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Discovering time-trends of the German populations exposure to contaminants by analysis of human samples of the German Environmental Specimen Bank (ESB)

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Highlights

- German ESB archives biological material of about 500 young adults annually since 35 years.
- Real-time monitoring (RTM) of HBM parameters in freshly taken samples reveals exposure situation directly.
- Archived samples can be used for retrospective analysis of new appearing contaminants.

- Human biomonitoring (HBM) data of the German ESB identify long-term trends in exposure.
- German ESB is a valuable tool to reveal necessity and success of regulatory measures.

Abstract

The German Environmental Specimen Bank (ESB) is a monitoring instrument of the German Federal Ministry for the Environment, Nature Conservationand Nuclear Safety. The permanent biobank facility is run since 1981 containing environmental and human samples from Germany. All samples are collected according to standard operating procedures (SOP). An annually standardized collection of human samples at four different regional sites of the country has been established since 1997. Routine sampling is done once a year, recruiting healthy non occupationally exposed students aged 20-29 years, in an equal gender distribution. The number of participants recruited is approximately 120 students per site and year. Directly after the annual sampling process, the human samples are analyzed for selected environmental chemicals. The time-trends of lead in blood, mercury and pentachlorophenol in 24 h-urine and polychlorinated biphenyls in plasma demonstrated a decrease of exposure during the last two decades by about 40 – 90 percent. In parallel retrospective studies using cryo-archived samples revealed increasing time trends of emerging chemicals used as substitutes for regulated toxicants.

The data demonstrates the great relevance of the ESB for the health related environmental monitoring and shows the importance of human biomonitoring as a tool in information based policy making.

Keywords Human biomonitoring; HBM; biobank; environmental specimen bank, chemical exposure

Introduction

The German Environmental Specimen Bank (ESB) is an important element of the health-related environmental monitoring system in Germany. The ESB is an archive of environmental and human samples for monitoring and evaluating the general quality of the environment as well as the body burden of chemicals in young adults in Germany (Wiesmüller et al. 2007; Wiesmüller and Gies 2011). A focus of the ESB is assessing and documenting human exposure to relevant contaminants by concomitant analysis in the year of sampling as well as retrospective analysis of archived samples to evaluate time-trends. Data generated from the ESB samples enables the surveillance of phased-out and emerging chemicals in humans and the environment. The evaluation of long-term trends in human chemical exposure is important for the development and success control of legal regulations and restrictions.

The human part of the German ESB was founded in 1974 in Münster (Western Germany) and started with a pilot phase, in which conditions of optimal storage, systematic recruitment and sampling procedures were examined (Kemper et al. 1993). Regular sampling and storage of human specimens in the biobank started in 1981. After the German reunification the spatial sampling design was extended successively up to four sampling sites by including the cites Halle/Saale (Central-East Germany) in 1995, Greifswald (Northern Germany) in 1996 and Ulm (Southern Germany) in 1997 (Figure 1).

While in the early years human specimen like scalp and pubic hair were also sampled, the focus lies now on standardized sampling of whole blood, blood plasma and 24 h-urine. All samples are cryo-preserved at temperatures below -130° C. Exposure-relevant behaviours of each volunteer are recorded by standardized, self-administered online-questionnaires. Aliquots of each sample are analysed for selected human biomonitoring (HBM) parameters within the year of sampling. Due to this approach resulting in a prompt discovery of the exposure situation of the recent year it was called “real-time monitoring” (RTM).

This study presents the time-trends of lead in blood, mercury and pentachlorophenol in 24 h-urine and polychlorinated biphenyls in plasma during the last two decades.

Material and methods

Recruitment of volunteers and sampling have been described in detail before (Lermen et al. 2014). Briefly, once a year volunteers are recruited via online registration targeting healthy students aged 20-29, studying at the sampling sites. A container for collecting 24 h-urine is sent in combination with an information kit by parcel. The number of participants recruited is approximately 120 students per site and year, in an equal gender distribution.

Personal metadata (e.g. sex, age, place of birth, body height, body weight), medical history (e.g. health and dental status, and medication) as well as information about exposure relevant behaviour (e.g. nutrition, alcohol consumption, smoking habits, use of body care products) and other sources of exposure (e.g. home and living surrounding situation) are collected using a standardized, self-administered online questionnaire, that is in part verified by the field operators in a face-to-face interview.

In the period 1997-2004, 24 h-urine, blood (whole blood and blood plasma), scalp and pubic hair, and salivary samples had been collected; since 2005 the number of matrices was reduced and only urine, whole blood and blood plasma are being collected. Since 2013 sampling has been conducted under highly standardized conditions in a mobile lab unit (Lermen et al. 2014). This guarantees the same facility and infrastructure specifically adapted to the needs of the sampling procedure of the German ESB and an identical treatment of all samples at all sampling sites. Blood samples are prepared (plasma extraction) and portioned immediately after withdrawal. After sample preparation cholesterol, triglycerides, total protein, and creatinine are measured in plasma samples immediately. In 24 h-urine samples, volume, density, conductivity and creatinine content are measured immediately after delivery. All samples are stored temporarily in a mobile liquid nitrogen tank at < -130°C for transportation to the permanent

biobank in Münster-Wolbeck (Lermen et al. 2014). All respective processes including quality assurance and quality control are accredited according to DIN EN ISO/IEC 17025 and certificated according to DIN EN ISO 9001.

Until 2005, the human specimens were kept at a temperature of -80 to -85°C. Since 2006, the specimens have been stored under stable deep-freezing conditions in the gaseous phase above liquid nitrogen at a temperature of < -130°C (Wiesmüller and Gies, 2011).

After completing all sampling processes of the year, laboratory analyses are performed sequentially (real-time-monitoring, RTM). Until 2010, 64 inorganic elements and 6 organic parameters (hexachlorobenzene in plasma (HCB-P), the polychlorinated biphenyl congeners PCB138, PCB153 and PCB180 in plasma, pentachlorophenol in plasma (PCP-P) and urine (PCP-U)) were analyzed (Wiesmüller and Gies 2011). Since 2011, the RTM parameter spectrum was reduced to 11 inorganic elements, while for organic chemicals the focus was put on retrospective monitoring. Recent RTM parameters are lead in blood (Pb-B), copper (Cu-P) selenium (Se-P) and zinc in plasma (Zn-P), total arsenic (As_{total}-U), cadmium (Cd-U), calcium (Ca-U), copper (Cu-U), mercury (Hg-U) and strontium (Sr-U) in urine. Inorganic elements are measured by different inductively coupled plasma mass spectrometry (ICP-MS) techniques (HR-ICP-MS, ICP-DRC-MS) and atomic absorption spectrometry (AAS). The analytical procedures of the recent RTM parameters are listed in Table 1. The analysis of PCP, HCB, and PCB congeners was done after liquid liquid extraction by gas chromatography mass spectrometry (GC-MS). For the determination of PCP a derivatisation was performed using diazomethane before GC-MS analysis. Most of the analytical procedures are based on methods approved by the DFG Working Group for the Analyses of Hazardous Substances in Biological Materials (Göen et al. 2012b). To avoid bias of analytical reproducibility, all samples of the year are analyzed after randomization.

Sampling, analyzing, and archiving are performed according to ESB standard operating procedures (Lermen et al. 2015 a,b,c). All analyses are conducted under conditions of quality

control and quality assurance. Reproducibility control is performed by analyzing of quality control samples in each series and evaluation by control charts. Accuracy is controlled by the analysis of certified reference materials (CRM) and the successful participation in the German External Quality Assurance Scheme (GEQUAS, Göen et al. 2012a).

Results

The time-trends of the internal exposure to environmental chemicals assessed by RTM analyses are displayed for lead in blood, mercury in urine, the polychlorinated biphenyl congener PCB153 in plasma and pentachlorophenol in urine in Figures 2-5. Moreover, the time-trend of the urinary levels of the trace element calcium is presented in Figure 6.

Figure 2 shows blood lead levels for both the period of parallel sampling at four sites (since 1997) and the previous term of sampling at Münster only. The time course demonstrates a clear and distinct decrease during the 1980s, which continued subsequent however with a less steep slope. Nevertheless, since 2010 blood lead levels (geometric mean, GM) are nearly on a constant level with only small fluctuation between the years. The blood lead level (GM) of the participants in Münster decreased by about 87 % from 78.0 µg/L in 1981 to 10.1 µg/L in 2017. From 1997 to 2017 the lead concentration in blood of the participants at all four sites decreased by about 48 % from approximately 21.2 to approximately 10.9 µg/L (Lermen et al. 2018a).

In 2017, the geometric means of Pb-B in the populations of the different sampling sites ranged from 10.1 µg/L (Münster) to 12.0 µg/L (Greifswald).

Figure 3 shows the mercury concentration in 24 h-urine of the participants at all four sampling sites for the period 1997-2017. The time course of this parameter demonstrates a considerable decline of the exposure at all four sites. However, the comparison between the sites demonstrates different magnitudes of exposure to mercury, in which the participants at Münster and Ulm show comparably lower exposure than the other sites. The GM values range in 1997 from 0.329 µg/L to 0.564 µg/L and decreased to a range of 0.040 µg/L to 0.059 µg/L in 2017.

Figure 4 shows the concentration of PCB153 in blood plasma of the participants at all four sampling sites for the period 1997-2010. For the internal level of this organic contamination a clear decrease within the observation period was shown. The PCB153 level in plasma (GM) of the participant at all four sites decreased from approximately 0.49 µg/L in 1997 to approximately 0.18 µg/L in 2010 by about 63 %. The parameter did not show a clear difference between the individual sampling sites.

Figure 5 shows the concentrations of PCP in urine of the participants at all four sampling sites for the period 1997-2010. For the urinary level of this contaminant a distinct decrease was found over the investigated period. The GM of the urinary PCP level of all participants decreased by about 90 % from approximately 0.73 µg/L in 1997 to approximately 0.07 µg/L in 2010. The parameter did not show a clear difference between the individual sampling sites.

Figure 6 shows the time-trends for calcium in urine at the four sampling sites. For the urinary level of this essential trace element a decline was also found within the observation period. The overall GM decreased from 109 mg/L in 1997 to 62 mg/L in 2017. No difference was found between the four sampling sites.

Discussion

The observed decrease of blood lead levels within the last three decades proves the success of the ban of lead in gasoline in Germany as well as worldwide. A decrease of blood lead levels due to a ban of lead was also observed in other European and other industrial countries (Bierkens et al. 2011; Muntner et al. 2005). Tsoi and coworkers (2016) analysed and compared the blood lead levels in the eight NHANES surveys of the US population since 1999. They found a distinct decline of blood lead levels in children (1-5 years), adolescents (6-19 years), adults (20-59 years) and the elderly population (≥60 years). For the adult US population the mean blood lead level declined from 12.6 µg/L in 1999/2000 to 7.1 µg/L in 2013/2014 in females and from 20.6 µg/L in 1999/2000 to 10.2 µg/L in 2013/2014 in males, which is very similar to the

results in the German ESB. A further exploration of the ESB data revealed significantly lower blood lead levels in women compared to men too as well as lower levels in individuals with low alcohol consumption. Both effects have been demonstrated in other studies too (Lermen et al. 2018a). Whereas the gender effect is clearly attributed to the different haematocrit between men and women, the reason for the effect of alcohol is still unclear. However, it is well known that regular alcohol consumption increases the corpuscular erythrocyte volume (Kilo et al. 2016), which may increase the capability for lead absorption.

The decreasing trend of urinary mercury levels may be explained by different reasons. First, there are several indications that the general environmental mercury contamination in Germany decreased during the last decades (Rüdel et al. 2010). The urinary mercury levels in ESB RTM samples are in the same range with exposure levels in the Czech Republic found between 2001-2003, which revealed a GM of mercury in urine samples of adults of 0.61 µg/g creatinine (Batáriova et al. 2006). Second, an impact of dental amalgam on the urinary mercury level has to be considered. Halbach et al. (2000) demonstrated that the removal of dental amalgam resulted in a clear decline of urinary mercury levels. Moreover, data of the German Environmental Survey of 1998 revealed a dependence of urinary mercury on the number of amalgam fillings (Becker et al. 2003). Thus, the reduction of dental amalgam applications during the last decades may contribute to the decline of the urinary excretion of mercury too. The indicated difference between individuals of the eastern part of Germany and western Germany is also confirmed by the data of the German Environmental Surveys GerES II and GerES III (Schulz et al. 2007). A more detailed exploration of the ESB data for mercury will be published elsewhere (Lermen et al., in preparation).

The decline of the PCB153 concentration in plasma samples of young adults in Germany is in accordance with the ban of the production and application of polychlorinated biphenyls in most industrial countries from the end of the 1970s up to the end of the 1980s (ECC 1985). The decrease of PCB153 and other PCB congeners was found in other studies of individuals of the

population in Germany too (Fromme et al. 2015; Lackmann et al. 1996; Schettgen et al. 2015). Moreover, the decline of the exposure to these compounds was also demonstrated for the Swedish population by two independent studies (Hardell et al. 2010; Axmon et al. 2008). Hardell and coworkers (2010) investigated the time trend of persistent compounds in plasma samples withdrawn from 537 Swedish individuals (controls in different human cancer studies) during the five years period 1993-1997 and found a statistical decline of the internal PCB exposure by 7.2 % per year. Axmon and coworkers (2008) compared the persistent organochlorine pollutants in the serum samples of young male citizens of Sweden taken in 2000 (n=274) and 2004 (n=200). They found a distinct decline of the internal exposure to PCB153. The median value of PCB153 was 0.31 µg/L in 2000 and 0.09 µg/L in 2004, which demonstrates a decline of 18 % per year.

The decrease of urinary PCP levels within the last two decades is also in compliance with the German regulation for the ban of the use of pentachlorophenol as biocide in 1989. The decrease of PCP exposure was also demonstrated by the course of the urinary PCP levels in the German Environmental Surveys (Schulz et al. 2007).

The decline of the concentration of the essential trace element calcium in the 24 h samples of the ESB cannot be explained by time trend of calcium exposure. However, it has to be emphasized that the urinary volume excreted by the participants during 24 hours has been increasing, which resulted in a dilution of the urinary samples compared to previous sampling dates. After correction of the urinary calcium level by the 24 h-urine volume the trend was not observed anymore (data not shown). A detailed exploration of the physiological standard parameters including calcium level and 24 h-urine volume will be presented elsewhere (Lermen et al. 2018b).

Additionally to the RTM programme, other human biomonitoring parameters were determined in retrospective studies by special requests and indications in the archived samples. By this approach, the decline of the exposure of the German population to previously prominent, but banned perfluorinated compounds and to primary prominent plasticizers, e.g. di-(2-

ethylhexyl)phthalate and di-n-butylphthalate (DEHP and DnBP), as well as the emerging of substitutes, e.g. di-(2-propylheptyl)phthalate (DPHP) and 1,2-cyclohexane dicarboxylic acid diisononyl ester (DINCH), has been revealed (Göen et al. 2011; Koch et al. 2017; Schröter-Kermani et al. 2013; Schütze et al. 2014; Schütze et al. 2015).

In principle, two independent coexisting approaches are used for the retrospective or prospective monitoring of the exposure of populations to environmental contaminants: (1) retrospective HBM surveys of representative subsets of a population, e.g. NHANES and GerES, and (2) bio-banking of human materials of a representative population subset at a certain date or period of time. Repetitive HBM surveys request high efforts of sampling logistics and analytical costs but provide a direct control of temporal trends. In general, HBM survey samples are usually not archived beyond the survey term and thus do not allow a supplement of additional parameters afterwards. In contrast, bio-banks of human samples needs efforts for sampling only once and for the cryo-archive operation but enables a retrospective exploration of the exposure to contaminants of emerging concern in the future. The German ESB is an unique tool, which achieves to combine both approaches through the continuous yearly sampling and archiving of human specimens. The RTM analysis enables a direct control of temporal trends like the repetitive HBM surveys while the ESB archives allow a retrospective examination of new emerging questions as well. Certainly, the exploration of the German ESB data is limited due to unrepresentative sampling and the restriction on a special age interval. On the other hand, students are regarded as a proper subgroup for analysing general trends of exposure because they are a homogenous subgroup with similar socio-demographic features, high mobility, and little or no occupational exposures.The special design of the ESB also helps to control important factors like the half-life of the HBM parameter. The half-lifes of HBM parameters range from a few hours for some elements and organic metabolites up to several years for persistent compounds, e.g. polychlorinated biphenyls and perfluorinated compounds. Generally, persistent compounds are determined in blood or blood plasma regularly, whereas non-persistent

parameters are determined in urine. While spot urine concentration of short-time parameters can in principle be affected by an inhomogenous partition of the exposure during the day, any effect of the circadian fluctuation on the results is avoided by gathering 24 h urine sampling. This approach assures the monitoring of the total daily elimination and, moreover, allows for the estimation of the daily intake using the concentration of the HBM parameter, the volume of the 24h urine sample and the excretion fraction factor of the urinary excreted parameter.

Conclusions

Time trend analyses of the internal exposure to chemicals by HBM supplies useful and reliable information on the success of regulatory measures. The data revealed the effect of the ban and restriction of chemicals in consumer products and industrial goods, e.g. the ban of lead in gasoline or the ban of the production and use of polychlorinated biphenyls in the EU. They demonstrate the great relevance of the ESB as one component of the German health related environmental monitoring system (Kolossa-Gehring et al. 2012) and show the importance of human biomonitoring as a tool in information based policy making.

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Conflict of interest

The authors declare no conflict of interest.

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References

Axmon, A., Hagmar, L., Jönsson, B.A.G., 2008. Rapid decline of persistent organochlorine pollutants in serum among young Swedish males. Chemosphere 70, 1620-1628. https://doi.org/10.1016/j.chemosphere.2007.07.077

Barátiová, A., Spevácková, V., Benes, B., Cejchanová, M., Smid, J., Cerna, M., 2006. Blood and urine levels of Pb, Cd and Hg in the general population of the Czech Republic and proposed reference values. Int. J. Hyg. Environ. Health 209, 359-366. https://doi.org/10.1016/j.ijheh.2006.02.005

Becker, K., Schulz, C., Kaus, S., Seiwert, M., Seifert, B., 2003. German Environmental Survey 1998 (GerES III): environmental pollutants in the urine of the German population. Int. J. Hyg. Environ. Health 206, 15-24. https://doi.org/10.1078/1438-4639-00188

Bierkens, J., Smolders, R, Van Holderbeke, M., Cornelis, C., 2011. Predicting blood lead levels from current and past environmental data in Europe. Sci. Total Environ. 409, 5101-5110. https://doi.org/10.1016/j.scitotenv.2011.08.034

ECC – Council of the European Communities, 1985. Council Directive 85/467/EEC of 1 October 1985, amending for the sixth time (PCBs/PCTs) Directive 76/769/EEC on the approximation of the laws, regulations and administrative provisions of the Member States relating the restrictions on the marketing and use of certain dangerous substances and preparation.

Fromme, H., Albrecht, M., Appel, M., Hilger, B, Vökel, W., Liebl, B, Roscher, E., 2015. PCBs, PCDD/Fs and PBDEs in blood samples of a rural population in South Germany. Int. J. Hyg. Environ. Health 218, 41-16. https://doi.org/10.1016/j.ijheh.2014.07.004

Göen, T., Dobler, L., Koschorreck, J., Müller, J., Wiesmüller, G.A., Drexler, H., Kolossa-Gehring, M., 2011. Trends of the internal phthalate exposure of young adults in Germany – Follow-up of a retrospective human biomonitoring study. Int. J. Hyg. Environ. Health 215, 36-45; http://doi.org/10.1016/j.ijheh.2011.07.011

Göen, T., Schaller, K.H., Drexler, H., 2012a. External quality assessment of human biomonitoring in the range of environmental exposure levels. Int. J. Hyg. Environ. Health 215, 229-232. http://doi.org/10.1016/j.ijheh.2011.08.012

Göen, T., Eckert, E., Schäferhenrich, A., Hartwig, A., 2012b. Allocation of reliable analytical procedures for human biomonitoring published by the DFG Senate Commission for the Investigation of Health Hazards of Chemical Compounds in the Work Area. Int. J. Hyg. Environ. Health 215, 233-237. http://doi.org/10.1016/j.ijheh.2011.08.013

Halbach, S., Welzl, G., Kremers, L., Willruth, H., Mehl, A., Wack, F.X., Hickel, R., Greim, H., 2000. Steady-state transfer and depletion kinetics of mercury from amalgam fillings. Sci. Total Environ. 259, 13-21. https://doi.org/10.1016/S0048-9697(00)00545-3

Hardell, E., Carlberg, M., Nordström, M, van Bavel, B., 2010. Time trends of persistent organic pollutants in Sweden during 1993-1997 and relation to age, gender, body mass index, breast-feeding and parity. Sci. Total Environ. 408, 4412-4419. https://doi.org/10.1016/j.scitotenv.2010.06.029

Kemper, F.H., 1993. Human organ specimen banking – 15 year of experience. Sci. Total Environ. 139-140, 13-25.

Kilo, S., Göen, T., Eckert, E., Drexler, H., 2016. Evaluation of biomarkers of regular alcohol consumption in an occupational setting. Int. Arch. Occup. Environ. Health 89, 1193-1203; http://doi.org/10.1007/s00420-016-1155-1

Koch, H.M., Rüther, M., Schütze, A., Conrad, A., Pälmke, C., Apel, P., Brüning, T., Kolossa-Gehring, M., 2017. Phthalate metabolites in 24-h urine samples of the German Environmental Specimen Bank (ESB) from 1988 to 2015 and acomparison with US NHANES data from 1999 to 2012. Int. J. Hyg. Environ. Health 220 (2), 130-141. https://doi.org/10.1016/j.ijheh.2016.11.003

Kolossa-Gehring, M., Becker, K., Conrad, A., Schröter-Kermani, C., Schulz, C., Seiwert, M., 2012. Environmental surveys, specimen bank and health related environmental monitoring in Germany. Int. J. Hyg. Environ. Health 215(2), 120-126. http://doi.org/10.1016/j.ijheh.2011.10.013

Lackmann, G.M., Göen, T., Töllner, U., Schaller, K.H., Angerer, J., 1996. PCBs and HCB in serum of full-term German neonates. The Lancet 348, 1035; https://doi.org/10.1016/S0140-6736(05)64967-7

Lermen, D., Schmitt, D., Bartel-Steinbach, M., Schröter-Kermani, C., Kolossa-Gehring, M., von Briesen, H., Zimmermann, H., 2014. A new approach to standardize multicenter studies: mobile lab technology for the German Environmental Specimen Bank. PLOS ONE 9(8), e105401. https://doi.org/10.1371/journal.pone.0105401

Lermen, D., Bartel-Steinbach, M., Jost, N., 2015a. German Environmental Specimen Bank (ESB) - Guideline for Sampling and Sample Processing 24h-Sampling Urine. https://www.umweltprobenbank.de/en/documents/publications/23843

Lermen, D., Bartel-Steinbach, M., Jost, N., 2015b. German Environmental Specimen Bank (ESB) - Guideline for sampling and sample processing whole blood and blood plasma. https://www.umweltprobenbank.de/en/documents/publications/23863

Lermen, D., Bartel-Steinbach, M., Jost, N., 2015c. German Environmental Specimen Bank (ESB) - Guideline for Characterization of Human Samples: Analysis of Clinical Chemical Parameters. https://www.umweltprobenbank.de/en/documents/publications/23883

Lermen, D., Göen, T., Bartel-Steinbach, M., Eckard, R., Conrad, A., Rüther, M., Von Briesen, H., Kolossa-Gehring, M., 2018a. Lead exposure of young adults in Germany – Long-term Trend and influencing factors by means of human biomonitoring in samples from the German Environmental Specimen Bank. Int. J. Hyg. Environ. Health, submitted

Lermen, D., Bartel-Steinbach, M., Gwinner, F., Conrad, A., Weber, T., von Briesen, H., Kolossa-Gehring, M., 2018b. Trends in Characteristics of 24-h Urine Samples and their Relevance for Human Biomonitoring Studies – 20 Years of Experience in the German Environmental Specimen Bank. Int. J. Hyg. Environ. Health, submitted

Muntner, P., Menke, A., DeSalvo, K.B., Rabito, F.A., Batuman, V., 2005. Continued decline in blood lead levels among adults in the United States: The National Health and Nutrition

Examination Surveys. Arch. Intern. Med. 165, 2155-2161. https://doi.org/10.1001/archinte.165.18.2155

Rüdel, H., Fliedner, A., Kösters, J., Schröter-Kermani, C., 2010. Twenty years of element analysis of marine biota within the German Environmental Specimen Bank – a thorough look at the data. Environ. Sci. Pollut. Res. 17, 1025-1034. https://doi.org/10.1007/s11356-009-0280-8

Schettgen, T., Alt, A., Esser, A., Kraus, T., 2015. Current data on the background burden to the persistent organochlorine pollutants HCB, p,p'-DDE as well as PCB 138, PCB 153 and PCB 180 in plasma of the general population in Germany. Int. J. Hyg. Environ. Health 218, 380-385. https://doi.org/10.1016/j.ijheh.2015.02.006

Schröter-Kermani, C., Müller, J., Jürling, H., Conrad, A., Schulte, C., 2013. Retrospective monitoring of perfluorocarboxylates and perfluorosulfonates in human plasma archived by German Environmental Specimen Bank. Int. J. Hyg. Environ. Health 216, 633-640. https://doi.org/10.1016/j.ijheh.2012.08.004

Schulz, C., Conrad, A., Becker, K., Kolossa-Gehring, M., Seiwert, M., Seifert, B., 2007. Twenty years of the German Environmental Survey (GerES): human biomonitoring--temporal and spatial (West Germany/East Germany) differences in population exposure. Int. J. Hyg. Environ. Health 210(3-4), 271-297. https://doi.org/10.1016/j.ijheh.2007.01.034

Schütze, A., Kolossa-Gehring, M., Apel, P., Brüning, T., Koch, H.M., 2014. Entering markets and bodies. Increasing levels of the novel plasticizer Hexamoll DINCH in 24h urine samples from the German Environmental Specimen Bank. Int. J. Hyg. Environ. Health 217, 421-426. https://doi.org/10.1016/j.ijheh.2013.08.004

Schütze, A., Gries, W., Kolossa-Gehring, M., Apel, P., Schröter-Kermani, C., Fiddicke, U., Leng, G., Brüning, T., Koch, H.M., 2015. Bis-(2-propylheptyl)phthalate (DPHP) metabolites emerging in 24 h urine samples from the German Environmental Specimen Bank (1999-2012). Int. J. Hyg. Environ. Health 218, 559-563. https://doi.org/10.1016/j.ijheh.2015.05.007

Tsoi, M.F., Cheung, C.L., Cheung, T.T., Cheung B.M.Y., 2016. Continual decrease in blood lead level in Amercans: United States National Health Nutrition and Examination Survey 1999-2014. Am. J. Med. 129, 1213-1218. https://doi.org/10.1016/j.amjmed.2016.05.042

Wiesmüller GA, Gies A., 2011. Environmental specimen bank for human tissues. Encyclopedia Environ. Health, 507-527. https://doi.org/10.1016/B978-0-444-52272-6.00450-5

Wiesmüller, G.A., Eckard, R., Dobler, L., Günsel, A., Oganowski, M., Schröter-Kermani, C., Schüter, C., Gies, A., Kemper, F.H., 2007. The environmental specimen bank for human tissue a spart of the German Environmental Specimen Bank. Int. J. Hyg. Environ. Health 210, 299-305. http://doi.org/10.1016/j.ijheh.2007.01.036

Captions

Figure 1: Geographic location of the sampling sites of the human part of the German Environmental Specimen Bank (ESB)

Figure 2: RTM results (geometric mean) of lead in blood of the period 1981-2017 (split up among the sampling sites)

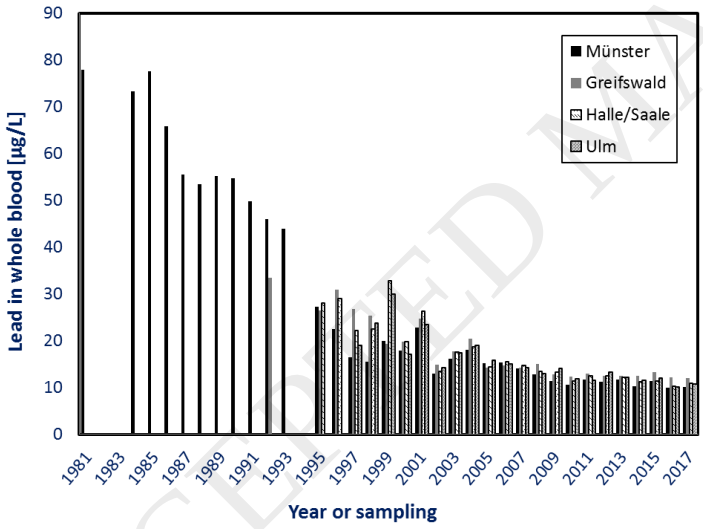

Figure 3: RTM results (geometric mean) of mercury in urine of the period 1997-2017 (split up among the sampling sites)

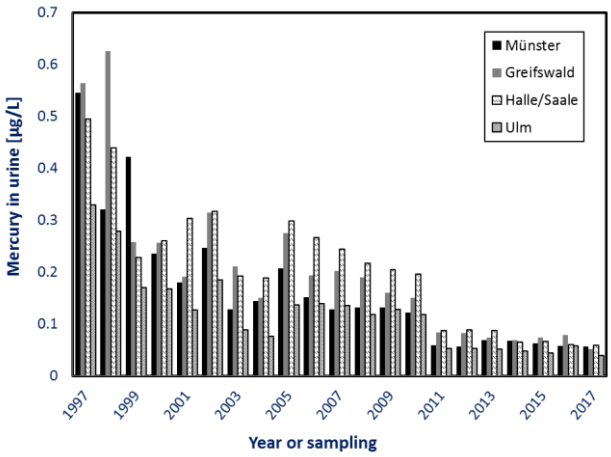

Figure 4: RTM results (geometric mean) of PCB153 in plasma of the period 1997-2010 (split up among the sampling sites)

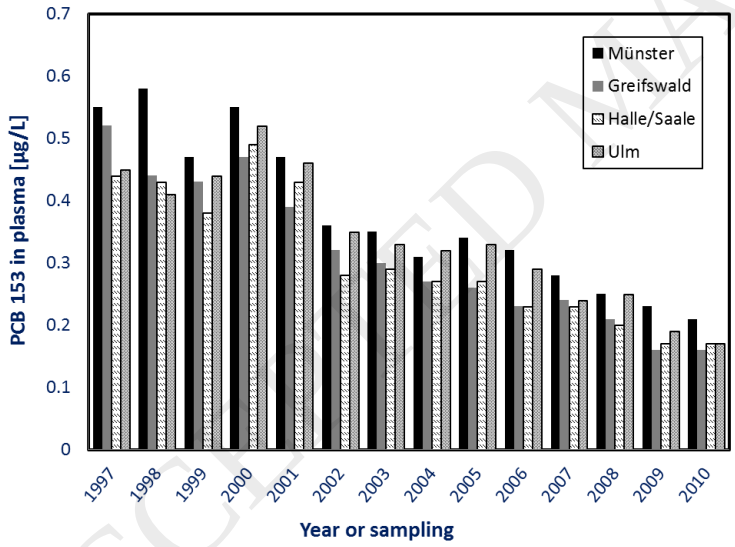

Figure 5: RTM results (geometric mean) of PCP in urine of the period 1997-2010 (split up among the sampling sites)

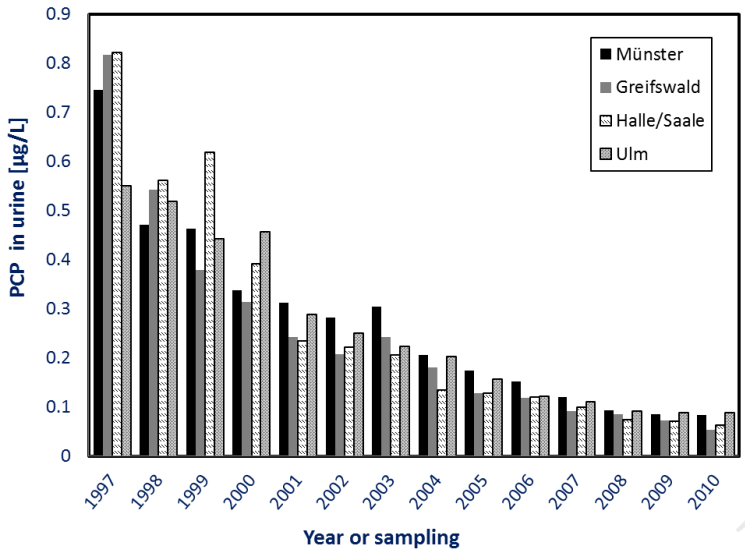

Figure 6: RTM results (geometric mean) of calcium in urine of the period 1997-2017 (split up among the sampling sites)

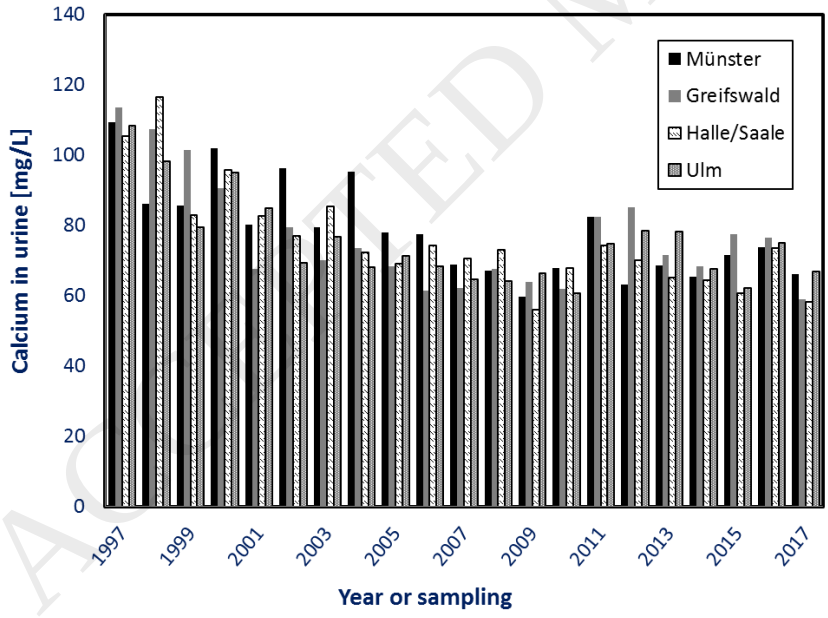

Table 1: RTM parameters (since 2011) and the analytical techniques applied

Element	Matrix	Analytical procedure
As_{total}	24 h-urine	ICP-DRC-MS
Ca	24 h-urine	ICP-DRC-MS
Cd	24 h-urine	ICP-DRC-MS
Cu	24 h-urine	ICP-DRC-MS
Cu	Blood plasma	ICP-DRC-MS
Hg	24 h-urine	FIMS-AAS
Pb	Whole blood	ICP-DRC-MS
Se	Blood plasma	ICP-DRC-MS
Sr	24 h-urine	ICP-DRC-MS
Zn	24 h-urine	ICP-DRC-MS
Zn	Blood plasma	ICP-DRC-MS

ICP-DRC-MS: Inductively Coupled Plasma Mass Spectrometry with Dynamic Reaction Cell

FIMS-AAS: Flow Injection Mercury System Atom Absorption Spectrometry