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Assessment of Exposure to Organic and Inorganic Pollutants in Children's Hair

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Abstract

A new approach is developed to determine people's exposure to organic and inorganic pollutants through hair analysis. This study involved 44 children. Two strands of hair were analyzed: to seek organic pollutant by liquid chromatography coupled with mass spectrometry and flight time (LC-MS-QTOF) and inorganic pollutant by inductively coupled plasma spectrometry with mass detector (ICP-MS). For organic pollutants, 100% of children show traces with an average of 38 toxicants per child with a predominance of synthetic intermediates (18%), insecticides (16%) and fungicides (13%). The 5 most commonly found products are metaldehyde (43 times), sorbic acid (43 times), hymecromone (42 times), hydroxychinolin (37 times) and pyrocatechol (34 times). For, 100% of children show inorganic pollutants (Metal Trace Element) at a rate at the limit of the toxic values we call “CONTROLLED LEVEL” with an average of 16 metal trace elements (MTE) per child. The MTE most often found are: aluminum, titanium, nickel, antimony, arsenic (43 times). For some elements, we define for the first-time the values in the hair.

Keywords

Pollutants, Exposure, Hair, Children, Organic, Inorganic, Synthetic Intermediate, Pesticides, Element Trace Metallic, Metal

1. Introduction

The World Health Organization has assessed pesticides as "very dangerous" and may have acute and chronic toxic effects, especially on children [1]. WHO also indicates that it does not have enough data to assess the impact of pesticides on our health, as exposure can be occupational or industrial, but also through the consumption of food and water [2].

Metals represent 4 of the 10 chemicals that are a major public health concern according to WHO [3]. The dispersion of metals in the environment no longer follows the old schemes of the "contaminated industrial site" or "waste disposal" type contaminated by plants. An example of metallic trace elements found in the environment is the use of synthetic marble to make kitchens: the resulting dust is potentially toxic [4]. There is now a second way that is becoming increasingly important in terms of the quantities produced and used around the world: inorganic nanoparticles. The quantities used in France are particularly high and exceed 100 000 tones/year [5].

Long-term exposure to these various pollutants leads to disruptions in our metabolic and hormonal systems [6, 8], leading to chronic diseases such as autoimmune diseases like diabetic, thyroiditis, obesity [9, 12], cancers [13, 14] and other infertility [15, 17] or neurodegenerative disorders [18, 20].

How to assess our pollutants impregnation? The study will determine the exposures to organic and inorganic pollutants. The work aims to identify the most common pollutants in children's hair. This screening will cover more than 1,700 organic and 40 inorganic pollutants in hair, which is the best matrix to reveal long-term exposure to one or more pollutants [21].

2. Methods

2.1. Population Studied

The population studied concerns 44 children of both sexes from different French regions of urban and rural. The ages range from 8 to 14 years old. We did not divide the sample by gender. To keep the children's anonymity, the Laboratory did not know the children's names, age, sex and address. The children's hair was collected in specific conical tubes of about 3 cm representing an average of 3 months of exposure. The samples are taken in duplicate and placed in different tubes, the first tube is used for LC-MS-QTOF analysis and the second tube for ICP-MS analysis.

2.2. Organic Pollutants Analysis

These analyses were performed on a liquid chromatography model 1260 (LC) coupled with a mass spectrometer and a flight time model 6545 (MS-QTOF) (Agilent, France).

The separation column used is a NUCLEOSHELL® Bluebird RP 18 column, 2.1x mm ID, 100 mm length and 2.7 µm particle size (Macherey Nagel, Germany).

The hair is washed with ultrapure water, then methanol. And then the hair is ground with a acetonitrile/acetone mixture 7V/3V. The extraction is done with a stirrer for 2 hours. The extract is evaporated using the SpeedVac centrifugal vacuum evaporator (Thermo-Fisher, France). The dry extract is taken up by 100 µl from a mixture of water / methanol 8V/2V. This eluate is filtered on a ChromFil cartridge (Macherey Nagel, Germany) and injected into LC-MS-QTOF.

For LC, the flow rate is set at 0.5 mL / min, the column temperature is set at 50°C and the injection volume is 10 µL. For eluent A: formic acid at 0.1% in ultrapure water 18.2 Mega Ohms, for eluent B: formic acid at 0.1% in Methanol LC/MS LGR (Labkem, Labbox, Spain). The gradient program defined as follows: in 5 min from 5 % to 100 % B, hold for 1.0 min, in 0.1 min to 5 % B, hold 5 % B for 3.9 min.

The data were processed in exact mass with the "MassHunter Workflow" software in relation to the "MassHunter Pesticides PCDL" database (Agilent, France).

Although the research is qualitative, we used a pesticide control purchased from LGC, Molsheim, France to evaluate a minimum impregnation of 0.1 ng/mg of hair. The signal obtained represents a minimum area of 100 000. Below this value, we consider that the pesticide or the pollutant is in the form of a trace: its clinical impact is low.

2.3. Inorganic Pollutants Analysis

These analyses were performed on a 7800 inductively coupled plasma mass spectrometer (ICP-MS) (Agilent, France).

The calibration curves extend from 0.001 µg/L to 25 µg/L and is performed with a dilution solution comprising 0.2% TraceMetal nitric acid (Fischer, UK), 0.1% Triton X100 (Agilent, France) in ultrapure 18 Mega Ohm water. To build these calibration curve, we use solution standard solutions No. IV-ICPMS-71A, No. IV-ICPMS-71B, No. MSHG-10PPM (Inorganic Venture, Canada). An exact amount of hair (20 to 40 mg) is mineralized in 69% TraceMetal Grade nitric acid (Fischer, UK). The solution obtained is diluted to 100th with solution standard solutions No. IV-ICPMS-71A, No. IV-ICPMS-71B, No. MSHG-10PPM (Inorganic Venture, Canada). The data is reprocessed with Agilent's "ICP Offline Data Analysis" software.

For our interpretation of the rates found in children’s hair, we used two publications[22, 23] that have established ranges of values for several elements in current population.

3. Results

3.1. Organic Pollutants

All children's hair contained organic pollutants, we found an average of 38 pollutants per child (table 1). We identified 265 different pollutants on a Agilent database of more than 1,700.

The 5 most commonly found products are Metaldehyde (43 times), sorbic acid (43 times), hymecromone (42 times), hydroxychinolin (37) and Pyrocatechol (34 times). We also found carbamates, parabens, phthalates and bisphenol A.

Table 1. *summary of organic pollutants found in children’s hair screening by LC QTOF.*

Matched pollutant found			
Totaly (44 children)	**Minimum (number/chlid)**	**Maximum (number/chlid)**	**Average (number/chlid)**
1669	18	62	38

Table 2. *Percent of persistent organic pollutant’s natures have been identified in children’s hair.*

Organic pollutants Nature	Number oftimes detected	Pourcent
Synthesis intermediary	292	18%
Insecticide agent	258	16%
Fungicide agent	213	13%
Herbicide agent	150	9%
Natural component	119	7%
Degradation product	118	7%
Food additive	97	6%
Deworming	94	6%
Growth regulator	48	3%
Chemical compound	46	3%
Molluscicide	43	3%
Medication	43	3%
Synergist	29	2%

Organic pollutants Nature	Number oftimes detected	Pourcent
Avicide	28	2%
Insect repellent	23	1%
Rodenticide agent	22	1%
Artefact	13	1%
Acaricide agent	11	1%

These products, distributed according to their "nature" (Table 2), show the predominance of "synthetic intermediates" (18%) widely used in chemistry and well known for their long-term toxic effects. They are not researched and therefore rarely present in the biological matrices of the population. Next come insecticides (16%) and fungicides (13%), which are more generally identified, as shown in the leaflet produced in 2017 by the French “Mutuelle Sociale Agricole (MSA)” and published on their website [24].

3.2. Inorganic Pollutants

First, we have separated the metal trace elements (MTE) into two parts: those that induce high toxicity at low levels (CRITICAL-MTE) and those that induce high toxicity at high levels (NON-CRITICAL-MTE). Table 3 summarizes the CRITICAL-MTE, these elements should only be found in the state of ultra-trace in the hair minimum value and NON-CRITICAL-MTE.

Second, we have established a threshold called: " CONTROLLED LEVEL ", we consider for the "CRITICAL-MTE" that the " CONTROLLED LEVEL " applies when the rate of the element is higher than the minimum value found in both articles[22, 23] and for the "NON-CRITICAL-MTE" that the " CONTROLLED LEVEL " applies when the rate of the element is higher than the maximum value found in both articles[22, 23]. Table 2 shows the analysis elements and the selected CONTROLLLED LEVEL.

***Table 3.** distinction between CRITICAL-MTE and NON-CRITICAL-MTE.*

Metal Trace Elements	CONTROLLED LEVEL	Elements
CRITICAL MTE (17)	above the minimum threshold	Al, Ti, V, Cr, Ni, As, Sr, Mo, Cd, Sn, Sb, Te, Ba, Hg, Th, Pb, U
NON-CRITICAL MTE (32)	above the maximum threshold	Be, B, Na, Mg, K, Ca, Mn, Fe, Co, Cu, Zn, Ga, Se, Zr, Ag, Cs, La Ce, Pr, Nd, Sm, Eu, Gd, Dy, Ho, Er, Tm, Yb, Hf, Ta, W, Th

All children have metal trace elements (MTE) in their hair. The quantities found were compared to our threshold called: "CONTROLLED LEVEL" (table 4).

***Table 4.** The CONTROLLED LEVELS for Metals Trace Elements in the hair.*

Metal Trace Elements	CONTROLLED LEVEL (µg/g)
Beryllium	0.0120
Boron	3.3000
Sodium	670.0
Magnésium	141.0
Aluminium	0.100
Potassium	420.0
Calcium	2890
Titanium	0.04
Vanadium	0.001
Chromium	0.03
Manganèse	2.41
Iron	23.0
Cobalt	0.1400
Nickel	0.0020
Copper	96.0
Zinc	209.0
Gallium	0.0680
Arsenic	0.0015
Selenium	1.84
Strontium	0.14
Zirconium	1.21
Molybdenum	0.01
Silver	1.96
Cadmium	0.004
Tin	0.007
Antimony	0.003
Tellurium	0.0003
Cesium	0.0019
Barium	0.05
Lanthanum	0.1060
Cerium	0.1640
Hafnium	0.0370
Tantalum	0.0200
Tungsten	0.0210
Mercury	0.053
Thallium	0.0001
Lead	0.13
Thorium	0.0044
Uranium	0.002

This study also focused on elements not yet studied and measured in the hair matrix. Table 5 proposes values for these elements.

100% of children are polluted with an average of 16 metallic trace elements at "CONTROLED LEVEL" with a maximum of 21 and a minimum of 11 elements.

***Table 5.** Hair values for some Metal Trace Elements not available until now.*

Metal Trace Elements	Values (µg/g)			
	Average	Minimum	Maximum	Median
Praséodyme	0.0007	0.0001	0.0027	0.0005
Néodyme	0.0024	0.0003	0.0104	0.0017
Samarium	0.0014	0.0003	0.0061	0.0012
Europium	0.0005	0.0001	0.0023	0.0004
Dysprosium	0.0014	0.0002	0.0126	0.0007
Holmium	0.0004	0.0001	0.0031	0.0002
Erbium	0.0011	0.0001	0.0096	0.0006
Thulium	0.0003	0.0001	0.0021	0.0002
Ytterbium	0.0022	0.0003	0.0159	0.0009

The most commonly found elements are aluminum, titanium, nickel, antimony, arsenic (97.7%), followed by tin and barium (95.4%). Mercury, vanadium and uranium was found in 90.9% of cases, lead 81.8% and cadmium in 61.4% (table 6).

Table 6. *Percent of the Metal Trace Elements with CONTROLLED LEVEL have been identified in hairs of 44 children.*

Metal Trace Elements	CONTROLLED LEVEL found	
	Number of times detected as positive	Percent
Beryllium	0	0%
Boron	0	0%
Sodium	0	0%
Magnésium	0	0%
Aluminium	43	97.73%
Potassium	0	0%
Calcium	0	0%
Titanium	43	97.73%
Vanadium	40	90.91%
Chromium	35	79.55%
Manganese	0	0%
Iron	0	0%
Cobalt	5	11.36%
Nickel	43	97.73%
Copper	0	0%
Zinc	1	2.27%
Gallium	0	0%
Arsenic	42	95.45%
Sélénium	0	0%
Strontium	41	93.18%
Zirconium	0	0%
Molybdènium	39	88.64%
Silver	0	0%
Cadmium	27	61.36%
Tin	42	95.45%
Antimoiny	43	97.73%
Tellure	18	40.91%
Cesium	6	13.64%
Baryum	42	95.45%
Lanthanum	3	6.82%
Cerium	0	0%
Gadolinium	1	2.27%
Hafnium	1	2.27%
Tantale	0	%
Tungsten	9	20.45%
Mercury	40	90.91%
Thallium	6	13.64%
Lead	36	81.82%
Thorium	10	22.73%
Uranium	40	90.91%

4. Discussion

To evaluate the impregnation of pollutants, we chose the hair matrix that reflects this chronic exposure. Our study was carried out on 44 children from different French regions, with rural and urban lifestyle.

4.1. Organic Pollutants

This work shows an average presence of 38 organic pollutants per child. The results are also different from one child to another, so this reflects each child's habits (food, environment, clothing…).

Synthesis intermediates and insecticides are the most common biomarkers. These analyses make it possible to establish children's exposure to pollutants in the environment by establishing a kind of profile. Very few studies have been carried out on screening research into traces of organic pollutants in children's hair.

4.2. Inorganic Pollutants

This work shows an average presence of 16 metallic trace elements at CONTROLLED LEVEL and per child. The results are also different from one child to another, so this reflects each child's habits (food, environment, clothing…).

Aluminum, titanium, nickel, antimony, arsenic are the most common biomarkers but we found also tin, barium, mercury, vanadium, uranium, lead and cadmium. These analyses make it possible to establish children's exposure to pollutants in the environment by establishing a kind of profile. Very few studies have been carried out on screening research into traces of inorganic pollutants in children's hair. The values found are comparable to those found in the two publications of Goullé et al [22] and Rodushkina et al [23].. To our knowledge for the new elements, we are the only ones to have brought values (table 5).

Let us remember that our children are in full neural, hormonal and growth process development. The accumulation of different pollutants and their interactions lead to a disruption in these complex development processes. These changes will therefore have important consequences in the future, as shown by the increasing evolutions of autoimmune diseases and cancers, infertility and neuronal development problems (decrease IQ). This impact will be more or less significant depending on whether the exposure is spaced over time (chronic toxicity) and the level (abundance) of pollutants found.

This developed approach is therefore to screen for organic molecules and trace elements in the hair. The scientific literature does not show such a similar comprehensive approach involving a search for organic and inorganic pollutants. The research should be as broad as possible in order to best reflect the toxicity of the pollutants found individually but also in interaction with each other.

The knowledge of the various pollutants, associated with our database (toxicity and sources of pollutants) allows us to react and modify our behavior and habits (e. g. presence of parabens or metals present in cosmetics, plasticizers in plastic containers, flame retardants in clothing mainly...) in order to eliminate pollutants from our environment.

5. Conclusion

The knowledge of the various pollutants, associated with our database (toxicity and sources of pollutants) allows us to react and modify our behavior and habits (e. g. presence of parabens or metals present in cosmetics, plasticizers in plastic containers, flame retardants in clothing mainly...) in order to eliminate pollutants from our environment.

This work will serve to provide a starting point for all scientists who will want to quantify and explain the emergence of diseases due to these "pollutants" in the coming years. Our children are fragile, they are perfect and privileged targets, they are the guinea pigs of the chemical world.

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