Associations between phthalates exposure and health outcomes of newborns and pregnant women in Taiwan

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Physicochemical properties

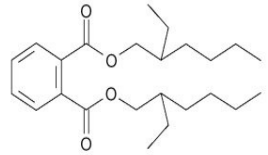


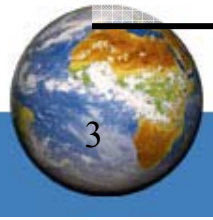
Table A Physicochemical properties of six phthalates from this study.

Compounds	Dimethyl phthalate	Diethyl phthalate	Di-*n*-butyl phthalate	Butyl benzyl phthalate	Di(2-ethylhexyl) phthalate	Di-*n*-octyl phthalate
Abbreviations	DMP	DEP	DBP	BBzP	DEHP	DOP
Chemical formula	$C_{10}H_{10}O_4$	$C_{12}H_{14}O_4$	$C_{16}H_{22}O_4$	$C_{19}H_{20}O_4$	$C_{24}H_{38}O_4$	$C_{24}H_{38}O_4$
Molecular weight	194.19	222.23	278.35	312.35	390.62	390.54
Melting point (˚C)	5.5	– 40.5	– 35	– 35	– 47	– 46
Boiling point (˚C)	282	295~302	340	370	386	390
Vapor pressure (25˚C, mmHg)	1 (100˚C)	1.65×10^{-3}	2.7×10^{-5}	6×10^{-7}	1×10^{-7}	1×10^{-7}
Solubility (mg L^{-1}, 25˚C)	4000	1000	13	0.7	0.3	3
Log Kow	1.6	2.5	4.5	4.9	7.5	8.1
Log Koc	1.9–2.6	1.8–2.7	3.1–4.2	3.2–4.2	4.9–6.0	6.3

Ref.: Sabljic et al., 1995; Staples et al., 1997; Api 2001; ATSDR, 2001, 2002; Kavlock et al., 2002.

Table B Application of different PAEs in commercial products.

PAEs	Applications
DEP	Perfume, cosmetics, shampoo, shower gel
DBP	PVC , food film, nail polish, perfume, latex adhesive, cellulose plastic, solvent of dye
DEHP	PVC flooring, carpet, wall covering, out layer of electric cord, artificial leather, waterproof cloth, car seat, raincoat, sealants, toy, wrapping, adhesive, pacifier, blood bag
BBP	PVC tubing, PVC flooring, adhesives
DMP	Cellulose plastic, hair preparations

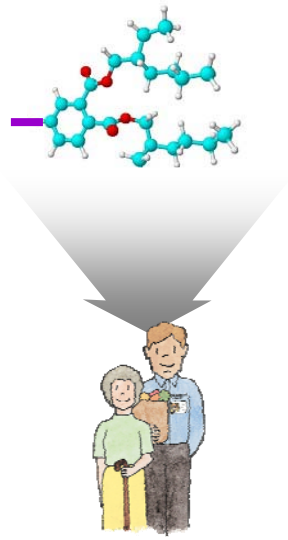


Application

- ***Phthalates*** **(PAEs) are used as plasticizers to plastics to make them soft and flexible, to cosmetics as a vehicle for fragrance, and to other products.**

- **Plastic Products**
- **Cosmetics**
- **Building Materials**
- **Cables and Wires**

- **Ω Food Contact Materials**
- **Ω Automotive Applications**
- **Ω Medical Devices**
- **Ω Toys**



Ref.: Api, 2001; ATSDR, 1995, 2001, 2002; Kavlock et al.,2002, 2006.

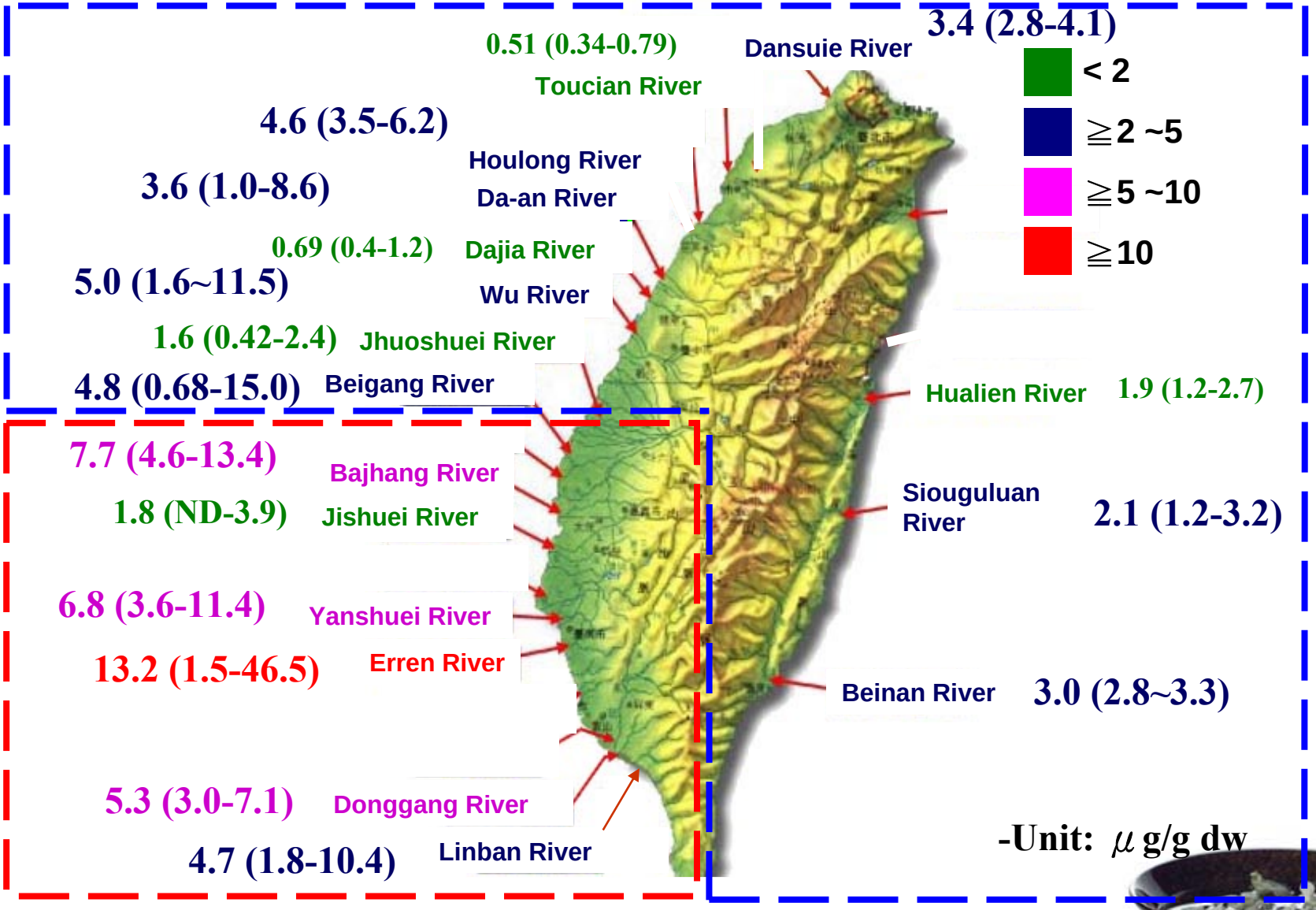


Figure A DEHP levels in sediment during low-flow season from 17 Taiwan's principal rivers.

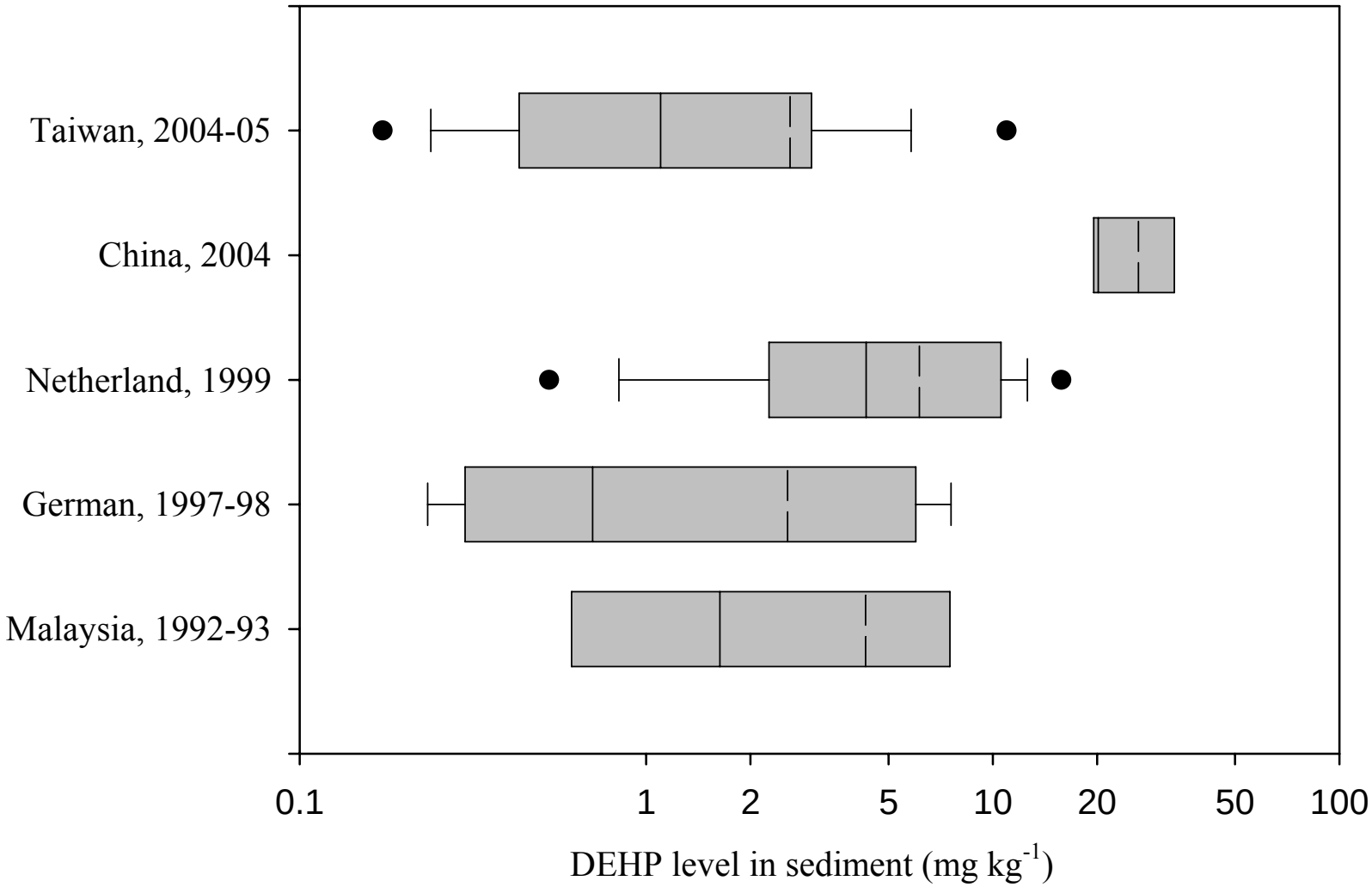


Figure B Comparison of DEHP concentration (mg kg^{-1}) in river sediment between this study and others (Mean value was showed by dash line).

Ref: Tan et al., 1995; Peijnenburg and Struijs, 2006; Fromme et al., 2002; Sha et al., 2007.

Table C Concentrations of phthalates (μg/g d. w.) in fish muscle (single and pooled samples).

Species	N	DEHP	DBP	DOP	BBzP	DEP	DMP
Single sample							
Oreochromis miloticus niloticus (吳郭魚)	12	33.6 (1.4-129.5)	0.23 (ND[1]-0.67)	0.08 (ND-0.4)	0.74 (ND-3.8)	0.11 (ND.-0.32)	0.03 (ND-0.17)
Liza subviridis (大鱗鯔; 豆仔)	7	61.8 (1.7-253.9)	0.10 (ND-0.31)	0.109 (ND-0.58)	6.37 (ND-26.8)	0.05 (ND-0.2)	0.27 (ND-1.8)
Pooled sample							
Oreochromis miloticus niloticus (吳郭魚)	3 (3, 12)	5.1 (2.6-7.6)	<0.05	ND	ND	0.26 (<0.05-0.49)	<0.05
Zacco platypus (平頷鱲;溪哥)	3 (3, 3, 5)	32.9 (1.5-92.7)	0.06 (<0.05-0.12)	ND	9.4 (ND-28.2)	0.06 (<0.05-0.97)	ND
Acrossocheilus paradoxus (石斑)	3 (3, 4, 8)	13.6 (1.2-33.1)	0.26 (<0.05-0.7)	ND	ND	0.03 (ND-0.07)	ND

[1]:ND: not detectable; ; the method detection limits (MDL) were as followed: 0.011 μg/g for DMP, 0.010 μg/g for DEP, 0.008 μg/g for DBP, 0.006 μg/g for BBP, 0.010 μg/g for DEHP, 0.008 μg/g for DOP

- **BSAF (Biota-Sediment Accumulation Factor) = (PAEs conc. in fish/ lipid content in fish)÷ (PAEs conc. in sediment/ organic carbon in sediment)**

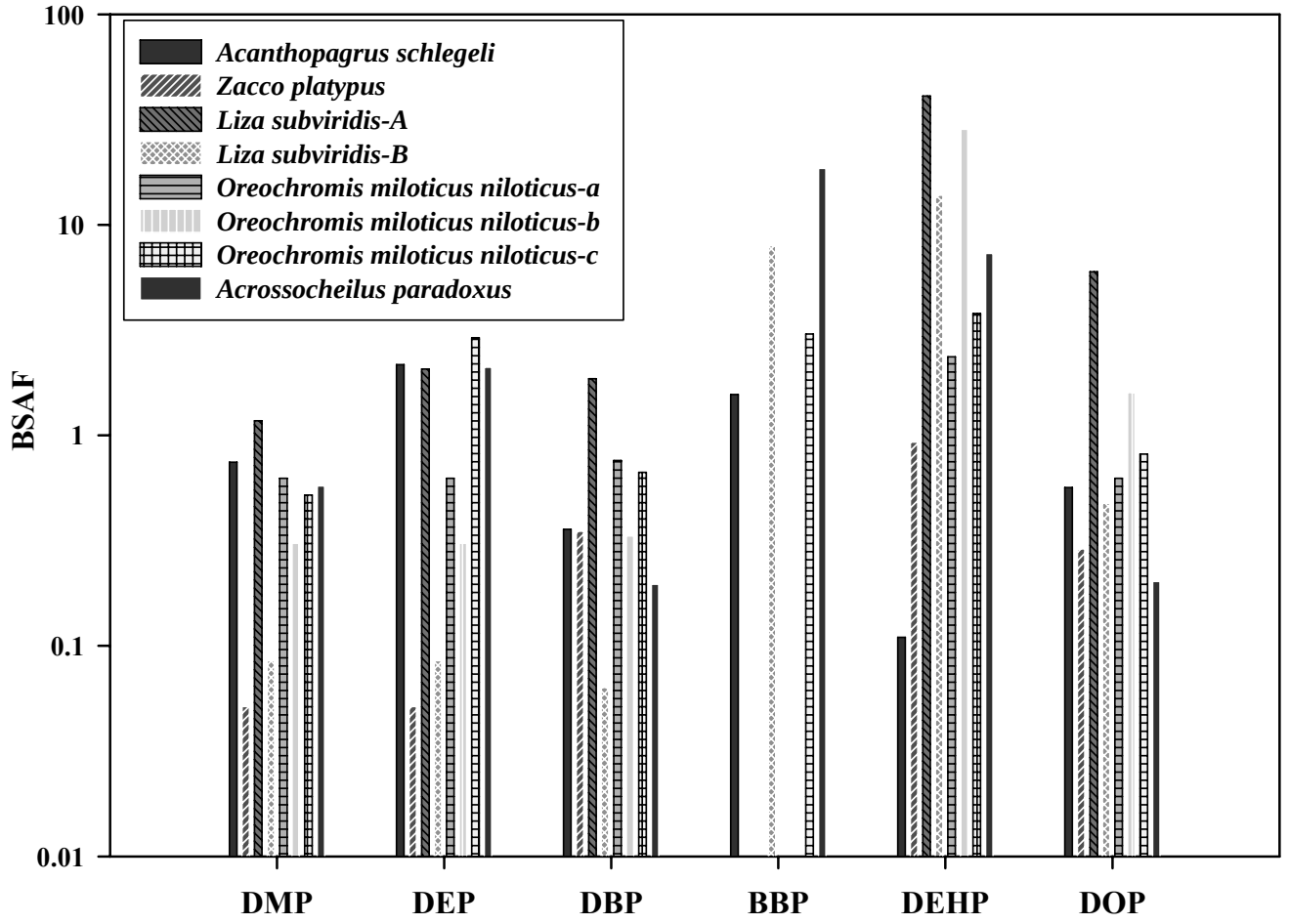


Figure C Bio-sediment accumulation factor (BSAF) of six phthalates in different fish species.

Table D Spearman correlation coefficients between DEHP and DBP level in sediment and several aquatic factors (N=64 for low and high-flow seasons; N=128 for all seasons).

Parameters	DEHP			DBP		
	Low-flow season	High-flow season	All season	Low-flow season	High-flow season	All season
DO	-0.254*	-0.583*	-0.286*			
BOD_5	0.520*	0.766*	0.567*			
SS	-0.101*	-0.205*	-0.171*			
NH_3-N	0.402*	0.775*	0.479*			
Temperature	0.302*	-	-			
pH	-0.274*	-0.348*	-0.263*			
COD	0.333*	0.712*	0.412*			
E.coli	0.712*	0.656*	0.541*			

✓**BOD, NH_3-N, COD, and E. coli indicate that the biological and chemical pollution of rivers.**

✓**Consistent results of their relationship to DEHP in sediment (r = 0.41~0.57) reveal that they may decrease the bio-degradation process of DEHP in the sediment by decreasing dissolved oxygen in the river.**

*: $p<0.05$.

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Occurrence of phthalates in sediment and biota: Relationship to aquatic factors and the biota-sediment accumulation factor

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RMSE=0.3978

the bio-degradation process

Figure D Multiple regression of DEHP level in sediment and water quality index in each river from 17 Taiwan principal rivers (R^2=0.453, p<0.001, N=125; Y_{DEHP} in sediment=4.478-0.317 X_{SS}+0.248 X $_{NH3-N}$-2.808 X $_{Water\ temperature}$ +0.258 X $_{COD}$)-All season. All the parameters were log-transferred.

Objectives

Part I: Association between Phthalate Exposure and Thyroid Hormone in Pregnant Women

- To determine the concentrations of urinary phthalate monoesters in pregnant women.
- To investigate the association between phthalate exposure and thyroid hormone during pregnancy.

Objectives

Part II: Association between Prenatal Exposure to Phthalates and the Health of Newborns

- To determine the concentrations of amniotic fluid phthalate monoesters in pregnant women.
- To evaluate the association between prenatal exposure of the fetus to phthalates in amniotic fluid and maternal urine and the health of newborns.

Objectives

Part III: Phthalates may alter thyroid hormones *in utero*: A follow-up study

- To follow-up our previously established participants in order to determine the correlation between phthalate exposure and thyroid hormones in pregnant women and newborns.

Framework

➢ To choose the susceptible population from the possibly highest phthalate exposure population in Taiwan

Pregnant Women

- Conc. of urinary m-PAEs
- Conc. of m-PAEs in serum
- Serum thyroid hormone (TSH, T_4, T_3, FT_4)
- Demographic characteristics (Age, gestation, smoke etc.)

Newborns

- Conc. of m-PAEs in amniotic fluid
- Conc. of m-PAEs in cord blood
- Cord blood thyroid hormone (TSH, T_4, T_3, FT_4)
- Health status of newborns (Anogenital distance, birth wt., etc.)

Association between Phthalate Exposure and Health Effects of Pregnant Women and Newborns in Taiwan

- Correlation between urinary m-PAEs and thyroid hormone in pregnant women.
- Correlation between amniotic fluid m-PAEs and the health of newborns.
- Correlation between cord blood m-PAEs and thyroid hormone in newborns.
- Correlation of m-PAEs among amniotic fluid, urine, serum and cord blood.

- **Provide an insight of potential health effects of phthalate exposure in pregnant women and newborns.**

Materials and Methods

Interview- 83 subjects

-alpha fetal protein (AFP) ↓

-free β-hCG ↑

-maternal age > 35 yrs

-undergo amniocentesis

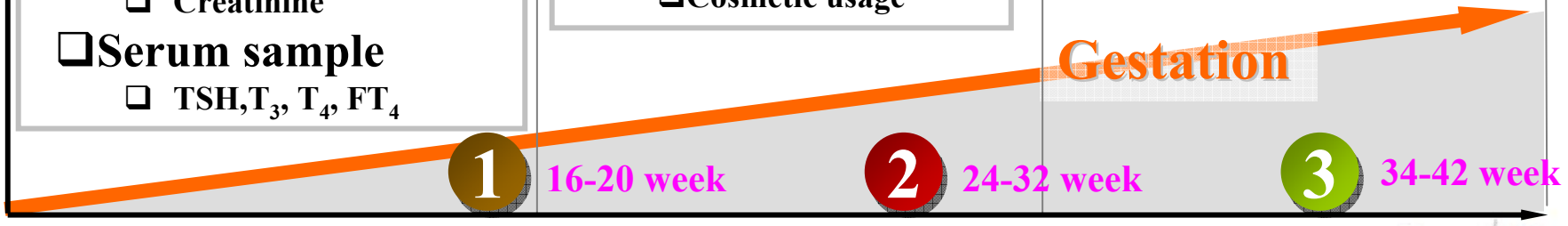
- ❑Informed consent
- ❑Amniotic fluid
  - ❑ Phthalate monoesters
- ❑ Urine sample
  - ❑ Phthalate monoesters
  - ❑ Creatinine
- ❑Serum sample
  - ❑ TSH, T_3, T_4, FT_4

Follow up-76 subjects

- ❑ Urine sample
  - ❑Phthalate monoesters
  - ❑Creatinine
- ❑ Serum sample
  - ❑TSH, T_3, T_4, FT_4
- ❑ Questionnaire
  - ❑Age, BMI, gestation
  - ❑Food consumption
  - ❑Cosmetic usage

Follow up-65 subjects

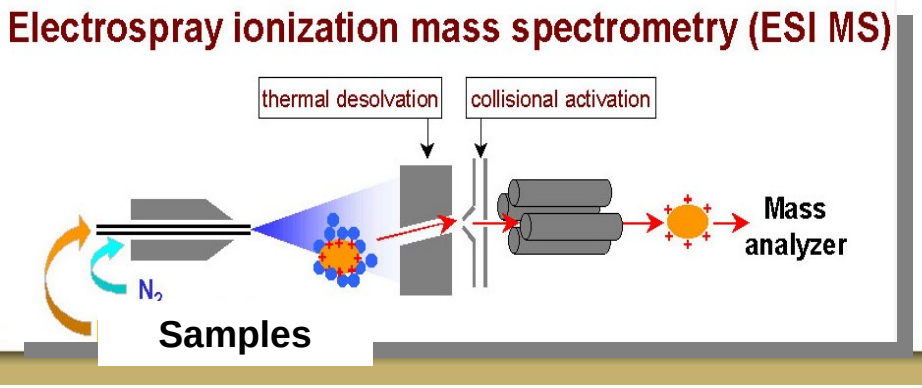
- ❑ Newborns' health
  - ❑Anogenital distance
  - ❑Gestational age
- ❑ Urine sample
  - ❑Phthalate monoesters
- ❑ Serum &Cord blood
  - ❑TSH, T_3, T_4, FT_4
  - ❑Phthalate monoesters



N Sampling

Measured Parameters

- **Phthalate monoesters**
  - monomethyl phthalate (MMP)
  - monoethyl phthalate (MEP)
  - monobutyl phthalate (MBP)
  - monobenzyl phthalate (MBzP)
  - mono-2-ethylhexyl phthalate (MEHP)
- **Sample**
  - Urine/ Amniotic fluid
  - Serum
- **Analytical method**
  - LC-MS/MS (Agilent 1100/API 3000)



1 mL sample thawed, sonicated

↓

250 μL ammonium acetate (1M, pH=6.5)
50 μL $^{13}C_4$-labeled standard (100 ng/mL)
5 μL β-glucuronidase (200 unit/mL)

↓

Incubation at 37°C for 90 min

↓

1 mL acidic buffer 0.14 M NaH_2PO_4

↓

Loaded to SPE cartridge (3 mL/60 mg)

↓

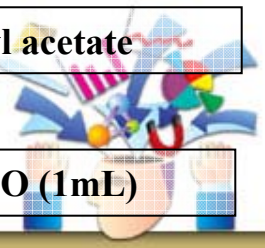
Washed with 2 mL formic acid (0.1M) and 1 mL H_2O

↓

Eluted with 2 mL ACN, 2 mL ethyl acetate

↓

The residue was resuspended in H_2O (1mL)



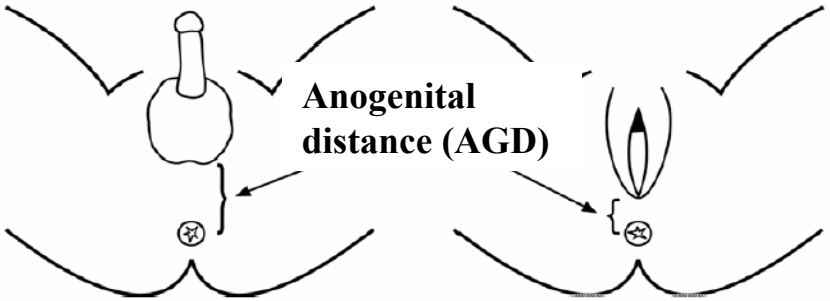
Measured Parameters

- **Thyroid hormones**
  - Thyroid stimulating hormone (TSH)
  - Thyroxin (T_4)
  - Triiodotyronine (T_3)
  - Free thyroxin (FT_4)
- **Questionnaires**
  - Age, BMI, gestation age, Smoking, Medical care etc.
  - Food consumption
  - Cosmetic usage
  - Personal care product usage
  - Building characteristics

- **Physical examination**
  - Anogenital distance (AGD)
  - Birth weight
  - Birth height
  - Gestational age
  - Anogenital index (AGI)

AGI-W= AGD/ birth weight.

AGI-L= AGD/ birth length.



-Salazar-Martinez et al., 2004.

Table I-1 Demographic characteristics of study participants (*n* = 76).

Characteristics	Mean ± SD
Age (years)	33.6 ± 3.3
Body mass index (BMI)	20.9 ± 2.5
Duration of gestation (weeks)	27.9 ± 2.3
Pregnancies and births	
Number of current pregnancy	1.9 ± 1.0
Number of current birth	1.5 ± 0.6
Smoking status (*n*/%)	
Active smoker	0/0
Passive smoker	14/18.4
Non-smoker	62/81.6
Alcohol drinker (*n*/%)	0/0
New decoration of living/ working place during previous 1 year (*n*/%) [a]	
Home	
Moving to new decorated house	7/9.2
Just decorated	4/5.3
Workplace	
Moving to new decorated workplace	2/2.6
Just decorated	6/7.9
Medical care during previous 3 months (*n*/%)	
Blood transfusion	3/3.9
Intravenous drip	3/3.9
Oxygen mask	1/1.3

[a] Moving to new decorated house or workplace means moved into a new building. Just decorated means the place you live or work have decorated recently such as painting or changing the floor.

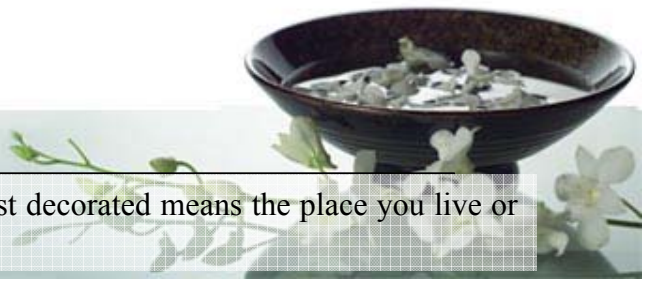


Table I-2 Levels of five urinary phthalates monoester in pregnant Taiwanese women and comparison with other studies (μ g/g creatinine).

Urinary phthalate monoesters[a]	Percentile								Median (range)		
	n	Min	5th	25th	50th	75th	95th	Max	US pregnant women[c]		US female population[d]
Creatine unadjusted (ng/ml)[b]											
MBP	76	13.2	21.6	40.6	81.8	131.0	368.0	580.0	–	> 2.5	30.0 (5.8-167)
MBzP	76	0.9	0.9	0.9	0.9	0.9	33.4	35.3	–	< 10 ↑	16.0 (2.4-103)
MEP	76	0.7	2.20	13.1	27.7	52.4	2346.0	5466.0	–	< 6	174.0 (28-2230)
MEHP	76	5.85	7.21	13.1	20.6	38.6	273.0	381.0	–	> 6.5	3.0 (ND-21.6)
MMP	76	0.7	0.7	0.7	4.3	14.7	87.8	237.2	–		–
Creatinine-adjusted (μg/g creatinine)											
MBP	76	57.8	88.9	127.0	195.0	339.0	839.0	1901.0	42.6 (21.3–105)	> 4.5	28.6 (10.6–131)
MBzP	76	0.5	0.8	2.0	3.7	6.0	24.0	69.9	12.1 (5.6–120)	< 4	14.7 (4.84–80.0)
MEP	76	5.0	8.3	27.0	68.0	205.0	4414.0	13 299.0	236 (26.7–5520)	< 3.5	157 (42.7–1920)
MEHP	76	12.2	15.8	31.4	60.8	121.0	885.0	1251.0	4.6 (1.8–449)	> 13	3.33 (ND–16.3)
MMP	76	0.4	0.9	3.7	10.8	34.9	263.0	363.0	–		–

[a]MBP, monobutyl phthalate; MBzP, monobenzyl phthalate; MEP, monoethyl phthalate; MEHP, mono-2-ethylhexyl phthalate and MMP, monomethyl phthalate. [b]Detection limit (LOD) of phthalate monoesters were: MBP, 5; MBzP, 1.8; MEP, 1.4; MEHP, 0.9; MMP, 1.4 ng/ml, respectively. Half of LOD was calculated as the detected value below the LOD. [c]New York pregnant women 18–35 years old ($n = 25$). [d]Data from NHANES 1999–2000 included children age 6 and older) ($n = 1326$, range: 10th–95th percentile).

Table I-3 Distribution of thyroid hormones in pregnant women.

Hormones [a]	Percentile					Reference range
	Min	5th	50th	95th	Max	
T_3 (ng/dL)	72.6	86.3	132.0	209.0	246.0	84.6-202.0
T_4 (μg/dL)	4.39	5.31	8.85	11.2	13.6	5.13-14.1
Free T_4 (ng/dL)	0.46	0.69	0.93	1.25	1.35	0.93-1.7
TSH (μ IU/mL)	0.22	0.31	1.06	3.4	5.19	0.27-4.2

[a]The analytic sensitivities of T_3, T_4, free T_4, and TSH were 19.5 ng/dL, 0.42 μg/dL, 0.023 ng/dL and 0.014 μlU/mL, respectively; the coefficient variations of T_3, T_4, free T_4, and TSH were 2.9%, 4.2%, 3.1%, and 3.0%, respectively.

Table I-4 Spearman correlation coefficients [a] between thyroid hormone, age, BMI, duration of gestation and urinary phthalate monoester levels (n = 75).

	TSH	T_3	T_4	FT_4	Age	BMI	Gestation
TSH	1						
T_3	0.145	1					
T_4	**0.291***	**0.709***	1				
FT_4	0.172	**0.299***	**0.761***	1			
Age	0.203	**–0.307***	**–0.218***	–0.008	1		
BMI	–0.044	0.309	0.137	–0.010	–0.106	1	
Gestation	–0.106	–0.018	0.038	0.057	–0.029	–0.131	1
MBP	0.079	–0.234	**–0.248***	**–0.368***	0.082	**0.081***	–0.098
MEP	–0.082	–0.019	–0.039	0.017	0.080	**0.095***	0.199
MEHP	-0.060	–0.067	–0.100	–0.090	0.166	–0.030	**0.030***
MBzP	–0.080	–0.084	0.034	–0.007	–0.064	–0.027	0.118
MMP	–0.078	–0.259	–0.089	0.007	–0.038	–0.006	–0.004

[a] Spearman correlation coefficients, *: $p < 0.05$.

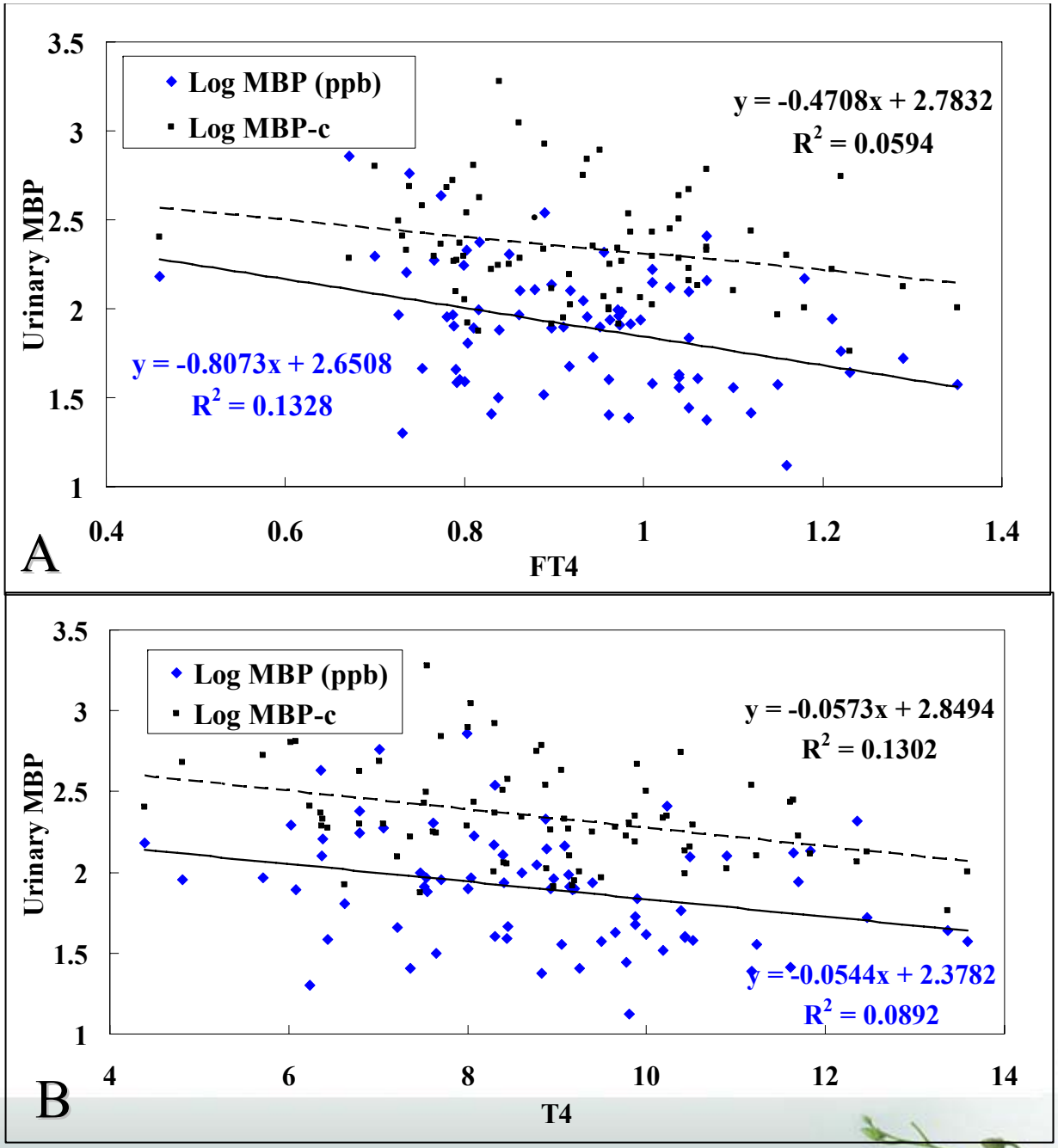


Figure I-1. Correlation between (A) free thyroxine (FT_4), (B) thyroxine (T_4) and log MBP (ppb) and MBP-c level in pregnant women at the 2nd trimester.

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Associations between urinary phthalate monoesters and thyroid hormones in pregnant women

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MBP	**-0.110**	**<0.001**	**-0.112**	**0.003**
MEP	**0.026**	**0.124**	**0.013**	**0.398**
MEHP	**-0.015**	**0.474**	**-0.007**	**0.814**
MBzP	**0.022**	**0.232**	**0.032**	**0.224**
MMP	**0.016**	**0.165**	**0.015**	**0.347**

[a] One outlier was excluded because of hypothyroidism (n=75).

→Multiple regression: FT_4： β = -0.110, R^2=0.24, p<0.05 T_4： β=-0.112; R^2=0.187, p<0.05

Table II-1 Physical examination of male and female newborns (*n* = 65).

Health status	Males (n=33)	Females (n=32)	P-value[3]
Birth weight (g)	3171 (1678-4260)	3002 (2000-3935)	0.156
Birth length (cm)	49.6 (42.0-56.0)	48.3 (42.5-53.5)	0.023*
Gestational age (weeks)	39.0 (35.3-41.7)	38.4 (33.6-40.3)	0.020*
Ano-genital distance (AGD) (mm)[1]	23 (10-36)	16 (7-23)	<0.001*
AGI-W(mm/kg)[2]	7.16 (3.18-13.09)	5.37 (2.30-8.96)	0.001*
AGI-L ($\times 10^3$)[2]	4.55 (2.17 -7.35)	3.29 (1.41 -5.07)	<0.001*

[1] The AGD of two girls and boys, respectively, were not available.

[2] AGI=ano-genital index; AGI-W= AGD/birth weight; AGI-L=AGD/ birth length.

[3] *: $p<0.05$

Table II-2 Concentrations of five phthalate monoesters in amniotic fluid from female and male infants (ng/mL, n=64)[1].

Female infants	Min	10th	50th	90th	Max
MBP	39.3	45.6	85.5	134.6	192.0
MEHP	ND[2]	5.0	24.0	91.1	148.0
MEP	ND	ND	ND	3.9	6.5
MBzP	ND	ND	ND	84.1	233.0
MMP	ND	ND	ND	ND	2.92
Male infants	**Min**	**10th**	**50th**	**90th**	**Max**
MBP	28.4	44.3	81.3	127.8	145.0
MEHP	ND[2]	2.6	22.1	100.6	110.0
MEP	ND	ND	ND	4.4	7.7
MBzP	ND	ND	ND	87.9	104.0
MMP	ND	ND	ND	ND	ND

[1]One amniotic fluid sample was failed during analysis.

[2]ND=not detected, detection limit of MMP, MEP, MBP, MBzP and MEHP were as follow: 1.4, 1.0, 1.4, 1.4, 0.9 ng/mL

Table II-3 Concentrations of maternal urinary phthalate monoesters in the 1st trimester (ng/mL, n=64)[1].

	Min	10th	50th	90th	Max
Female fetus group					
MBP	8.9	26.9	78.0	30.9	541.0
MEHP	ND	11.8	24.6	68.6	1140.0
MEP	ND	3.0	22.8	288.2	415.0
MBzP	ND	ND	3.0	16.8	845.0
MMP	ND	ND	7.1	19.9	48.4
Male fetus group					
MBP	19.4	28.1	79.6	232.6	524.0
MEHP	6.0	11.9	26.3	120.3	151.0
MEP	3.6	4.9	19.1	324.4	1420.0
MBzP	ND	ND	2.5	13.9	98.5
MMP	ND	ND	6.8	20.3	26.9

ND = not detectable. The detection limits for MMP, MEP, MBP, MBzP, and MEHP were 1.4, 1.0, 1.4, 1.4, and 0.9 ng/mL, respectively.

[a] One amniotic fluid sample failed during analysis.

■ **It is indicated that current levels of exposure to phthalate in pregnant women may only partly represent *in utero* fetal exposure**

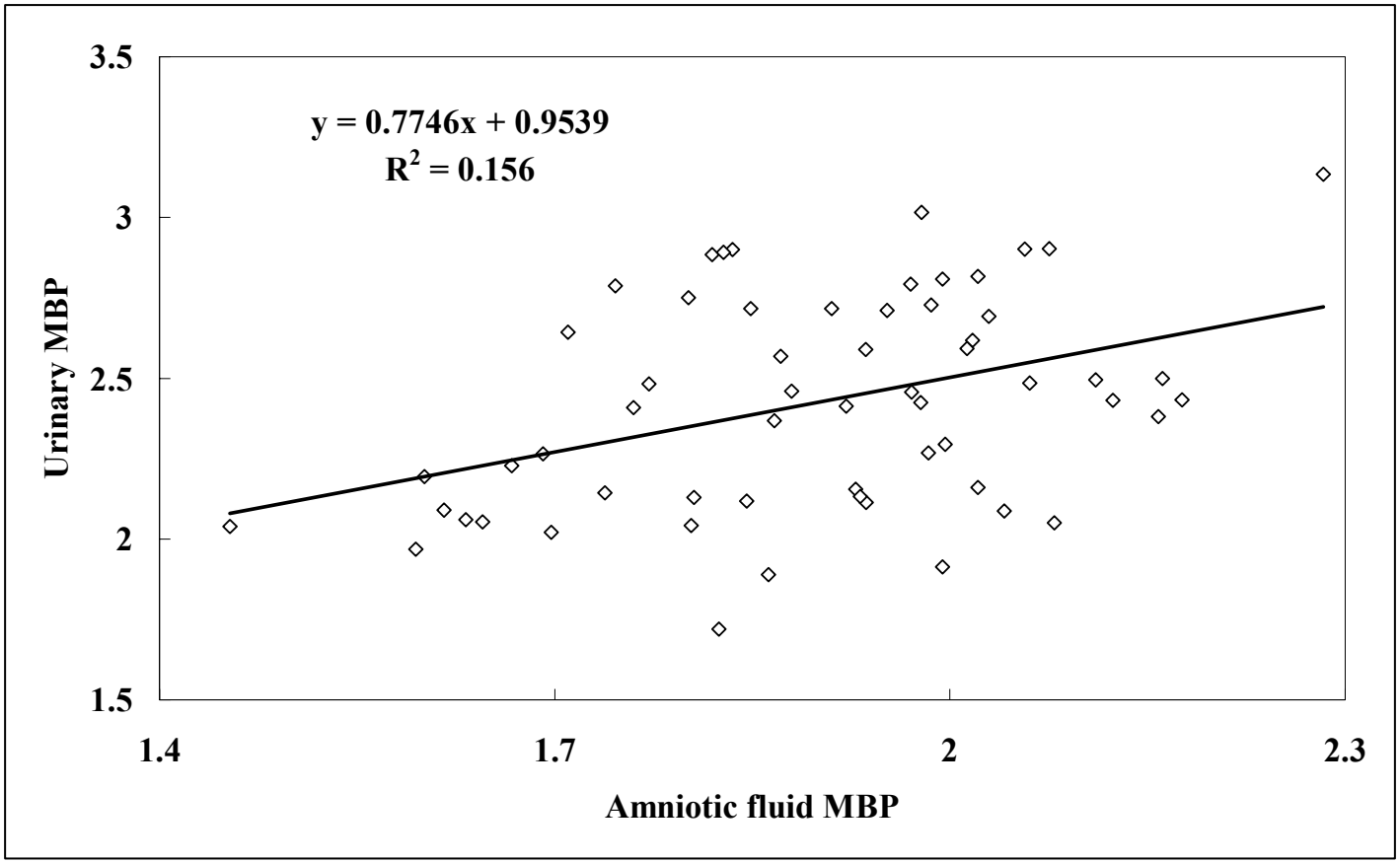

Figure II-1 Distribution plot of MBP levels in maternal urine and amniotic fluid (n=59). All the data were log-transformed.

Table II-4 Distribution of health status in females categorized by median levels of phthalate monoester in amniotic fluid (-AF) (*n* = 31).

Health status in female infants	MBP-AF[2]			MEHP-AF[3]		
	Low[4] (n=15)	High[4] (n=16)	P-value[1]	Low[4] (n=15)	High[4] (n=16)	P-value[1]
Birth weight (g)	2810±439	3172±398	0.031*	2918±448	3061±467	0.527
Birth length (cm)	47.3±2.3	49.2±1.7	0.018*	48.1±2.1	48.6±2.4	0.138
Gestational age (weeks)	38.1±1.5	38.7±1.0	0.205	38.4±0.9	38.4±1.6	0.591
AGD (mm)	17.6±4.0	13.9±3.8	0.024*	17.0±4.1	14.2±4.1	0.109
AGI-W (mm/kg)	6.2±1.6	4.5±1.5	0.007*	6.0±1.4	4.7±1.8	0.016*
AGI-L ($\times 10^3$)	3.7±0.9	2.8±0.8	0.008*	3.6±0.8	2.9±0.9	0.015*

[1]Wilcoxon Rank Sum Test, *: $p < 0.05$;
[2]AGD data were lacking for one female newborn in each group.
[3]AGD data were lacking for two female newborns in the high MEHP-AF group.
[4]Median levels of amniotic fluid MBP in low and high group were 67 and 104 ng/mL, respectively; those of MEHP were 9.5 and 38.8 ng/mL, respectively.

Table II-5. Distribution of health status in boys category by median levels of phthalate monoester in amniotic fluid. (*n* = 33).

Health status in male infants	MBP			MEHP		
	Low (n=16)	High (n=17)	P-value	Low (n=16)	High (n=17)	P-value
Birth weight (g)	3146±481	3194±469	0.640	3213±440	3131±515	0.815
Birth length (cm)	49.2±2.5	50.0±2.8	0.705	49.4±2.4	49.7±3.0	0.971
Gestational age (weeks)	39.1±1.0	38.9±1.4	0.396	39.1±0.9	38.9±1.5	0.787
AGD (mm)	21.2±6.5	24.1±5.7	0.234	23.3±6.8	22.2±5.6	0.606
AGI-W (mm/kg)	6.6±2.1	7.7±2.2	0.206	7.4±2.6	6.9±1.7	0.635
AGI-L ($\times 10^3$)	4.3±1.2	4.8±1.2	0.304	4.7±1.4	4.4±1.0	0.649

[1] One male infant of each group in MBP-AF and MEHP-AF, respectively, were lack of AGD.

Table II-6 Spearman correlation coefficients[1] between birth weight, birth length, gestational age, AGD, AGI-W, AGI-L, and phthalate monoester levels in amniotic fluid in female infants (n = 29)[2].

	MMP	MEP	MBP	MBzP	MEHP	BW	BL	GA
BW	0.15	0.07	0.16	–0.36	–0.17	1		
BL	0.29#	0.01	0.20	–0.35	–0.10	**0.83***	1	
GA	0.27	0.24	0.18	–0.22	–0.06	**0.43***	**0.48***	1
AGD	–0.42	–0.10	–0.31#	0.09	–0.13	–0.16	–0.22	**–0.46***
AGI-W	–0.41	–0.10	**–0.32***	0.32	0.03	**–0.58***	**–0.54***	**–0.54***
AGI-L	–0.44	–0.10	**–0.33***	0.15	–0.10	–0.27	–0.27#	**–0.50***

[1] *:p< 0.05; #: p<0.06.

[2] BW=birth weight, BL=birth length, GA=gestational age, AGD=ano-genital distance, AGI-W=AGD/BW, AGI-L=AGD/ BL

Negative associations among amniotic fluid MBP and AGI

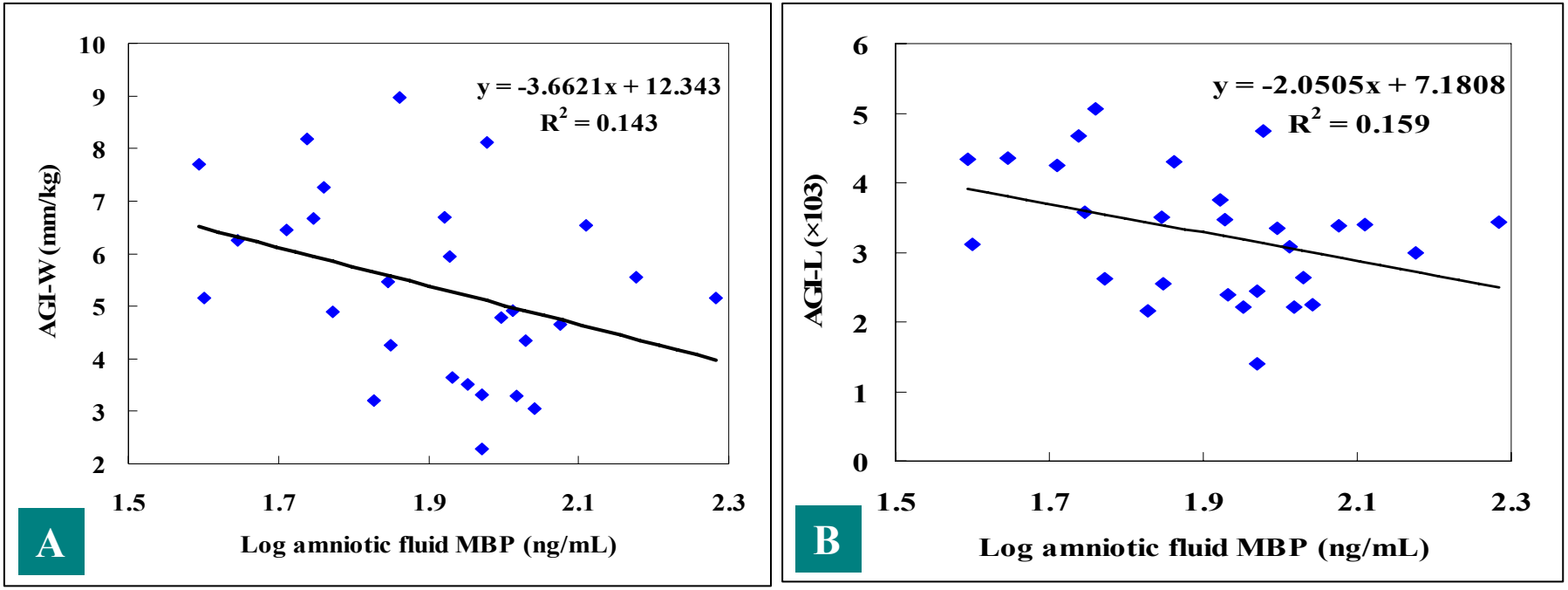

Figure II-2 (A) Linear regressions between MBP in amniotic fluid and AGI-W in females at birth (n=29, R^2=0.14, p=0.043); (B) Linear regressions between MBP in amniotic fluid and AGI-L in females at birth (n=29, R^2=0.16, p=0.032).

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Association between prenatal exposure to phthalates and the health of newborns ☆

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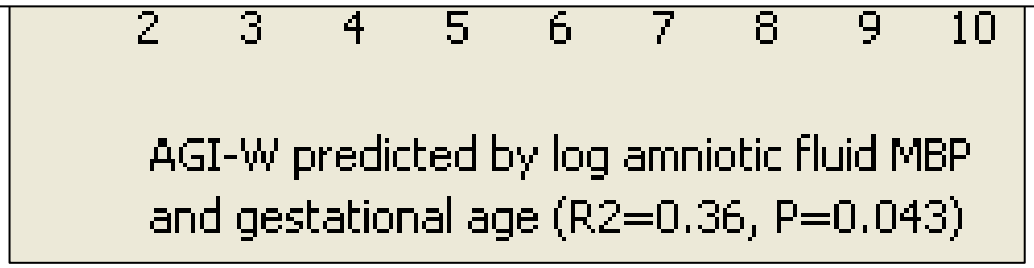
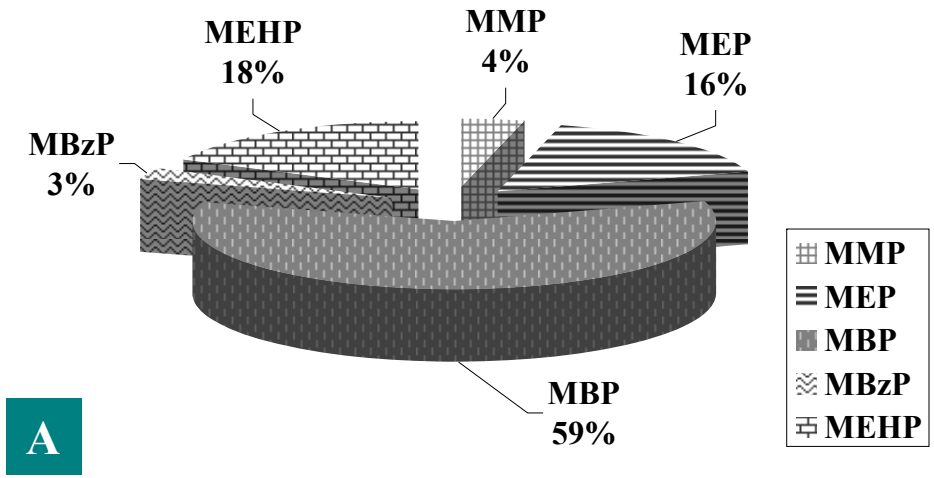


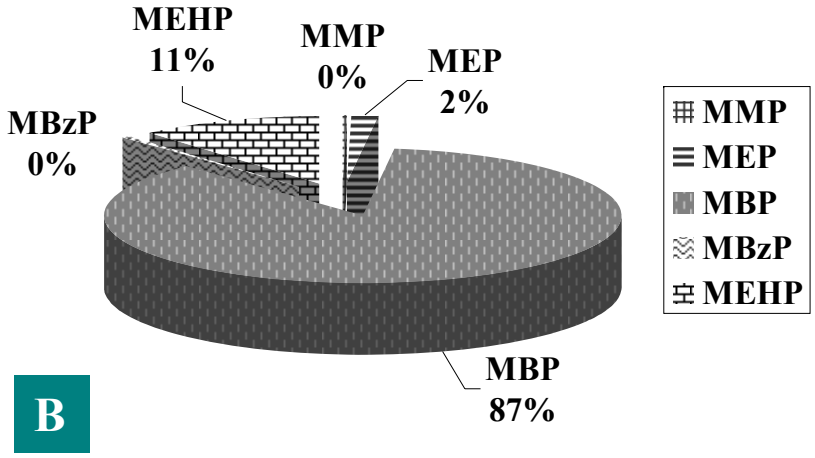
Figure III-3 Multiple regression of AGI, MBP in amniotic fluid and gestational age adjustment of gestational age and other phthalate monoesters (amniotic fluid MBP: β = –2.73, p = 0.041; gestational age: β = –0.899, p = 0.006) .

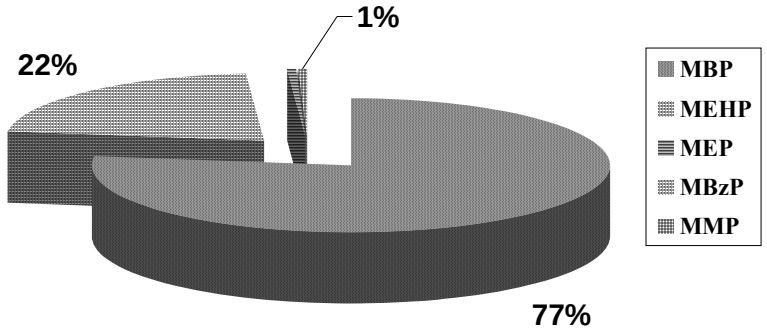
Table III-1 Concentrations of phthalate monoesters in urine, serum and cord blood at delivery (ng/mL, n=61).

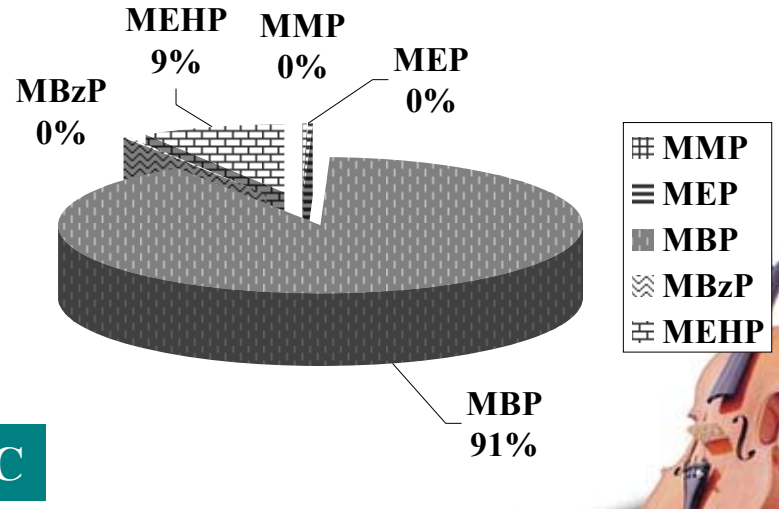
Mono-PAEs	Urine		Serum		Cord blood	
	Median (Range)	10-90th	Median (Range)	10-90th	Median (Range)	10-90th
MBP[a]	114 (25.4-1830)	36.9-550.6	158.0 (59.6-1080)	64.9-413.0	256.0 (65.2-815)	97.4-604.8
MEHP	40.2 (3.6-958)	8.4-152.0	21.0 (9.2-99.2)	11.7-37.1	24.7 (11.0-665.0)	14.2-65.9
MEP	36.4 (ND[b]-1980)	4.6-236.8	2.8 (ND-26.5)	ND-6.3	ND (ND-9.3)	ND-3.4
MBzP	5.7 (ND-218.0)	1.9-49.2	ND (ND-10.1)	ND-2.8	ND (ND-26.8)	ND-3.6
MMP	8.3 (ND-169)	1.7-38.0	ND (ND-3.7)	ND-2.4	ND (ND-13.3)	ND-ND

[a]MBP = monobutyl phthalate; MBzP = monobenzyl phthalate; MEP = monoethyl phthalate; MEHP = mono-2-ethylhexyl phthalate; MMP = monomethyl phthalate. [b]Detection limit (LOD) of phthalate monoesters were: MBP, 1.4; MBzP, 1.4; MEP, 1.0; MEHP, 0.9; MMP, 1.4 ng/mL, respectively. Half of LOD was calculated as the detected value below the LOD.









- **Figure III-1 Contribution profiles of phthalate monoesters in maternal urine (A), serum (B), and cord blood (C) (n=60).**

Table III-2 Concentrations of thyroid hormones[1] in maternal serum during pregnancy and cord blood at delivery (n=61).

Serum Thyroid Hormones	First-trimester		Second-trimester		Third-trimester		Cord blood	
	Median	Range	Median	Range	Median	Range	Median	Range
TSH (μIU/mL)	1.13	0.01-4.38	1.06	0.23-5.19	2.08	0.38-6.07	7.05	1.63-289
T_3 (ng/dL)	123	70.2-230	134	72.6-246	140	82.4-277	56.3	35.1-84.6
T_4 (μg/dL)	8.26	3.37-12.1	9.01	4.39-13.4	9.62	3.60-16.9	7.66	3.65-11.7
FT_4 (ng/dL)	0.95	0.62-1.29	0.93	0.46-1.29	0.99	0.33-1.31	1.13	0.49-1.45

[1]Reference values for thyroid hormones in Taiwan: TSH: 0.27-4.2; T_3: 84.6-202.0; T_4: 5.13-14.1; FT_4: 0.93-1.7. The analytic sensitivities of T_3, T_4, free T_4, and TSH were 19.5 ng/dL, 0.42 μg/dL, 0.023 ng/dL and 0.014 μIU/mL, respectively. The coefficient variations of T_3, T_4, free T_4, and TSH were 3.0%, 4.1%, 3.2%, and 2.9%, respectively.

Table III-3 Correlation [a] coefficients between fetal thyroid hormones and phthalate monoesters in cord blood samples (n=60)[a]

	Maternal age	Gestational age	MBP-C[b]	MEHP-C[b]	MEP-C[b]	MBzP-C[b]	MMP-C[b]
Maternal age	1	0.208	-0.073	0.007	0.045	-0.135#	0.112
Gestational age	0.208	1	0.028	0.016	0.150	0.229	0.063
TSH	0.176	-0.396	-0.277#	0.109	0.108	-0.084	-0.043
T_4	0.078	0.0343	-0.157#	-0.045	-0.026	0.038	-0.136
T_3	-0.133	-0.069	0.079	0.211	0.183	-0.053	-0.003
FT_4	-0.112	0.103	0.162	-0.062	0.049	0.003	-0.109
TSH×T_4	0.183	-0.389	-0.230*	0.096	0.124	-0.101	-0.072
TSH×FT_4	0.169	-0.390	-0.214#	0.095	0.128	-0.109	-0.069
FT_4/T_3	0.071	0.084	-0.027	-0.213	-0.104	0.006	-0.096
FT_4/T_4	-0.200	0.061	0.346**	0.017	0.066	-0.049	0.102
T_3/T_4	-0.072	-0.162	0.133#	0.147	0.122	-0.057	0.091
T_4/TSH	0.019	0.287	0.089	0.000	-0.131	0.108	-0.002

[a] Spearman correlation: **p<0.01, *p<0.05, #p<0.1. [b]-C: Phthalate monoesters in cord blood sample.

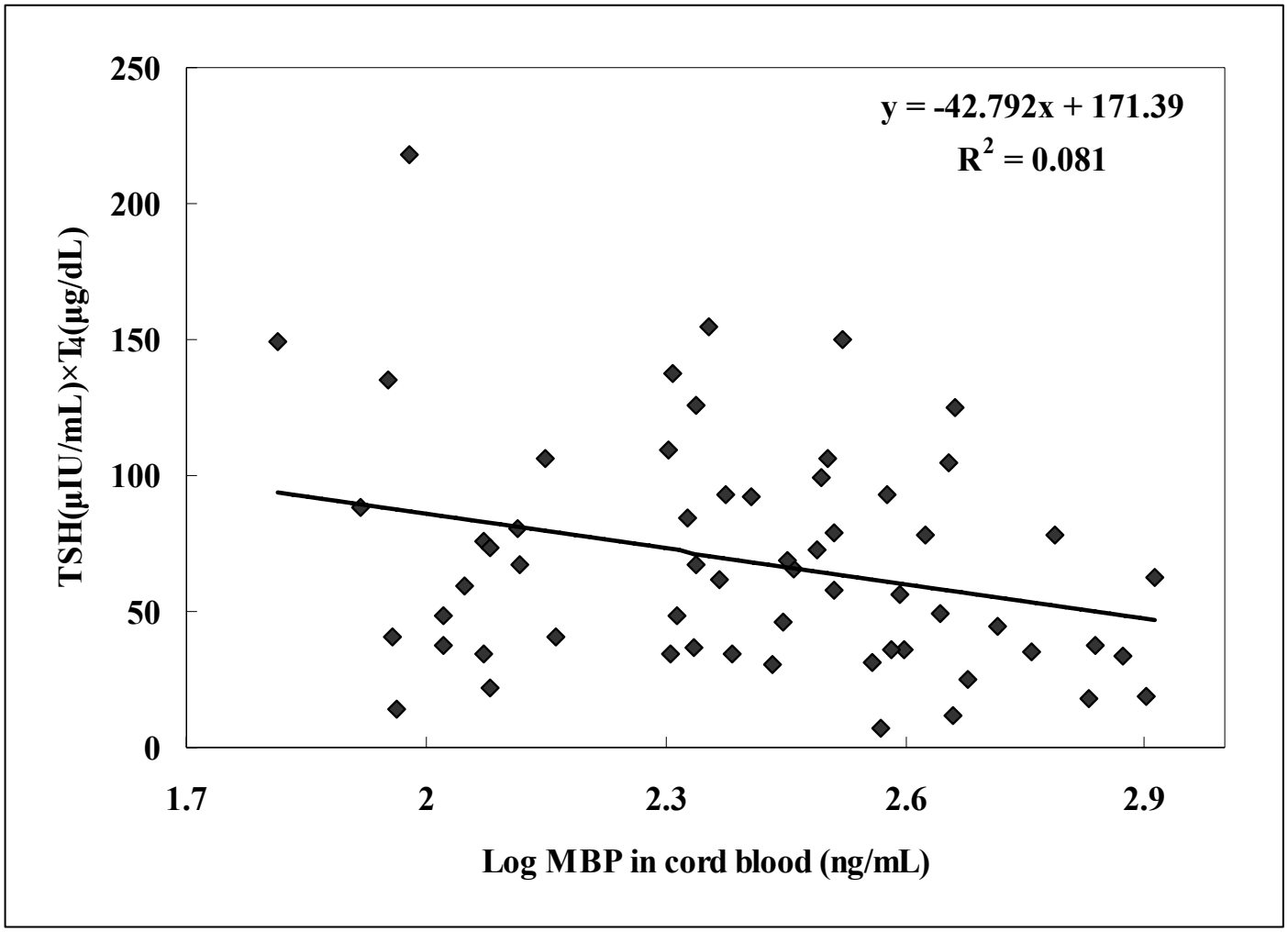


Figure III-2 Linear correlation between log MBP levels and TSH×T_4 in cord blood samples (R^2=0.081, p=0.028, n=60).

Table III-4 Multivariate regression between TSH and T_4 and their corresponding phthalate monoesters in cord blood (n=60)[a]

Variables	TSH (μIU/mL)		TSHxT4		TSHxFT4		FT4/T4		T_4 (μg/dL)	
	Estimate[a]	Prob > |t|	Estimate[a]	Prob > |t|	Estimate[a]	Prob > |t|	Estimate[a]	Prob > |t|	Estimate[a]	Prob > |t|
Intercept	3.49	0.010	171.4	0.004	20.6	0.001	0.123	0.006	20.4	0.017
MBP	**-0.217**	**0.044***	**-42.8**	**0.028***	-4.49	0.075#	**0.036**	**0.004****	**-1.71**	**0.036***
MEHP	–	–	–	–	–	–	–	–	–	–
Maternal age	–	–	–	–	–	–	-0.002	0.087#	0.113	0.106
Gestational age	-0.045	0.117	–	–	–	–	–	–	-0.315	-0.104
R^2	0.087	0.044	0.092	0.028	0.054	0.075	0.171	0.005	0.115	0.046

—, Excluded from model.

[a]Values are beta coefficients except for R^2; # $p < 0.10$. * $p < 0.05$. ** $p < 0.01$.

Possible Hypotheses

- **First, we postulated phthalate exposure may affect fetal thyroid function indirectly via decreased thyroid hormones from their mother.**
  - **Animal studies have shown that *in utero* exposure to thyroid hormone affects the regulation of the hypothalamus-pituitary-thyroid axis in newborns (Alonso et al., 2007; Piosik et al., 1997).**
  - **Our previous study showed decreased FT_4 and T_4 levels with increasing urinary MBP levels in pregnant women during the second trimester (Huang et al., 2007).**
- **Second, phthalate monoesters penetrate the human placenta and may have direct effects on fetal thyroid function.**
  - **Some toxicological studies have shown that certain phthalates, such as DBP and DEHP, are possible thyroxine antagonists that affect the transportation, secretion, and action of thyroid hormones in adult animals (Hinton et al., 1986; Poon et al., 1997; Sugiyama et al., 2005).**
  - **Although little information is available about phthalate exposure *in utero* and its effects on fetal thyroid function, some studies (Huang et al., 2007; Meeker et al., 2007) have shown possible negative effects in humans.**

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  - All stuffs


Thanks for your attention!
Comments!