6%)	64 (44.4%)	<5	<5	21 (6.6%)	0 (0%)	<5	20 (4.3%)	0 (0%)	<5
25-29	41 (22.3%)	43 (29.9%)	<15	<5	88 (27.7%)	6 (19.4%)	<30	69 (14.8%)	7 (26.9%)	<5
30-34	67 (36.4%)	31 (21.5%)	33 (42.3%)	5 (41.7%)	145 (45.6%)	6 (19.4%)	49 (48.0%)	196 (42.0%)	15 (57.7%)	24 (35.8%)
>=35	40 (21.7%)	6 (4.2%)	30 (38.5%)	<5	64 (20.1%)	19 (61.3%)	23 (22.5%)	182 (39.0%)	<5	35 (52.2%)
Maternal Education										
Up To High school degree, GED or Equivalent	33 (17.9%)	80 (55.6%)	8 (10.3%)	0 (0%)	35 (11.0%)	<5	17 (16.7%)	27 (5.8%)	0 (0%)	<5
Some college, no degree	40 (21.7%)	41 (28.5%)	24 (30.8%)	<5	59 (18.6%)	12 (38.7%)	11 (10.8%)	<85	<5	5 (7.5%)
Bachelor's and above	111 (60.3%)	23 (16.0%)	46 (59.0%)	<8	211 (66.4%)	15 (48.4%)	66 (64.7%)	353 (75.6%)	<25	58 (86.6%)
Missing	0 (0%)	0 (0%)	0 (0%)	6 (50.0%)	13 (4.1%)	<5	8 (7.8%)	<5	0 (0%)	<5
Maternal Race										
Hispanic All	36 (19.6%)	<5	30 (38.5%)	7 (58.3%)	9 (2.8%)	10 (32.3%)	8 (7.8%)	38 (8.1%)	<5	9 (13.4%)
Non-Hispanic Black	21 (11.4%)	139 (96.5%)	<5	<5	0 (0%)	0 (0%)	15 (14.7%)	49 (10.5%)	<5	<5
Non-Hispanic Other	11 (6.0%)	<5	26 (33.3%)	<5	8 (2.5%)	<5	7 (6.9%)	40 (8.6%)	<5	21 (31.3%)
Non-Hispanic White	116 (63.0%)	0 (0%)	20 (25.6%)	<5	297 (93.4%)	17 (54.8%)	72 (70.6%)	339 (72.6%)	22 (84.6%)	32 (47.8%)
Missing	0 (0%)	0 (0%)	0 (0%)	0 (0%)	<5	0 (0%)	0 (0%)	<5	0 (0%)	<5
Parity										
Nulliparous	120 (65.2%)	57 (39.6%)	36 (46.2%)	5 (41.7%)	137 (43.1%)	0 (0%)	27 (26.5%)	225 (48.2%)	18 (69.2%)	62 (92.5%)
	64 (34.8%)	87 (60.4%)	42 (53.8%)	7 (58.3%)	157 (49.4%)	<25	<75	242 (51.8%)	8 (30.8%)	<5
Missing	0 (0%)	0 (0%)	0 (0%)	0 (0%)	24 (7.5%)	<5	<5	0 (0%)	0 (0%)	<5

Infant Sex										
Female	86 (46.7%)	66 (45.8%)	42 (53.8%)	7 (58.3%)	168 (52.8%)	12 (38.7%)	48 (47.1%)	255 (54.6%)	18 (69.2%)	32 (47.8%)
Male	98 (53.3%)	78 (54.2%)	36 (46.2%)	5 (41.7%)	150 (47.2%)	19 (61.3%)	54 (52.9%)	212 (45.4%)	8 (30.8%)	35 (52.2%)
Year of Birth										
1991-2000	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	181 (38.8%)	0 (0%)	0 (0%)
2001-2010	40 (21.7%)	0 (0%)	0 (0%)	0 (0%)	42 (13.2%)	6 (19.4%)	0 (0%)	286 (61.2%)	0 (0%)	0 (0%)
2011-2019	144 (78.3%)	144 (100%)	78 (100%)	12 (100%)	276 (86.8%)	25 (80.6%)	102 (100%)	0 (0%)	26 (100%)	67 (100%)
Total SRS Raw Score										
Mean (SD)	28.8 (18.4)	37.0 (22.1)	32.2 (15.2)	NA (NA)	28.1 (15.2)	52.0 (23.3)	29.6 (17.5)	25.6 (18.4)	28.4 (17.8)	29.0 (15.2)
Median [Min, Max]	25.0 [3.00, 130]	33.0 [8.00, 133]	30.0 [7.00, 71.0]	NA [NA, NA]	25.0 [2.00, 133]	51.0 [6.00, 108]	25.5 [6.00, 122]	21.0 [3.00, 129]	24.0 [6.00, 83.0]	27.0 [5.00, 91.0]
Non-Missing	166 (90.2%)	61 (42.4%)	53 (67.9%)	0 (0%)	318 (100%)	31 (100%)	102 (100%)	414 (88.7%)	26 (100%)	53 (79.1%)
Total SRS T-score										
Mean (SD)	48.9 (7.33)	48.8 (8.66)	50.4 (6.01)	NA (NA)	45.8 (6.39)	55.1 (8.48)	45.3 (6.79)	47.7 (7.46)	45.1 (7.46)	45.1 (5.85)
Median [Min, Max]	47.0 [38.0, 88.0]	47.0 [37.0, 85.0]	50.0 [40.0, 65.0]	NA [NA, NA]	45.0 [35.0, 89.0]	54.0 [39.0, 76.0]	44.0 [36.0, 81.0]	46.0 [38.0, 88.0]	43.0 [36.0, 66.0]	44.0 [36.0, 69.0]
Non-Missing	166 (90.2%)	61 (42.4%)	53 (67.9%)	0 (0%)	318 (100%)	31 (100%)	102 (100%)	414 (88.7%)	26 (100%)	53 (79.1%)
Child Age Group at SRS assessment										
SRS Second Edition (SRS-2) Preschool	0 (0%)	52 (36.1%)	0 (0%)	0 (0%)	238 (74.8%)	<22	102 (100%)	0 (0%)	<28	53 (79.1%)
SRS Second Edition (SRS-2) School Age	166 (90.2%)	9 (6.2%)	53 (67.9%)	0 (0%)	80 (25.2%)	<12	0 (0%)	414 (88.7%)	<5	0 (0%)
Missing	18 (9.8%)	83 (57.6%)	25 (32.1%)	12 (100%)	0 (0%)	0 (0%)	0 (0%)	53 (11.3%)	0 (0%)	14 (20.9%)

Clinical ASD dx										
No	175 (95.1%)	139 (96.5%)	<78	12 (100%)	0 (0%)	12 (38.7%)	0 (0%)	448 (95.9%)	<25	65 (97.0%)
Yes	<5	<5	<5	0 (0%)	<5	7 (22.6%)	0 (0%)	<20	<5	<5
Missing	<8	<5	0 (0%)	0 (0%)	<320	12 (38.7%)	102 (100%)	<5	<5	<5

Note that for protection of participant confidentiality in the ECHO data use agreement, we cannot report exact counts in cells with <5 participants.

eTable 3. Distributions of PFAS (untransformed values, µg/L) and % below the limit of detection (LOD).

Analyte	n	% above LOD	5th Percentile	25th Percentile	50th Percentile	75th Percentile	95th Percentile
EtFOSAA	833	59.8	0.014	0.023	0.5	1.2	2.94
NMFOSAA_MeFOSAA	1259	70.7	0.022	0.071	0.153	1.3	3.61
PFDA	1329	70.4	0.025	0.071	0.1	0.2	0.4
PFHXS	1428	99.3	0.281	0.7	1.2	2.1	5.3
PFNA	1428	99	0.144	0.3	0.5	0.7	1.393
PFOA ^a	1429	99.6	0.314	0.8	1.5	3.9	8.9
PFOS ^a	1428	99.8	0.902	1.919	3.35	16.725	39.46
PFUNDA	730	59.5	0.014	0.062	0.071	0.142	0.3

EtFOSAA, 2-(N-Ethyl-perfluorooctane sulfonamido) acetic acid; NMFOSAA_MeFOSAA, N-methyl perfluorooctane sulfonamido acetic acid; PFAS, per- and polyfluoroalkyl substances; PFDA, perfluorodecanoic acid; PFHXS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; PFUNDA, perfluoroundecanoic acid.

^a If a cohort had measured branched and linear chain isomers for PFOA or PFOS separately, the two were summed as total PFOA or PFOS in accordance with the CDC protocol.¹

Reference:

1. CDC. *National Health and Nutrition Examination Survey: 2013-2014 Data Documentation, Codebook, and Frequencies*. 2016. https://wwwn.cdc.gov/Nchs/Nhanes/2013-2014/SSPFAS_H.htm

eTable 4. Associations of PFAS and SRS Raw Scores from Single-Pollutant Models

PFAS analyte	SRS total raw score	
	n _{adj}	β_{adj} (95% CI) ^a
EtFOSAA	675	1.8 (-3.2, 6.8)
NMFOSAA_MeFOSAA	1032	1.3 (-2.0, 4.6)
PFDA	1096	2.1 (-1.3, 5.4)
PFHXS	1168	-2.0 (-5.1, 1.2)
PFNA	1168	3.6 (-0.3, 7.5)
PFOA	1169	0.6 (-3.5, 4.6)
PFOS	1168	0.4 (-3.6, 4.4)
PFuNDA	567	-0.4 (-4.8, 4.1)

CI, confidence interval; EtFOSAA, 2-(N-Ethyl-perfluorooctane sulfonamido) acetic acid; NMFOSAA_MeFOSAA, N-methyl perfluorooctane sulfonamido acetic acid; PFDA, perfluorodecanoic acid; PFHXS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; PFuNDA, perfluoroundecanoic acid; SRS, Social Responsiveness Scale.

^aAdjusted models included ECHO cohort, maternal age, race/ethnicity, educational attainment, parity, SRS version (preschool, school-age), and child sex at birth.

eTable 5. Associations between PFAS and SRS total T-score stratified by SRS-2 form completed

PFAS analyte	Pre-school form of SRS ^a		School age form of SRS ^b	
	n _{adj}	β _{adj} (95% CI) ^c	n _{adj}	β _{adj} (95% CI) ^c
EtFOSAA	195	-0.2 (-8.8, 8.5)	482	0.8 (-1.4, 2.9)
NMFOSAA_MeFOSAA	459	0.0 (-1.7, 1.7)	598	1.1 (-0.9, 3.0)
PFDA	449	-0.7 (-2.8, 1.3)	672	1.6 (-0.2, 3.3)
PFHXS	474	-0.8 (-2.7, 1.1)	719	-0.7 (-2.3, 0.9)
PFNA	474	1.0 (-1.2, 3.2)	719	1.7 (-0.5, 3.9)
PFOA	474	-1.3 (-3.5, 0.9)	720	1.6 (-0.7, 3.9)
PFOS	474	-0.8 (-2.9, 1.4)	719	1.2 (-1.1, 3.5)
PFuNDA	449	0.6 (-1.2, 2.4)	143	-3.2 (-8.2, 1.7)

CI, confidence interval; EtFOSAA, 2-(N-Ethyl-perfluorooctane sulfonamido) acetic acid; NMFOSAA_MeFOSAA, N-methyl perfluorooctane sulfonamido acetic acid; PFDA, perfluorodecanoic acid; PFHXS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; PFuNDA, perfluoroundecanoic acid; SRS, Social Responsiveness Score.

^aRestricted to participants who completed the pre-school version of the SRS (generally 2.5-4.5 years old)

^bRestricted to participants who completed the child version of SRS (generally 4-18 years old)

^c Adjusted models included ECHO cohort, maternal age, race/ethnicity, educational attainment, and parity. T-score is standardized for SRS version (preschool, school-age) and child's sex.

eTable 6. Sensitivity analysis dropping samples below the LOD for the specific analytes with >20% values below the LOD.

PFAS analyte	SRS total T-score	
	n _{adj}	β _{adj} (95% CI) ^a
EtFOSAA	431	0.5 (-1.8, 2.8)
NMFOSAA_MeFOSAA	753	1.2 (-0.4, 2.9)
PFDA	798	0.2 (-1.9, 2.2)
PFuNDA	355	-0.9 (-3.5, 1.7)

EtFOSAA, 2-(N-Ethyl-perfluorooctane sulfonamido) acetic acid; NMFOSAA_MeFOSAA, N-methyl perfluorooctane sulfonamido acetic acid; PFAS, per- and polyfluoroalkyl substances; PFDA, perfluorodecanoic acid; PFuNDA, perfluoroundecanoic acid; SRS, Social Responsiveness Score.

^a Adjusted models included ECHO cohort, maternal age, race/ethnicity, educational attainment, and parity. T-score is standardized for SRS version (preschool, school-age) and child's sex.

eTable 7. Associations of PFAS and T-scores on SRS subscales from single-pollutant models

PFAS analyte	SRS Social Communication and Interaction subscale (T-score)		SRS Restricted Interests and Repetitive Behavior subscale (T-score)
	n _{adj}	β_{adj} (95% CI) ^a	β_{adj} (95% CI) ^a
EtFOSAA	677	1.2 (-0.9, 3.2)	0.6 (-1.5, 2.7)
NMFOSAA_MeFOSAA	1057	0.7 (-0.6, 2.1)	0.1 (-1.3, 1.5)
PFDA	1121	1.1 (-0.3, 2.4)	0.3 (-1.1, 1.7)
PFHXS	1193	-0.8 (-2.0, 0.5)	-0.8 (-2.1, 0.6)
PFNA	1193	1.8 (0.2, 3.4)	1.1 (-0.6, 2.8)
PFOA	1194	0.8 (-0.9, 2.4)	-0.1 (-1.9, 1.6)
PFOS	1193	0.1 (-1.5, 1.8)	0.2 (-1.6, 1.9)
PFUNDA	592	-0.2 (-2, 1.7)	-0.2 (-2.1, 1.8)

EtFOSAA, 2-(N-Ethyl-perfluorooctane sulfonamido) acetic acid; NMFOSAA_MeFOSAA, N-methyl perfluorooctane sulfonamido acetic acid; PFAS, per- and polyfluoroalkyl substances; PFDA, perfluorodecanoic acid; PFHXS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; PFUNDA, perfluoroundecanoic acid; SRS, Social Responsiveness Score.

^aAdjusted models included ECHO cohort, maternal age, race/ethnicity and educational attainment, parity, and SRS version (preschool, school-age). T-score is standardized by SRS version (preschool, school-age) and child's sex.

eTable 8. Associations of PFAS and ASD diagnoses from single-pollutant models

PFAS analyte	ASD clinical diagnosis		
	n _{crude} / n _{adj}	OR _{crude} (95% CI) ^a	OR _{adj} (95% CI) ^b
EtFOSAA	716/710	1.5 (0.4, 5.5)	1.6 (0.4, 6.2)
NMFOSAA_MeFOSAA	821/809	2.1 (0.6, 7.1)	2.0 (0.6, 6.9)
PFDA	890/878	0.9 (0.3, 2.7)	0.9 (0.3, 3.1)
PFHXS	986/974	0.9 (0.3, 2.6)	0.8 (0.3, 2.3)
PFNA	986/974	0.7 (0.2, 2.7)	0.7 (0.2, 2.7)
PFOA	987/975	0.6 (0.2, 2.0)	0.5 (0.1, 2.2)
PFOS	986/974	0.6 (0.1, 2.3)	0.4 (0.1, 1.8)
PFUNDA	297/288	2.0 (0.3, 15.2)	3.2 (0.4, 28.3)

ASD, autism spectrum disorder; EtFOSAA, 2-(N-Ethyl-perfluorooctane sulfonamido) acetic acid; NMFOSAA_MeFOSAA, N-methyl perfluorooctane sulfonamido acetic acid; PFAS, per- and polyfluoroalkyl substances; PFDA, perfluorodecanoic acid; PFHXS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; PFUNDA, perfluoroundecanoic acid;

^aAdjusted for ECHO cohort.

^bAdjusted models included ECHO cohort, maternal age, race/ethnicity, educational attainment, parity, and child sex at birth.

eTable 9. Associations of PFAS and ASD diagnosis in mixture models using Bayesian Weighted Sums (n=809)

Weighted sums	ASD diagnosis	
	OR _{adj}	95% HPD ^a
Summed effect	1.4	(0.2, 10.2)
w(PFOA) ^b	0.17	(0.00, 0.44)
w(PFOS) ^b	0.16	(0.00, 0.45)
w(PFNA) ^b	0.16	(0.00, 0.44)
w(PFHxS) ^b	0.16	(0.00, 0.44)
w(NMFOSAA_MeFOSAA) ^b	0.18	(0.00, 0.48)
w(PFDA) ^b	0.16	(0.00, 0.44)
Individual association^c		
PFOA	1.1	(0.3, 3.2)
PFOS	0.9	(0.3, 2.8)
PFNA	1.1	(0.3, 3.3)
PFHxS	1.0	(0.4, 2.4)
NMFOSAA_MeFOSAA	1.4	(0.6, 3.9)
PFDA	1.1	(0.4, 2.6)

ASD, autism spectrum disorder; HPD, Highest Posterior Density; NMFOSAA_MeFOSAA, N-methyl perfluorooctane sulfonamido acetic acid; PFAS, per- and polyfluoroalkyl substances; PFDA, perfluorodecanoic acid; PFHxS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; PFuNDA, perfluoroundecanoic acid;

^aAdjusted models included ECHO cohort, maternal age, race/ethnicity, educational attainment, parity, and child sex at birth.

^bPercent (%) weight contributed by each mixture component to the summed association. Weight and 95% HPD reported to two decimal places.

^cCo-adjusted for other PFAS using semi-Bayesian shared mean approach.

eTable 10. Sex-stratified associations between PFAS and SRS total T-score

PFAS analyte	SRS total T-score		P _{interaction} ^b
	Males β_{adj} (95% CI) ^a	Females β_{adj} (95% CI) ^a	
EtFOSAA	1.1 (-2.1, 4.4)	0.3 (-2.2, 2.8)	0.34
NMFOSAA_MeFOSAA	-0.4 (-2.5, 1.6)	1.7 (0, 3.4)	0.06
PFDA	0.6 (-1.6, 2.7)	1.1 (-0.6, 2.8)	0.55
PFHXS	-0.7 (-2.7, 1.4)	-1 (-2.5, 0.5)	0.55
PFNA	2.2 (-0.3, 4.7)	0.7 (-1.3, 2.6)	0.79
PFOA	0.9 (-1.8, 3.5)	-0.3 (-2.2, 1.7)	0.20
PFOS	-0.8 (-3.5, 1.8)	1.3 (-0.6, 3.2)	0.02
PFUNDA	0.2 (-2.4, 2.9)	-0.3 (-2.4, 1.9)	0.81

EtFOSAA, 2-(N-Ethyl-perfluorooctane sulfonamido) acetic acid; NMFOSAA_MeFOSAA, N-methyl perfluorooctane sulfonamido acetic acid; PFAS, per- and polyfluoroalkyl substances; PFDA, perfluorodecanoic acid; PFHXS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; PFUNDA, perfluoroundecanoic acid; SRS, Social Responsiveness Scale.

^a Adjusted models included ECHO cohort, maternal age, race/ethnicity, educational attainment and parity and SRS version (preschool, school-age). T-score is standardized for child's sex.

^b P-value obtained from an interaction model which included both a first order term for sex and the cross-product term of child sex and PFAS analyte added to adjusted model. We considered P-interaction<0.05 to be statistically significant.

eTable 11. Sex-stratified associations between PFAS and SRS using Bayesian Weighted Sums

Weighted sums	SRS total T-score			
	Male		Female	
	β_{adj}	95% HPD ^a	β_{adj}	95% HPD ^a
Summed association	-0.6	(-4.1, 2.8)	1.5	(-1.2, 4.3)
w(PFOA) ^b	0.18	(0.00, 0.46)	0.14	(0.00, 0.39)
w(PFOS) ^b	0.16	(0.00, 0.44)	0.21	(0.00, 0.51)
w(PFNA) ^b	0.15	(0.00, 0.43)	0.15	(0.00, 0.42)
w(PFHxS) ^b	0.17	(0.00, 0.44)	0.12	(0.00, 0.37)
w(NMFOSAA_MeFOSAA) ^b	0.17	(0.00, 0.45)	0.20	(0.00, 0.51)
w(PFDA) ^b	0.17	(0.00, 0.44)	0.17	(0.00, 0.45)
Individual association^c				
PFOA	-0.9	(-4.4, 1.7)	-0.5	(-3.1, 1.6)
PFOS	1.9	(-1.6, 4.5)	1.9	(-0.6, 5.0)
PFNA	1.9	(-1.0, 5.1)	-0.1	(-2.5, 2.1)
PFHxS	-0.5	(-2.6, 1.4)	-1.3	(-3.3, 0.5)
NMFOSAA_MeFOSAA	-0.8	(-2.9, 0.9)	0.9	(-0.6, 2.5)
PFDA	-1.3	(-3.8, 0.8)	0.6	(-1.2, 2.3)

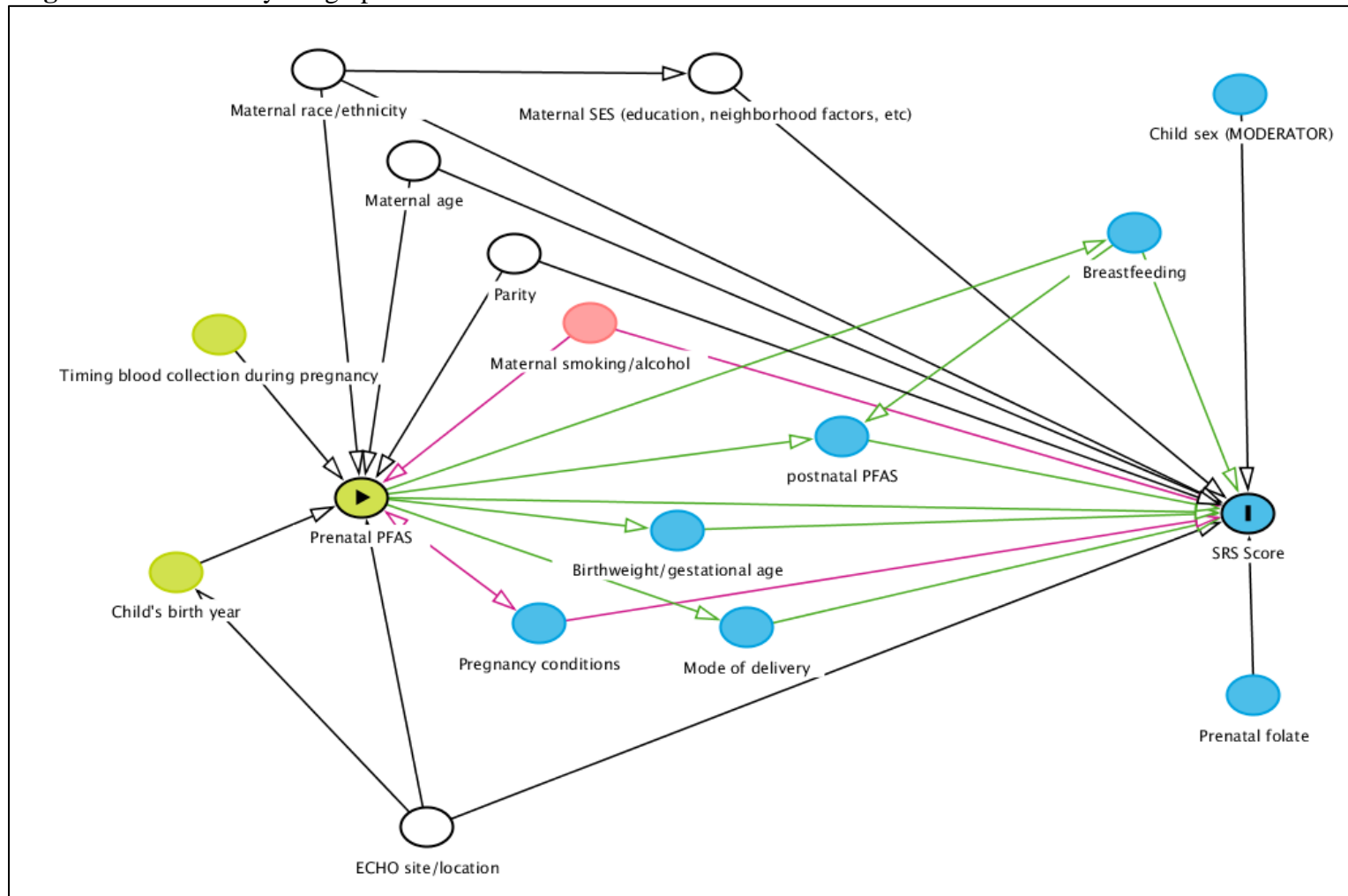
HPD, Highest Posterior Density; NMFOSAA_MeFOSAA, N-methyl perfluorooctane sulfonamido acetic acid; PFAS, per- and polyfluoroalkyl substances; PFDA, perfluorodecanoic acid; PFHxS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; PFuNDA, perfluoroundecanoic acid; SRS, Social Responsiveness Scale.

^a Adjusted models included ECHO cohort, maternal age, race/ethnicity, educational attainment, parity, and SRS version (preschool, school-age).

^b Percent (%) weight contributed by each mixture component to the summed association. Weight and 95% HPD reported to two decimal places.

^c Co-adjusted for other PFAS using semi-Bayesian approach

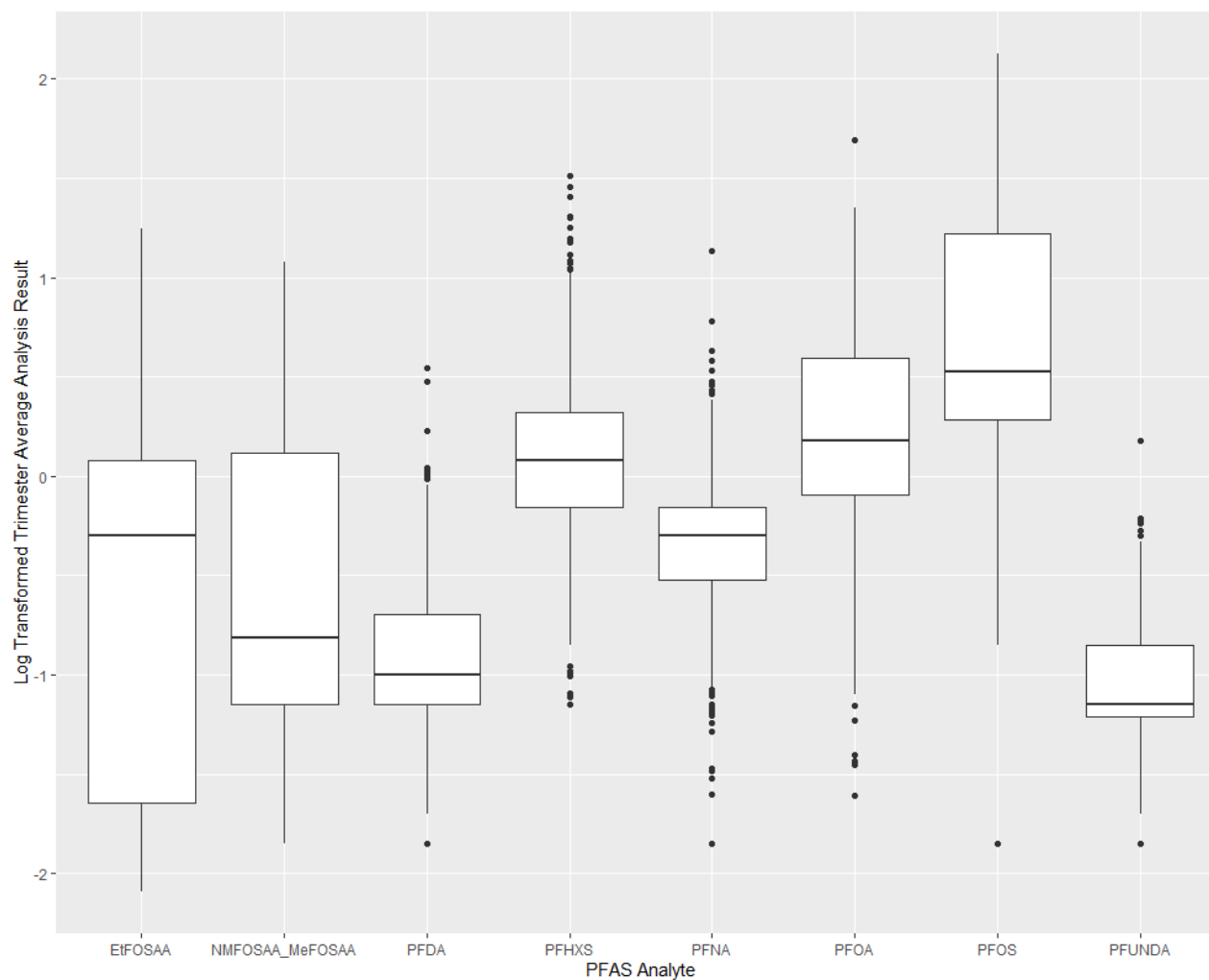
eFigure 1. Directed acyclic graph^a



ECHO, Environmental influences on Child Health Outcomes; SES, socioeconomic status.

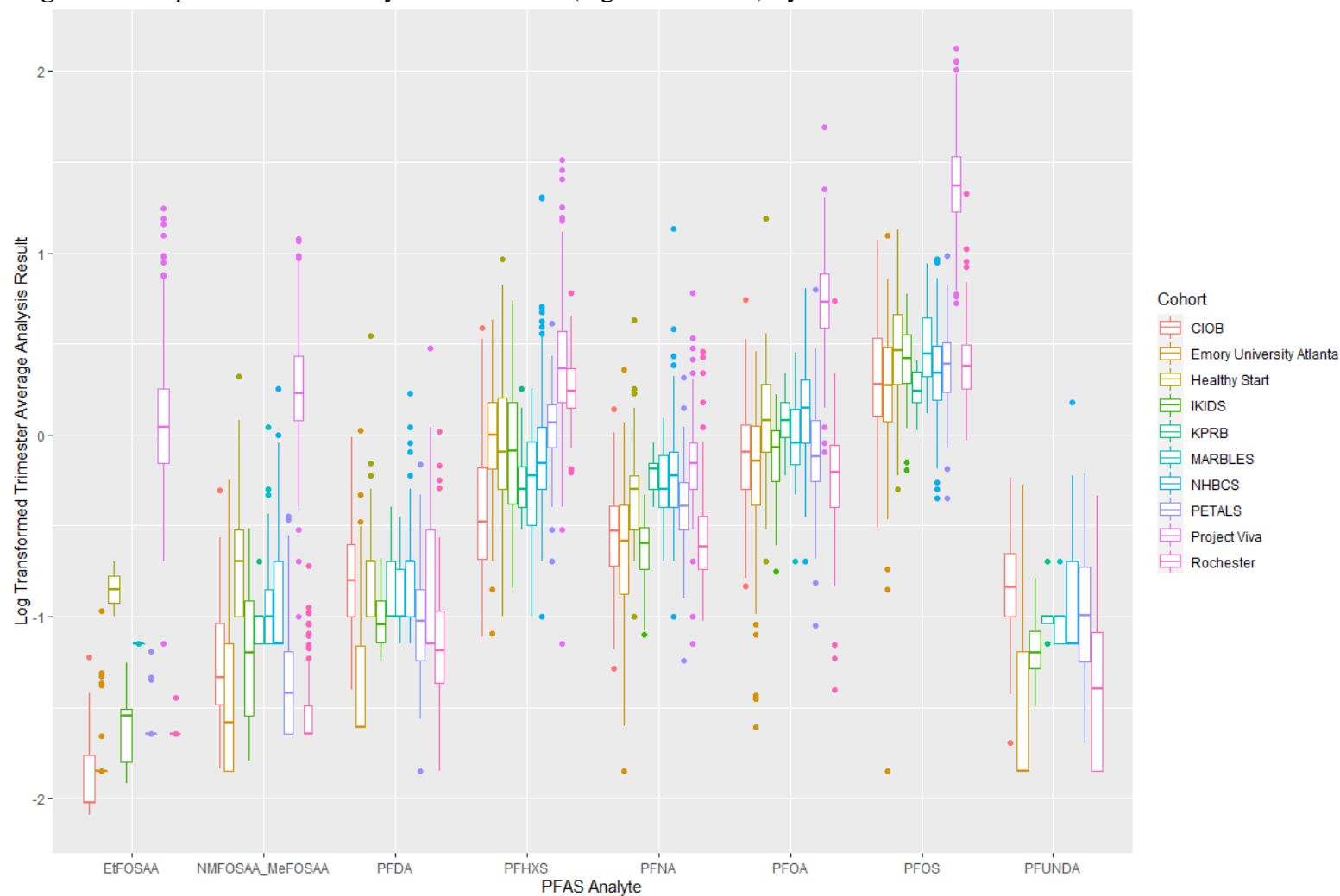
^a Potential mediators (e.g., birth weight, gestational age, pregnancy complications) were not included in the final analytic models.

eFigure 2. Boxplots of PFAS analytes (log-transformed) in the pooled ECHO sample



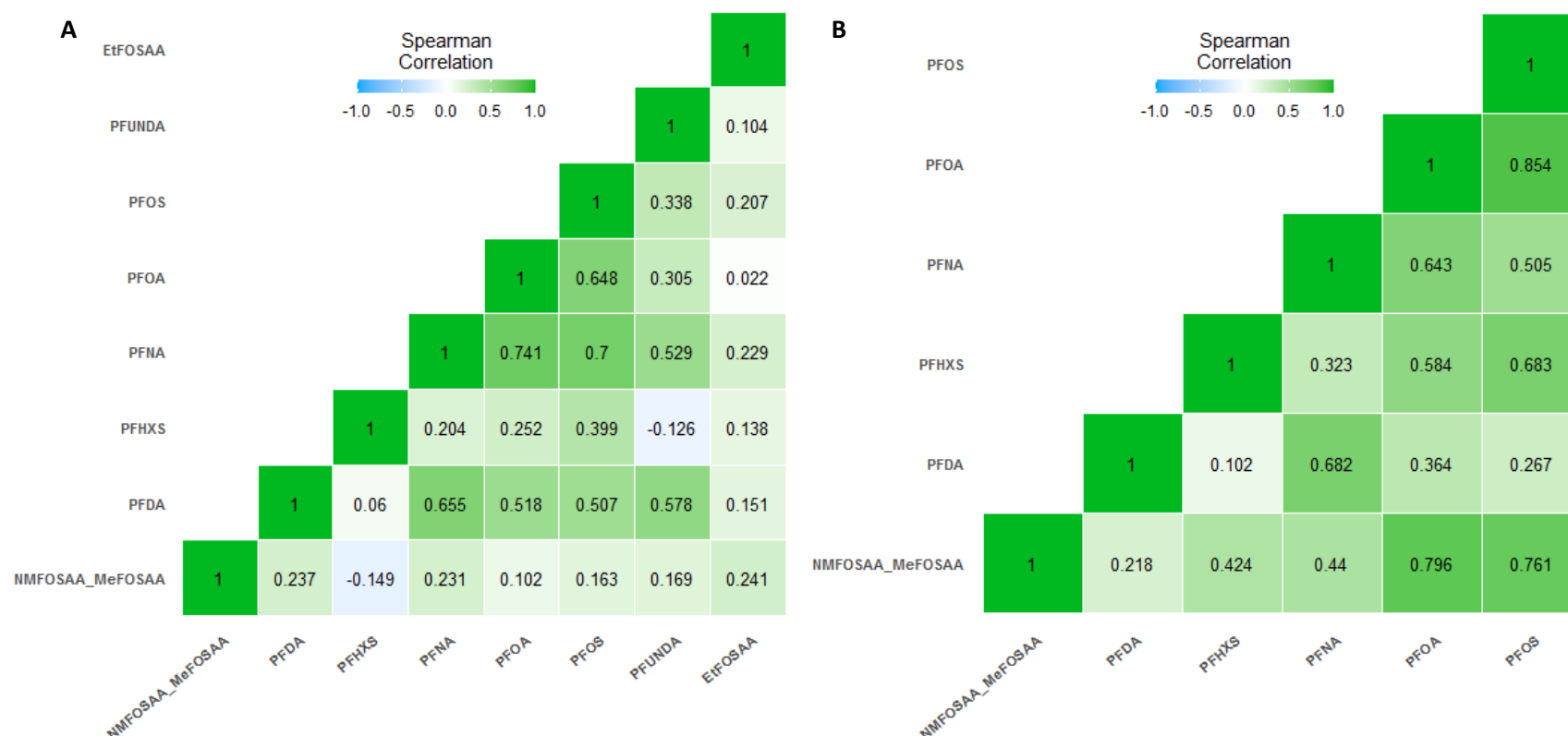
EtFOSAA, 2-(N-Ethyl-perfluorooctane sulfonamido) acetic acid; NMFOSAA_MeFOSAA, N-methyl perfluorooctane sulfonamido acetic acid; PFAS, per- and polyfluoroalkyl substances; PFDA, perfluorodecanoic acid; PFHXS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; PFUNDA, perfluroundecanoic acid.

eFigure 3. Boxplots of PFAS analyte distributions (log-transformed) by ECHO cohort



EtFOSAA, 2-(N-Ethyl-perfluorooctane sulfonamido) acetic acid; NMFOSAA_MeFOSAA, N-methyl perfluorooctane sulfonamido acetic acid; PFAS, per- and polyfluoroalkyl substances; PFDA, perfluorodecanoic acid; PFHXS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; PFUNDA, perfluoroundecanoic acid.

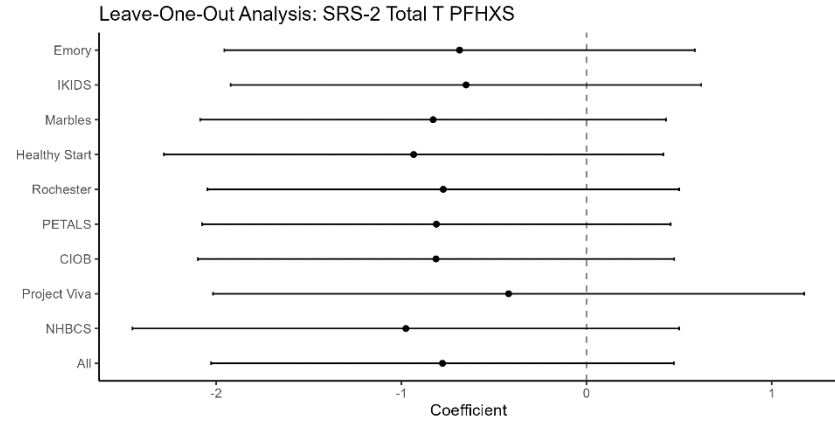
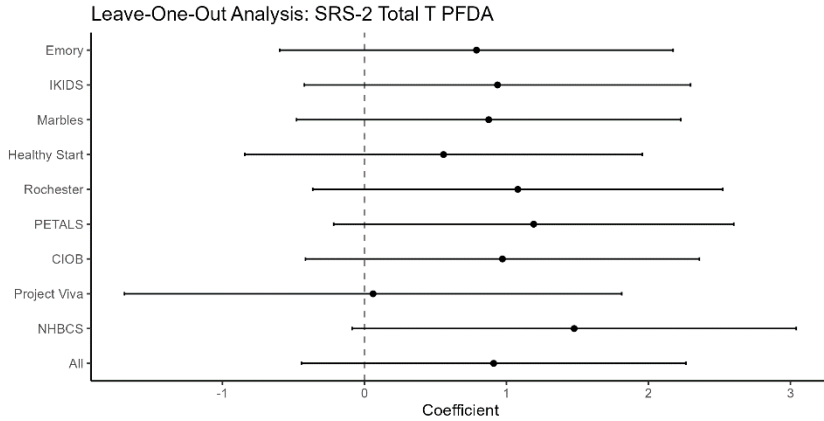
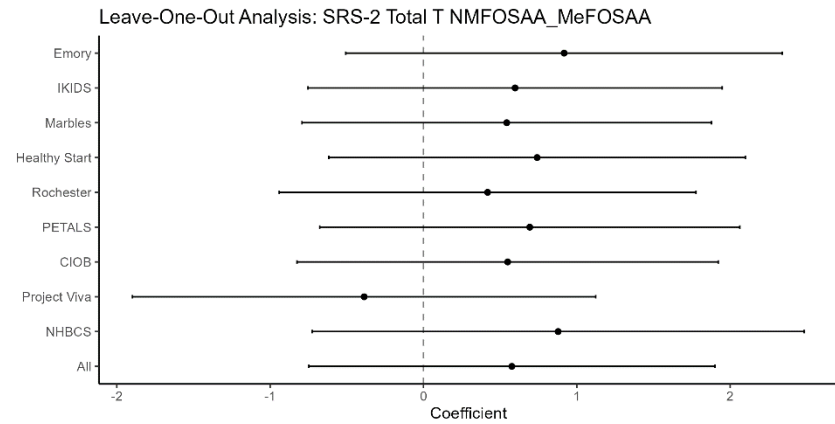
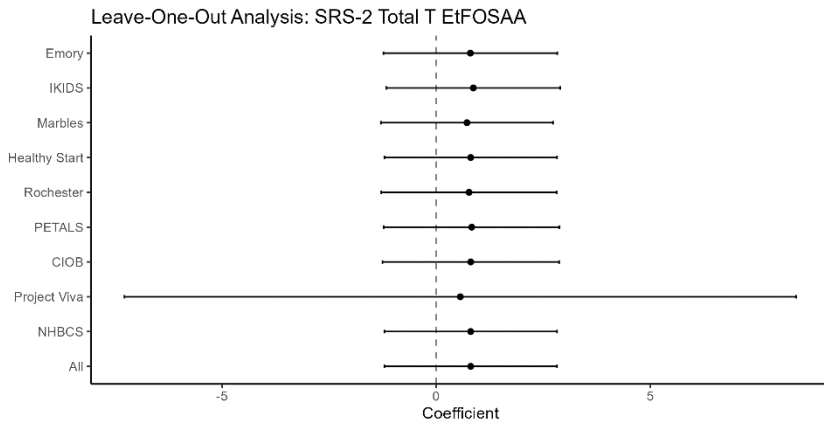
eFigure 4. Correlation plots of PFAS analytes

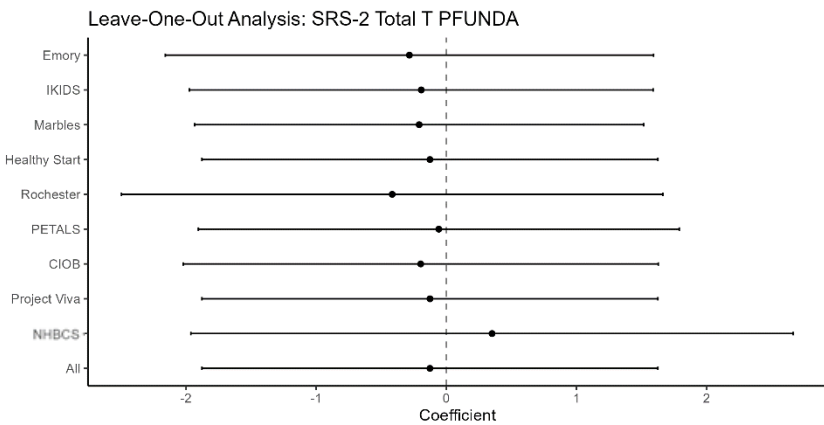
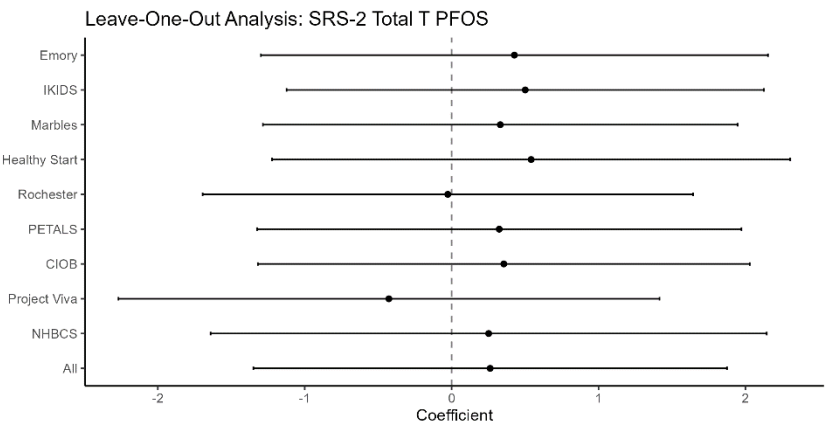
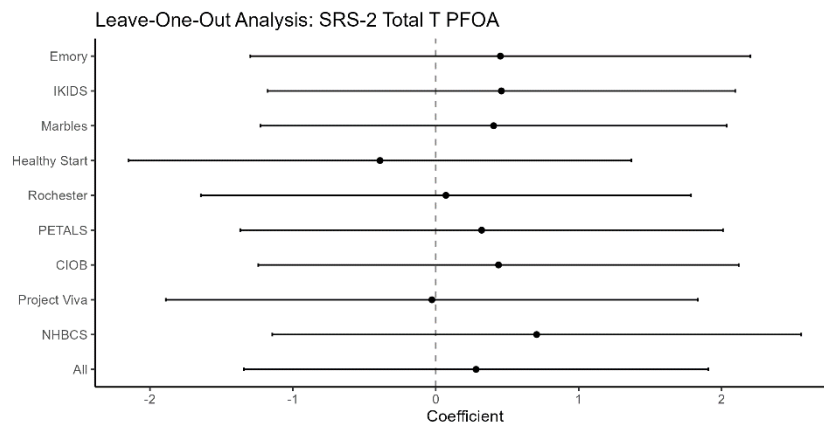
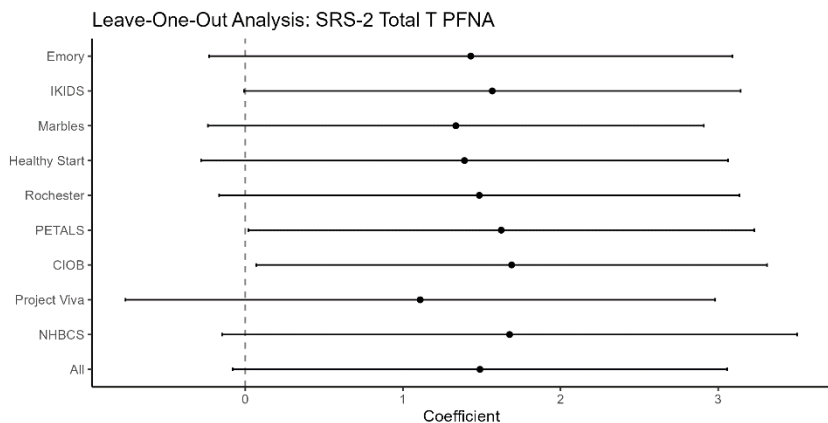


A) Correlation among all 8 PFAS passing original detection criteria (n=360 due to some analytes not being measured in all individuals); B) Correlation among 6 PFAS after using stricter detection criteria (n=1231).

EtFOSAA, 2-(N-Ethyl-perfluorooctane sulfonamido) acetic acid; NMFOSAA_MeFOSAA, N-methyl perfluorooctane sulfonamido acetic acid; PFAS, per- and polyfluoroalkyl substances; PFDA, perfluorodecanoic acid; PFHXS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; PFUNDA, perfluoroundecanoic acid.

eFigure 5. Coefficient plots of leave-one-out results for SRS total T-score. The Y-axis lists the name of the cohort that was left out of the regression model. All models were adjusted for ECHO cohort, maternal age, race/ethnicity, educational attainment and parity and SRS version (preschool, school-age). T-score is standardized by SRS version and child's sex.





EtFOSAA, 2-(N-Ethyl-perfluorooctane sulfonamido) acetic acid; NMFOSAA_MeFOSAA, N-methyl perfluorooctane sulfonamido acetic acid; PFAS, per- and polyfluoroalkyl substances; PFDA, perfluorodecanoic acid; PFHXS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; PFuNDA, perfluoroundecanoic acid; SRS, Social Responsiveness Scale.