Levels of environmental pollutants biomarkers in French parturient women

A pilot study of the Elfe Longitudinal mother-child cohort

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Background

The Elfe (Etude Longitudinale Française depuis l'Enfance) national cohort study (2011)

Elfe is an on-going national French cohort study that aims at assessing the impact of environmental exposures and socioeconomic or familial factors on children's development, health, and behaviour (Vandentorren et al 2009). One way of assessing prenatal exposure to environmental pollutants is the determination of biomarker levels in biological samples taken at the child's birth, from the mother or the umbilical cord. This approach will be applied by the French Institute for Public Health Surveillance on the Elfe study's biological collection, and constitutes the **perinatal part of the French Biomonitoring Program**, whose main objectives are :

- to describe the biological levels of environmental pollutant biomarkers among pregnant women and newborns;
- to establish national reference values when possible and;
- to identify and quantify the main factors associated with these observed levels.

Mother-child pairs were included in 2011 in the national cohort, at delivery. The results of biomarker determination will be available in 2014.

The Elfe pilot study (2007)

In October 2007, a pilot study was carried out to test the feasibility of the Elfe biological sample collection and to get a first estimate of some environmental pollutant biomarkers levels (Oleko et al 2011).

The aim of this work is to present the biomonitoring results from the Elfe 2007 pilot study and how they were used for the design of perinatal part of the French Biomonitoring Program.

Methods

Population of the Elfe Pilot study

Parturient women were included in October 2007, at the time of delivery, in 30 hospital maternities from 2 French regions, including 5 counties: Seine-Saint-Denis, Ardèche, Isère, Loire and Savoie. Inclusion criteria were to deliver a living child (single or twin birth) at least at 33 weeks of amenorrhea. Among the 571 women who delivered between October 1st and 4th according to these criteria, **296** agreed to participate to the biological part of the study.

Data collection

Information about maternal and child characteristics, or other covariates were collected through the mother's medical chart, face-to-face or telephonic interviews or self-administered questionnaires, at birth and 6 to 8 weeks after.

Biological samples and analysis

- Maternal blood and urine, as well as cord blood were collected by a midwife in the delivery room.
- Maternal hair and breast milk were collected by a trained interviewer during the days following delivery.
- Mature breast milk was collected at home by the mother, 1 month after delivery.

After collection, urine and milk samples were aliquoted in 10 mL polypropylene vials and stored at -80°C until biomarkers analysis. Analysed biomarkers, analytical method and limits of quantification are indicated in table 1.

Statistical analysis

When the percentage of left-censored values (<LOD or LOQ) was low (<15%), values below the LOD were replaced by LOD/2 and values between the LOD and LOQ were replaced by (LOD+LOQ)/2. When this percentage was higher (≥15%), the ROS method was used.

Statistical analyses were conducted using SAS version 9.1 and the SURVEY package in R version 2.12.

Results

Descriptive results (see table 1) show that

- More than 90% of the women who were included in the study had detectable levels of biomarkers for **BPA, phtalates, pyrethroid pesticides, dioxins, furans, PCBs, brominated flame retardants and perfluorinated compounds.**
- **Organotins, atrazine and propoxur** metabolites were detected in less than 25% of the women.
- Despite the wide use of **glyphosate**, its metabolites were not detected in any sample.

These results are similar to other previously published results among general population or pregnant women for most of the compounds.

TABLEAU Biomarker analysis in the Elfe pilot study, October 2007

Substances	Biomarkers	Matrix	Analytical Method	LOQ	% ≥ LOQ	Median
BPA (n=254)	Total BPA Free BPA	Urine	GC-MS	0.3 µg/L	**88.2%** 61.4%	2.5 µg/L 0.4 µg/L
Phtalates (n=279)	MnBP, MiBP, MEHP, 5OH-MEHP, 5oxo-MEHP, 5cx-MEPP, 2cx-MEPP	Urine	LC-MS/MS	0.5 µg/L	99.7% to **100%**	11.8 µg/L to 53.7 µg/L
Cotinine (n=210)	Cotinine	Urine	LC-MS/MS	0.07 µg/L	60%	0.26 µg/L
Organotins (n=273)	MBT, DBT, TBT, MOcT, DOcT, TOcT, MPhT, DPhT, TPhT	Urine	GC-ICP-MS	0.005 to 0.024 µg/L	0-24%	Not calculated (%<LOQ to high)
Pesticides (Pyr. n=247 Others n=238)	**Pyrethroids (**3-BPA, F-BPA, Br2CA, cis-Cl2CA, trans-Cl2CA)	Urine	HPLC-MS/MS	0.021 to 0.029 µg/L	83 to **96**% (F-PBA <5%)	0.08 to 0.32 F-PBA<LOQ
	Atrazine (Mercapturate, Desethyl, Desisopropyl, Desethyl-desisopropyl, Hydroxy, Hydroxy-desethyl, Hydroxy-desisopropyl, Hydroxy-desethyl-desisopropyl)		LC-MS/MS	0.003 to 0.90 µg/L	1.3 to 23.1%	All < LOQ
	Glyphosate / AMPA		HPLC-MS/MS	0.05 µg/L	0%	<LOQ
	Propoxur / 2-isopropoxyphenol		LC-MS/MS	0.065 to 0.08 µg/L	13.0% / 1.7%	<LOQ
Dioxins Furans PCBs (n=44)	**Dioxins** 2, 3, 7, 8 – TetraCDD 1, 2, 3, 7, 8 - PentaCDD 1, 2, 3, 4, 7, 8 - HexaCDD 1, 2, 3, 6, 7, 8 - HexaCDD 1, 2, 3, 7, 8, 9 - HexaCDD 1, 2, 3, 4, 6, 7, 8 - HeptaCDD OctaCDD	Mature breast milk	GC-HRMS	0.02 to 0.93 µg/L	95.5 to **100**%	1 to 59 pg/g lipids
	Furans 2, 3, 7, 8 - TetraCDF 1, 2, 3, 7, 8 - PentaCDF 2, 3, 4, 7, 8 - PentaCDF 1, 2, 3, 4, 7, 8 - HexaCDF 1, 2, 3, 6, 7, 8 - HexaCDF 1, 2, 3, 7, 8, 9 - HexaCDF 2, 3, 4, 6, 7, 8 - HexaCDF 1, 2, 3, 4, 6, 7, 8 - HeptaCDF 1, 2, 3, 4, 7, 8, 9 - HeptaCDF OctaCDF			0.02 to 0.33 µg/L	6.8 to **100**%	<LOQ to 10 pg/g lipids
	PCBs **DL** (77, 81, 126, 169, 105, 114, 118, 123, 156, 157, 167, 189) **NDL** (28, 52, 101, 138, 153, 180)			0.23 to 13041 µg/L	**DL** 23 to **100**% **NDL** 93 to **100**%	**DL** <LOQ to 14 ng/g lipids **NDL** 5 to 77 ng/g lipids
BFRs PFCs (n=48)	**PBDEs** (28, 47, 99, 100, 153, 154, 183, 209), **PBBs** (52, 101, 153), **HBCD** (α, β, γ)	Mature breast milk	GC-ICP-MS	0.09 to 1.5 µg/L	PBDEs and PBBs **100**% HBCD 17 to **100**%	PBDEs 0.03 to 0.63 ng/g lipids PBBs 0.02 to 0.16 ng/g lipids HBCDs <LOQ to 1.12 ng/g lipids
	PFCs (PFBA, PFPA, PFHxA, PFHpA, PFOA, PFNA, PFDeA, PFUnA, PFDoA, PFBS, PFHxS, PFHpS, PFOS, PFDeS)		LC-MS/MS	0.03 to 0.15 µg/L	PFOA. PFHxS. PFOS >**90**% Others : 0 to 12.5%	PFOA. PFHxS. PFOS 0.05 to 0.08 µg/L Others <LOQ

Further analysis show that

- For **phtalates and BPA**: our results also suggested a potential contamination of women with these compounds, from medical devices (catheterization or urine probes), at delivery (Vandentorren *et al* 2011).
- For **cotinine**: comparison between self-reported smoking status and biochemical values showed discrepancies (misclassification rate >16%), suggesting underreporting of smoking status during pregnancy by the mothers.
- For **dioxins, furans and PCBs**: comparison of our results to those from a previous study conducted in France in 1999 shows that levels of dioxins in maternal milk decreased from 40% in the time interval between the two studies.

Conclusions

This pilot study was conducted on a sample of women that provided results on maternal environmental exposure in France in 2007. On one hand, it was useful to select relevant biomarkers to include in the national study. On the other hand, the results allowed us to estimate the number of subjects to be analyse for each biomarker in order to determine reference values for the corresponding pollutants.

Main feedback from the pilot, used for the perinatal part of the French Biomonitoring Program

- Special precautions needed to prevent contamination during the biological collection.
- Usefulness of free BPA determination along with total BPA, to assess recent environmental contamination.
- Usefulness of cotinine biological values, to deal with misclassification errors in reported smoking status.
- No further investigation on organotins.
- Relevance of dioxins, furans and PCBs determination even if not used anymore.

References

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