

Supplementary Material

Association of exposure to perfluoroalkyl substances (PFAS) and phthalates with thyroid hormones in adolescents from HBM4EU aligned studies

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Figure S1. Directed acyclic graph (DAG) used to determine confounders for all models.

Figure S2. Mixture effect of PFAS/phthalate metabolites on thyroid hormones in males (n=317).

Figure S3. Mixture effect of PFAS/phthalate metabolites on thyroid hormones in females (n=406).

Table S1. Available limits of detection (LOD), limits of quantification (LOQ), and frequency of quantification (FOQ) for serum PFAS in HBM4EU aligned studies.

Study		PFHxA	PFOA	PFNA	PFBS	PFHxS	PFOS	PFPA	PFHpA	PFDA	PFUnDA	PFDoDA	PFHpS
FLEHS IV	LOD	0.10	0.00	0.01	0.01	0.01	0.01	0.00	0.00	0.01	0.00	0.01	0.00
	LOQ	0.02	0.02	0.02	0.04	0.02	0.04	0.01	0.10	0.03	0.01	0.04	0.01
	FOQ %	3.00	100	82.00	0.33	97.67	100	0.00	1.30	47.67	9.00	1.67	3.00
PCB	LOD	0.10	0.00	0.01	0.01	0.01	0.01	0.00	0.00	0.01	0.00	0.01	0.00
	LOQ	0.02	0.02	0.02	0.04	0.02	0.04	0.01	0.10	0.03	0.01	0.04	0.01
	FOQ %	99.7	99.7	93.8	0.75	99.7	100	52.7	90.4	67.8	42.5	0.003	82.5
BEA	LOD	-	-	-	-	-	-	-	-	-	-	-	-
	LOQ	0.25	0.16	0.16	0.10	0.34	0.33	0.16	0.20	0.20	0.20	0.12	0.25
	FOQ %	0.00	100	89.3	0.00	2.01	100	0.00	0.00	15.7	8.36	1.67	5.35

Table S2. Available limits of detection (LOD), limits of quantification (LOQ), and frequency of quantification (FOQ) for urinary phthalates/DINCH® in HBM4EU aligned studies.

		MBzP	MiBP	MnBP	MEHP	OH-MEHP	oxo-MEHP	cx-MEPP	MEP	MHNP	MCOP	OH-MiDP	cx-MiDP	MCHP	MOP	MnPEP	OH-MINCH	cx-MINCH
FLESH IV	LOD																	
	LOQ	0.20	0.50	0.50	0.50	0.20	0.20	0.50	0.50	0.20	0.20	0.20	0.20	*	*	*	0.20	0.20
	FOQ %	98.33	100	100	81.33	99.67	99.33	100	100	100	99.67	91.67	100				95.67	98.33
PCB	LOD	0.10	0.07	0.18	0.18	0.07	0.07	0.07	0.02	0.30	0.20	0.20		0.20	0.20		0.05	0.03
	LOQ	0.30	0.20	0.40	0.50	0.20	0.20	0.20	0.60	0.70	0.40	0.60		0.60	0.50		0.14	0.10
	FOQ %	97.64	98.95	99.30	97.91	99.65	100	99.30	100	100	98.95	13.59		1.05	1.05		100	99.30
BEA	LOD																	
	LOQ	0.20	1.00	1.00	0.30	0.20	0.20	0.20	0.50	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.05	0.05
	FOQ %	93.33	99.67	99.67	97.33	100	100	100	100	100	100	96.33	96.67	0	0	0.67	100	99

Table S3. Spearman correlation analysis between serum PFAS and urinary phthalate and DINCH® metabolites

	PFOA	PFNA	PFOS	MBzP	MiBP	MnBP	MEHP	OH-MEHP	oxo-MEHP	cx-MEPP	MEP	MHNP	MCOP	OH-MINCH	cx-MINCH
PFOA		0.54*	0.58*	-0.02	-0.06	-0.12*	-0.25*	-0.24*	-0.25*	0.05	-0.26*	-0.16*	-0.39*	-0.17*	-0.03
PFNA	0.54*		0.66*	0.06	-0.05	-0.24*	-0.18*	-0.28*	-0.10*	0.12*	0.03	-0.25*	-0.15*	0.02	0.14*
PFOS	0.58*	0.66*		-0.01	-0.07*	-0.13*	-0.17*	-0.20*	-0.19*	0.01	-0.15*	-0.12*	-0.26*	-0.09*	0.03
MBzP	-0.02	0.06	-0.01		0.34*	0.14*	0.17*	0.08*	0.32*	0.38*	0.09*	0.07	0.12*	0.09*	0.06
MiBP	-0.06	-0.05	-0.07*	0.34*		0.50*	0.37*	0.37*	0.39*	0.25*	0.21*	0.23*	0.22*	0.08*	-0.02
MnBP	-0.12*	-0.24*	-0.13*	0.14*	0.50*		0.43*	0.61*	0.35*	-0.02	0.10*	0.49*	0.28*	0.00	-0.19*
MEHP	-0.25*	-0.18*	-0.02*	0.17*	0.37*	0.43*		0.69*	0.72*	0.36*	0.28*	0.49*	0.52*	0.16*	0.00
OH-MEHP	-0.24*	-0.28*	-0.20*	0.08*	0.37*	0.61*	0.69*		0.77*	0.27*	0.21*	0.59*	0.48*	0.13*	-0.10*
oxo-MEHP	-0.26*	-0.10*	-0.19*	0.32*	0.39*	0.35*	0.72*	0.77*		0.63*	0.34*	0.39*	0.49*	0.22*	0.04
cx-MEPP	0.05	0.12*	0.01	0.38*	0.25*	-0.02	0.36*	0.27*	0.63*		0.14*	0.09*	0.16*	0.13*	0.21*
MEP	-0.26*	0.03	-0.15*	0.09*	0.21*	0.10*	0.28*	0.21*	0.34*	0.14*		0.13*	0.41*	0.20*	0.11*
MHNP	-0.16*	-0.25*	-0.12*	0.07	0.23*	0.49*	0.48*	0.59*	0.39*	0.09*	0.13*		0.66*	0.14*	-0.04
MCOP	-0.39*	-0.15*	-0.26*	0.12*	0.22*	0.28*	0.52*	0.48*	0.49*	0.16*	0.42*	0.66*		0.29*	0.10*
OH-MINCH	-0.17*	0.02	-0.09*	0.09*	0.08*	0.00	0.16*	0.13*	0.22*	0.13*	0.20*	0.14*	0.29*		0.81*
cx-MINCH	-0.03	0.14*	0.03	0.06	-0.02	-0.19*	0.00	-0.10*	0.04	0.21*	0.11*	-0.04	0.10*	0.82*	

*p<0.05

Table S4. Associations of PFAS and phthalate/DINCH® metabolites categorized into quartiles with thyroid hormones in adolescent from HBM4EU aligned studies (n=773).

Exposure biomarkers	Quartiles (ref: 1st quartile)	TSH			FT4			FT3			FT4/TSH		
		%	95% CI		%	95% CI		%	95% CI		%	95% CI	
			LL	UL		LL	UL		LL	UL		LL	UL
PFOA	Q2	-5.44	-14.77	4.92	-3.45*	-6.68	-0.10	0.31	-2.85	3.57	2.11	-8.75	14.25
	Q3	-4.46	-13.99	6.13	-7.16*	-10.30	-3.91	1.71	-1.52	5.05	-2.83	-13.27	8.87
	Q4	8.87	-2.18	21.16	-10.25*	-13.34	-7.04	2.38	-0.94	5.80	-17.55*	-26.56	-7.44
PFNA	Q2	-7.13	-16.58	3.39	-1.93	-5.41	1.68	0.66	-2.60	4.03	5.60	-6.05	18.71
	Q3	-5.29	-15.28	5.86	0.36	-3.33	4.20	0.32	-3.05	3.80	5.97	-6.14	19.64
	Q4	-1.05	-11.80	11.02	-0.94	-4.70	2.97	0.38	-3.10	3.98	0.11	-11.68	13.48
PFOS	Q2	-5.63	-15.01	4.78	0.56	-2.91	4.15	-0.72	-3.85	2.52	6.56	-4.89	19.39
	Q3	2.80	-7.67	14.46	-2.58	-6.03	0.99	0.78	-2.48	4.15	-5.24	-15.68	6.49
	Q4	3.36	-7.21	15.13	-4.30*	-7.70	-0.78	2.25	-1.08	5.68	-7.41	-17.65	4.10
MEP	Q2	-10.77*	-19.73	-0.82	8.33*	4.67	12.12	-3.35*	-6.42	-0.18	21.41*	8.31	36.10
	Q3	-8.93†	-18.09	1.25	10.66*	6.90	14.54	-4.36*	-7.41	-1.21	21.51*	8.37	36.25
	Q4	-8.81†	-18.02	1.44	12.05*	8.23	16.00	-4.19*	-7.26	-1.02	22.88*	9.53	37.85
MBzP	Q2	-2.70	-12.46	8.14	3.55†	-0.05	7.27	-3.24*	-6.32	-0.07	6.42	-5.13	19.39
	Q3	-4.68	-14.31	6.03	3.53†	-0.10	7.28	-3.78*	-6.86	-0.60	8.62	-3.27	21.96
	Q4	-7.97	-17.36	2.49	2.33	-1.29	6.08	-3.78*	-6.89	-0.56	11.19†	-1.09	25.00
MiBP	Q2	-6.16	-15.44	4.13	3.82*	0.27	7.50	-1.02	-4.14	2.19	10.64†	-1.21	23.91
	Q3	-5.40	-14.77	5.00	3.14†	-0.39	6.81	-0.63	-3.76	2.61	9.03	-2.67	22.14
	Q4	-10.94*	-19.89	-0.99	0.67	-2.83	4.30	-1.59	-4.74	1.67	13.03*	0.73	26.84
MnBP	Q2	-7.59	-16.77	2.59	1.90	-1.60	5.53	-1.56	-4.66	1.65	10.27	-1.58	23.56
	Q3	-5.47	-15.22	5.41	-0.91	-4.46	2.76	-0.87	-4.13	2.49	4.82	-6.90	18.00

	Q4	-5.71	-18.16	8.64	-3.02	-7.51	1.68	-2.66	-6.79	1.66	2.85	-11.84	19.98
MEHP	Q2	-5.40	-14.88	5.12	1.88	-1.66	5.55	0.90	-2.31	4.22	7.70	-3.95	20.78
	Q3	-11.76*	-20.65	-1.86	4.98*	1.30	8.78	-0.75	-3.93	2.54	18.96*	6.00	33.51
	Q4	-14.16*	-23.29	-3.95	3.50†	-0.33	7.47	-2.86†	-6.16	0.55	20.58*	6.72	36.23
	Q2	-14.58*	-23.01	-5.22	4.22*	0.64	7.92	-1.38	-4.48	1.83	22.00*	8.98	36.58
OH-MEHP	Q3	-9.69	-19.00	0.68	5.41*	1.63	9.34	-3.52*	-6.70	-0.24	16.73*	3.72	31.36
	Q4	-19.33*	-29.28	-7.98	3.50	-0.98	8.18	-4.34*	-8.14	-0.38	28.30*	11.20	48.02
	Q2	-1.55	-11.26	9.22	0.33	-3.09	3.87	2.23	-0.98	5.55	1.91	-8.92	14.03
oxo-MEHP	Q3	8.54	-2.24	20.52	-1.90	-5.27	1.59	3.69*	0.41	7.08	-9.62†	-19.30	1.21
	Q4	17.29*	5.53	30.37	-6.34*	-9.59	-2.97	4.07*	0.75	7.51	-20.15*	-28.77	-10.47
	Q2	2.44	-8.15	14.24	2.26	-1.41	6.06	-3.50*	-6.66	-0.23	-0.18	-11.36	12.42
cx-MEPP	Q3	-3.68	-14.00	7.87	0.05	-3.67	3.92	-1.31	-4.67	2.17	3.87	-8.19	17.52
	Q4	-0.95	-11.64	11.02	-0.65	-4.38	3.21	-1.89	-5.26	1.59	0.30	-11.42	13.58
	Q2	-3.76	-13.32	6.86	1.54	-1.95	5.15	0.18	-2.98	3.45	5.51	-5.86	18.24
MHNP	Q3	-2.27	-12.49	9.16	3.31†	-0.43	7.20	1.04	-2.33	4.52	5.71	-6.27	19.23
	Q4	0.09	-11.85	13.66	-1.13	-5.24	3.15	-0.23	-4.04	3.74	-1.23	-13.99	13.43
	Q2	-13.82*	-22.64	-4.00	9.07*	5.31	12.96	-0.74	-3.96	2.60	26.56*	12.65	42.18
MCOP	Q3	-13.07*	-22.16	-2.90	10.53*	6.62	14.57	-4.24*	-7.43	-0.94	27.14*	12.85	43.24
	Q4	-10.69*	-20.21	-0.03	14.24*	10.13	18.50	-3.30†	-6.58	0.10	27.91*	13.26	44.44
	Q2	-5.99	-15.32	4.37	3.92*	0.37	7.61	-2.02	-5.11	1.17	10.54†	-1.31	23.83
OH-MINCH	Q3	-9.69†	-18.70	0.31	6.10*	2.45	9.88	0.20	-2.98	3.48	17.48*	4.82	31.68
	Q4	-5.97	-15.31	4.41	5.12*	1.52	8.85	-0.73	-3.87	2.50	11.79†	-0.22	25.23
	Q2	-5.28	-15.03	5.58	1.77	-1.86	5.54	-1.08	-4.31	2.26	7.45	-4.52	20.91
cx-MINCH	Q3	-5.73	-15.57	5.26	2.73	-1.00	6.59	-0.22	-3.53	3.20	8.97	-3.35	22.85
	Q4	-0.02	-10.37	11.53	2.26	-1.42	6.07	2.27	-1.09	5.74	2.27	-9.19	15.18

Model adjusted by study or origin, age, sex, BMI z-score (continuous), household education, and urinary creatinine concentration (only for phthalates). Estimates (%) are expressed as the percentage of difference in thyroid hormone level for each one-quartile increase in the exposure biomarker. CI: Confidence interval; LL: Lower level; UL: Upper level. * $p \leq 0.05$; † $p < 0.10$

Table S5. Associations of serum PFAS and urinary phthalate/DINCH® metabolite concentrations with thyroid hormones excluding zBMI from the model (n=733).

Exposure biomarkers	TSH			FT4			FT3			FT4/TSH		
	%	95% CI		%	95% CI		%	95% CI		%	95% CI	
		LL	UL		LL	UL		LL	UL		LL	UL
PFAS												
PFOA	3.02	-4.47	11.09	-9.01*	-11.21	-6.76	1.13	-1.18	3.51	-11.68*	-18.60	-4.16
PFNA	0.92	-4.23	6.34	-0.59	-2.32	1.18	0.90	-0.71	2.54	-1.49	-6.94	4.28
PFOS	2.71	-2.61	8.31	-2.55*	-4.27	-0.80	1.43†	-0.21	3.10	-5.12†	-10.44	0.52
∑PFAS	3.44	-3.16	10.48	-4.53*	-6.60	-2.41	1.68	-0.35	3.76	-7.70*	-14.06	-0.86
Phthalate/DINCH® metabolites												
MEP	-1.78	-4.61	1.14	2.67*	1.68	3.66	-0.66	-1.55	0.24	4.53*	1.27	7.90
MBzP	-3.13*	-6.44	0.28	0.63	-0.54	1.80	-0.81	-1.86	0.25	3.88*	0.04	7.87
MiBP	-2.55	-6.48	1.53	0.46	-0.92	1.85	-0.66	-1.90	0.61	3.09	-1.41	7.80
MnBP	-3.70	-8.00	0.79	-0.35	-1.87	1.19	-1.36*	-2.74	0.03	3.48	-1.53	8.75
MEHP	-5.49*	-9.32	-1.50	1.66*	0.26	3.08	-1.61*	-2.86	-0.35	7.57*	2.85	12.50
OH-MEHP	-6.31*	-10.48	-1.95	1.33*	-0.21	2.90	-2.17*	-3.53	-0.79	8.16*	2.94	13.65
oxo-MEHP	-7.87*	-12.14	-3.40	2.88*	1.26	4.53	-1.90*	-3.32	-0.45	11.67*	6.08	17.56
∑DEHP	-7.88*	-12.29	-3.24	2.05*	0.38	3.75	-2.22*	-3.69	-0.73	10.77*	5.03	16.83
cx-MEPP	-2.84	-8.06	2.67	0.05	-1.78	1.92	-0.95	-2.62	0.74	2.98	-3.02	9.35
MHNP	2.34	-2.09	6.97	0.48	-1.00	1.98	0.06	-1.29	1.43	-1.82	-6.43	3.02
MCOP	-3.17†	-6.69	0.47	4.34*	3.09	5.60	-1.20*	-2.31	-0.07	7.76*	3.55	12.14
∑DiNP	-0.30	-4.62	4.22	3.04*	1.54	4.57	-0.59	-1.93	0.77	3.35	-1.50	8.45
OH-MINCH	-1.64	-5.45	2.32	1.89*	0.56	3.24	-0.20	-1.40	1.02	3.59	-0.76	8.13
cx-MINCH	1.66	-2.30	5.79	0.90	-0.43	2.26	0.14	-1.08	1.37	-0.75	-4.95	3.64

Σ DINCH -0.89 -5.03 3.43 **1.60*** **0.16** **3.06** 0.06 -1.25 1.38 2.51 -2.14 7.38

Models adjusted by study of origin, age, sex, household education, and urinary creatinine concentration (only for phthalate metabolites). Estimates (%) are expressed as the percentage of difference in thyroid hormone level for one log-unit increase in the exposure biomarker. CI: Confidence interval; LL: Lower level; UL: Upper level. *p \leq 0.05; †p<0.10

Table S6. Associations of PFAS and phthalate/DINCH® **metabolites** with thyroid hormones in adolescent males (n=327).

Exposure biomarker	TSH			FT4			FT3			FT4/TSH		
	%	95% CI		%	95% CI		%	95% CI		%	95% CI	
		LL	UL		LL	UL		LL	UL		LL	UL
Serum PFAS												
PFOA	6.55†	-4.54	18.94	-6.81*	-10.64	-2.81	-2.14	-6.52	2.44	-12.54*	-22.54	-1.25
PFNA	2.46	-4.43	9.85	-0.64	-3.28	2.08	0.87	-2.00	3.84	-3.03	-10.24	4.76
PFOS	3.00	-4.27	10.81	-1.35	-4.10	1.49	1.28	-1.75	4.41	-4.21	-11.68	3.88
Σ PFAS	5.32	-3.95	15.50	-2.93	-6.33	0.59	0.96	-2.85	4.91	-7.84	-16.79	2.07
Urinary phthalate/DINCH® metabolites												
MEP	-2.54	-6.20	1.26	2.64*	1.16	4.14	-1.12	-2.68	0.47	5.32*	0.96	9.86
MBzP	-4.98*	-9.52	-0.21	0.00	-1.88	1.92	-0.95	-2.96	1.10	5.24†	-0.33	11.13
MiBP	-3.68	-9.01	1.97	0.15	-2.03	2.38	-1.04	-3.36	1.34	3.97	-2.40	10.76
MnBP	-2.70	-8.33	3.28	1.24	-1.06	3.59	-1.71	-4.11	0.75	4.05	-2.61	11.15
MEHP	-7.94*	-12.98	-2.59	2.26*	0.05	4.52	-0.99	-3.31	1.39	11.08*	4.37	18.22
OH-MEHP	-13.47*	-19.24	-7.28	1.66	-1.07	4.47	-2.01	-4.84	0.91	17.48*	8.82	26.83
oxo-MEHP	-12.97*	-18.57	-6.98	2.51	-0.13	5.23	-0.89	-3.66	1.96	17.79*	9.43	26.78
Σ DEHP	-14.42*	-20.28	-8.13	2.14	-0.68	5.04	-1.49	-4.43	1.54	19.35*	10.33	29.12
cx-MEPP	-4.89	-11.33	2.01	-0.39	-3.05	2.34	-0.59	-3.46	2.35	4.74	-3.10	13.21
MHNP	-0.43	-5.85	5.29	0.91	-1.24	3.10	0.51	-1.80	2.87	1.35	-4.75	7.84
MCOP	-4.37†	-8.74	0.21	3.71*	1.90	5.56	0.19	-1.75	2.17	8.45*	3.02	14.18
Σ DINP	-2.68	-7.91	2.85	3.03*	0.88	5.22	0.49	-1.80	2.83	5.87†	-0.41	12.54
OH-MINCH	1.83	-3.36	7.30	1.26	-0.76	3.32	0.53	-1.63	2.74	-0.56	-6.17	5.39
cx-MINCH	3.38	-1.50	8.50	0.04	-1.81	1.93	0.61	-1.39	2.66	-3.23	-8.29	2.11

Σ DINCH 2.72 -2.76 8.51 1.01 -1.10 3.16 0.73 -1.54 3.05 -1.67 -7.48 4.51

Model adjusted by study or origin, age, zBMI, household education, and urinary creatinine concentration (only for phthalates). Estimates (%) are expressed as the percentage of change in thyroid hormone level for one log-unit increase in the exposure biomarker. CI: Confidence interval; LL: Lower limit; UL: Upper limit. *p \leq 0.05; †p<0.10

Table S7. Associations of PFAS and phthalate/DINCH® metabolites with thyroid hormones in adolescent females (n=406).

Exposure biomarker	TSH			FT4			FT3			FT4/TSH		
	%	95% CI		%	95% CI		%	95% CI		%	95% CI	
		LL	UL		LL	UL		LL	UL		LL	UL
Serum PFAS												
PFOA	1.34	-8.65	12.43	-10.21*	-12.87	-7.46	3.19*	0.75	5.68	-11.40*	-20.70	-1.00
PFNA	-0.63	-7.92	7.24	-0.33	-2.64	2.03	0.94	-0.83	2.74	0.30	-7.58	8.86
PFOS	2.62	-4.87	10.69	-3.20*	-5.41	-0.94	1.63†	-0.13	3.43	-5.67	-13.03	2.32
ΣPFAS	2.24	-6.83	12.20	-5.36*	-7.98	-2.67	2.30*	0.13	4.51	-7.44	-16.21	2.26
Urinary phthalate/DINCH® metabolites												
MEP	-1.73	-5.94	2.67	2.63*	1.29	3.99	-0.45	-1.46	0.56	4.44†	-0.34	9.46
MBzP	-1.92	-6.59	2.99	1.23	-0.26	2.76	-0.71	-1.82	0.42	3.21	-2.06	8.77
MiBP	-1.76	-7.29	4.10	0.63	-1.14	2.43	-0.46	-1.78	0.89	2.43	-3.75	9.01
MnBP	-3.89	-10.20	2.87	-1.73	-3.75	0.33	-1.13	-2.68	0.43	2.25	-4.96	10.00
MEHP	-4.47	-9.98	1.37	1.46	-0.37	3.32	-2.13*	-3.45	-0.78	6.21†	-0.34	13.20
OH-MEHP	-2.68	-8.42	3.42	1.30	-0.56	3.20	-2.18*	-3.54	-0.81	4.09	-2.48	11.11
oxo-MEHP	-4.65	-10.76	1.89	3.30*	1.24	5.40	-2.50*	-3.96	-1.00	8.33*	0.91	16.30
ΣDEHP	-4.13	-10.37	2.54	2.18*	0.11	4.30	-2.57*	-4.06	-1.06	6.59†	-0.83	14.57
cx-MEPP	-0.54	-8.58	8.21	0.55	-2.01	3.18	-1.25	-3.15	0.70	1.09	-7.66	10.68
MHNP	5.94†	-1.01	13.38	0.32	-1.75	2.44	-0.10	-1.66	1.49	-5.31	-11.97	1.87
MCOP	-1.55	-6.95	4.17	4.99*	3.25	6.75	-2.50*	-3.74	-1.24	6.64*	0.39	13.27
ΣDINP	2.85	-3.97	10.16	3.27*	1.15	5.44	-1.50	-3.04	0.07	0.41	-6.73	8.10
OH-MINCH	-4.86†	-10.27	0.87	2.65*	0.85	4.50	-0.66	-2.00	0.69	7.90*	1.35	14.88
cx-MINCH	-0.42	-6.60	6.17	2.22*	0.24	4.23	-0.13	-1.60	1.37	2.65	-4.18	9.97

ΣDINCH -4.68 -10.68 1.73 **2.47*** **0.45** **4.52** -0.37 -1.86 1.15 **7.49*** **0.25** **15.26**

Model adjusted by study or origin, age, zBMI, household education, and urinary creatinine concentration (only for phthalates).
 Estimates (%) are expressed as the percentage of change in thyroid hormone level for one log-unit increase in the exposure biomarker. CI: Confidence interval; LL: Lower limit; UL: Upper limit. *p≤0.05; †p<0.10

Table S8. Associations of PFHxS and the PFAS mixture with thyroid hormones in adolescents from FLEHS IV and PCB cohort (n=436)

	TSH			FT4			FT3			FT4/TSH		
	%	LL	UL	%	LL	UL	%	LL	UL	%	LL	UL
All (n=466)												
PFHxS	12.35*	2.03	23.72	1.06	-3.06	5.36	2.58	-1.50	6.83	-73.18	-97.42	178.96
PFAS mixture	5.10†	-0.42	10.92	0.41	-1.53	2.39	0.36	-1.45	2.21	-4.46	-10.02	1.45
Males (n=201)												
PFHxS	5.83	-3.99	16.67	-0.27	-4.21	3.83	1.44	-3.10	6.20	-5.77	-15.20	4.71
PFAS mixture	3.39	-4.26	11.65	1.24	-2.47	5.10	-1.38	-5.19	2.59	-2.08	-10.09	6.65
Females (n=265)												
PFHxS	7.69	-2.78	19.28	-1.53	-4.25	1.26	2.20*	0.00	4.44	-8.56	-17.86	1.79
PFAS mixture	5.39	-2.11	13.47	-0.44	-2.71	1.88	1.02	-1.02	3.09	-5.53	-12.75	2.28

Single-exposure model (PFHxS): Models adjusted by study of origin (FLEHS IV/PCB), age, sex, zBMI, and household education. Estimates (%) are expressed as the percentage of difference in thyroid hormone level for one log-unit increase in the exposure biomarker.

Mixture effect model (PFAS mixture components: PFNA, PFOS, PFOA, and PFHxS): Models adjusted by study of origin (FLEHS IV/PCB), age, sex, zBMI, and household education. Estimates (%) are expressed as percentage of difference in thyroid hormone level per each quartile increase in the mixture concentration.

CI: Confidence interval; LL: Lower level; UL: Upper level. *p≤0.05; †p<0.10

Table S9. Serum PFAS and urinary phthalate/DINCH® metabolites mixture effects on thyroid hormones excluding zBMI from the model (n=733).

	PFAS mixture			Phthalate/DINCH® metabolites mixture		
	%	LL	UL	%	LL	UL
TSH	1.36	-2.56	5.43	-4.72	-11.30	2.36
FT4	-2.59*	-3.99	-1.18	2.58*	0.43	4.79
FT3	0.71	-0.53	1.96	-2.12†	-4.31	0.12
FT4/TSH	-3.90†	-7.96	0.34	7.66†	-0.37	16.34

PFAS mixture components: PFNA, PFOS, PFOA; Phthalates/DINCH® metabolite mixture components: MEP, MBzP, MiBP, MnBP, MEHP, OH-MEHP, oxo-MEHP, cx-MEPP, MHNP, MCOP, OH-MINCH, cx-MINCH.

Models adjusted by study of origin, age, sex, household education, and urinary creatinine concentration (only for phthalate metabolites). Estimates (%) are expressed as percentage of difference in thyroid hormone level per each quartile increase in the mixture concentration. CI: Confidence interval; LL: Lower level; UL: Upper level. *p<0.05; †p<0.10

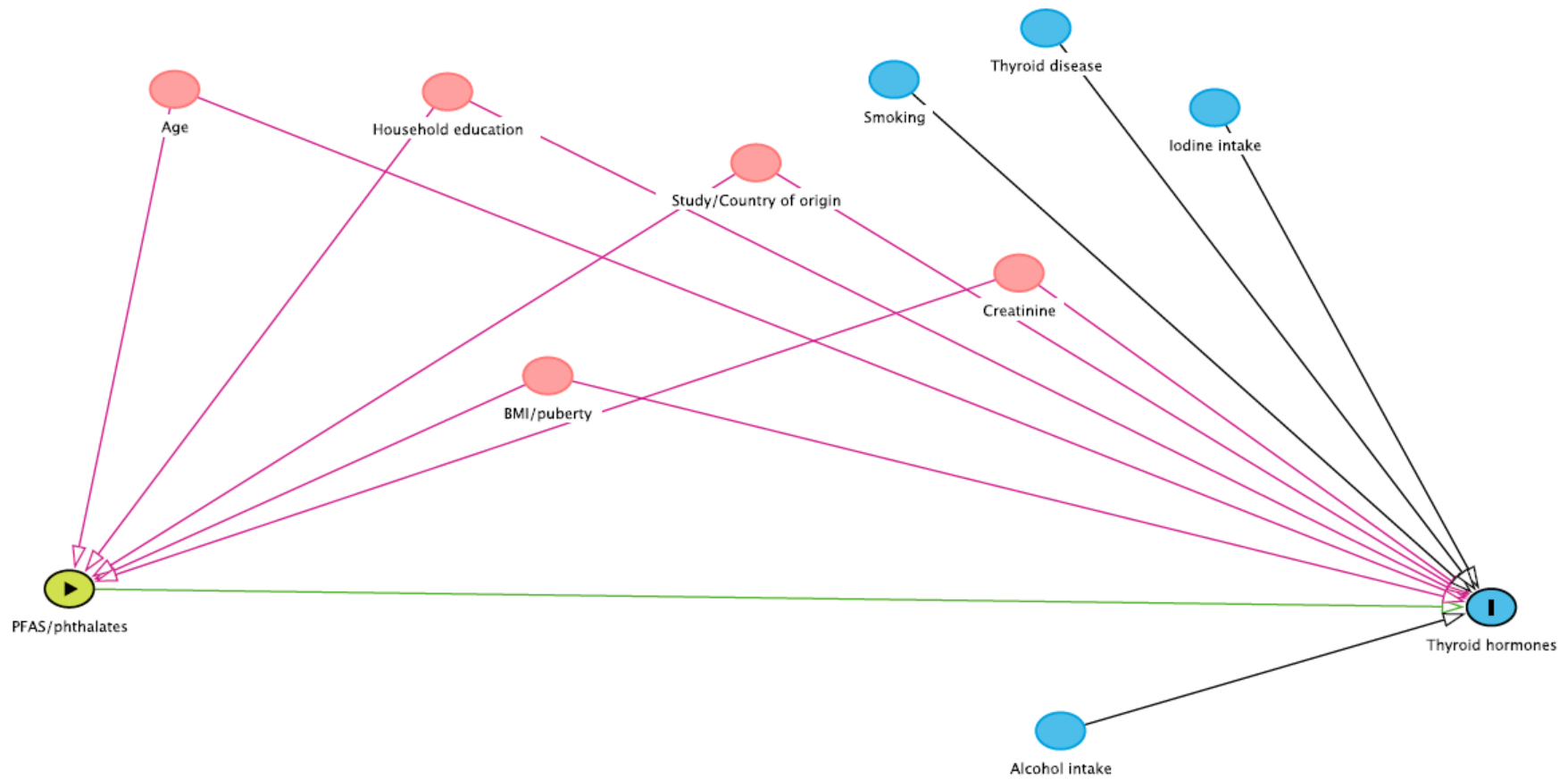


Fig S1. Directed acyclic graph (DAG) used to determine confounders for all models.

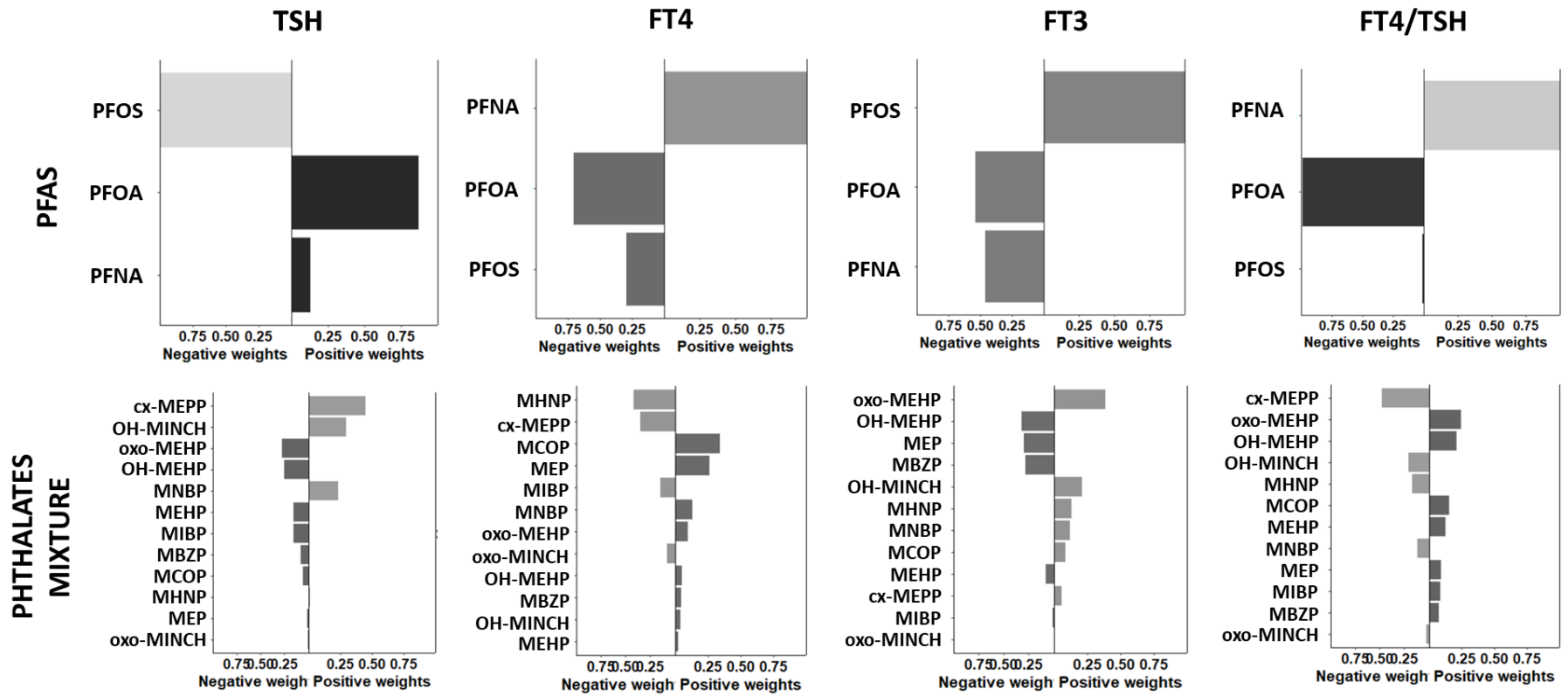


Figure S2. Mixture effect of PFAS/phthalate metabolites on thyroid hormones in males (n=327). PFAS mixture components: PFNA, PFOS, PFOA; Phthalates/DINCH® mixture components: MEP, MBzP, MiBP, MnBP, MEHP, OH-MEHP, oxo-MEHP, cx-MEPP, MHNP, MCOP, OH-MINCH, cx-MINCH.

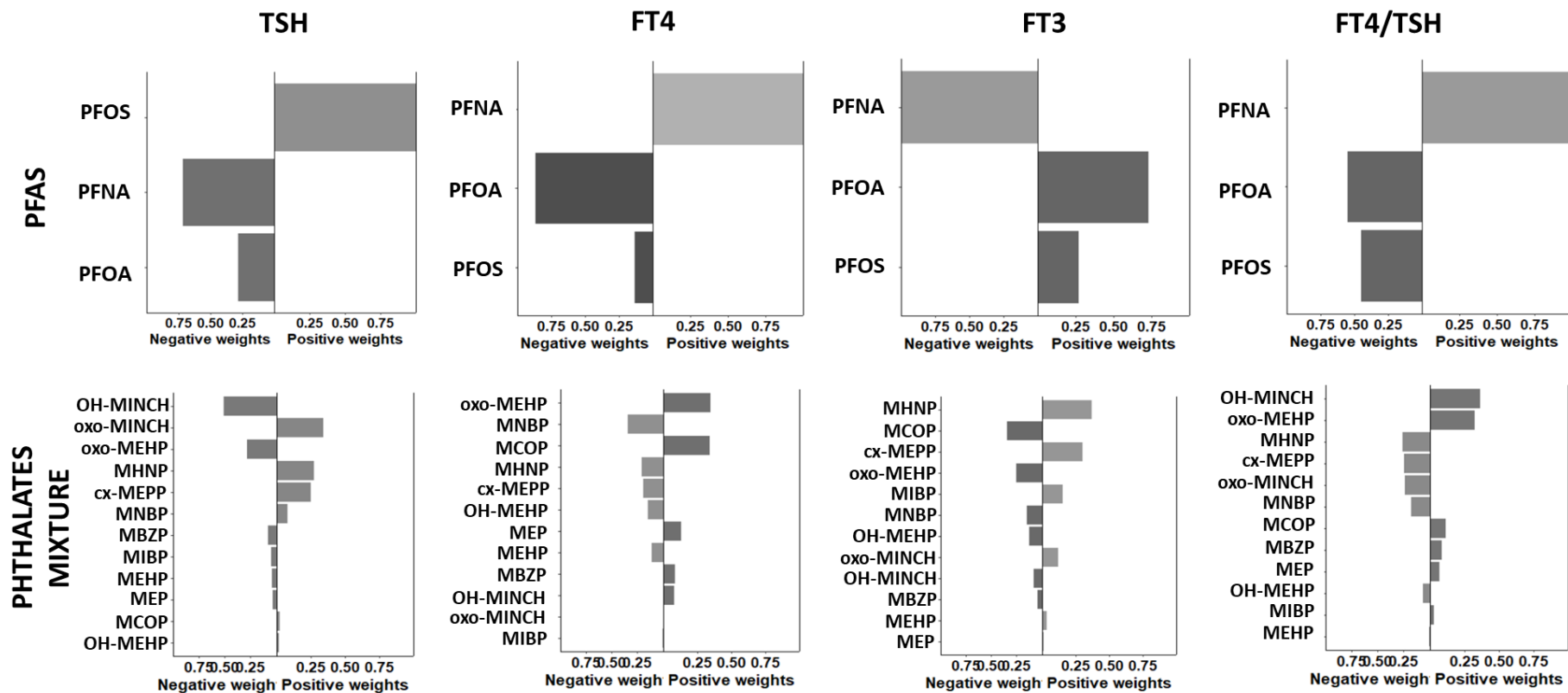


Figure S3. Mixture effect of PFAS/phthalate metabolites on thyroid hormones in females (n=406). PFAS mixture components: PFNA, PFOS, PFOA; Phthalates/DINCH® mixture components: MEP, MBzP, MiBP, MnBP, MEHP, OH-MEHP, oxo-MEHP, cx-MEPP, MHNP, MCOP, OH-MINCH, cx-MINCH

