# Human Biomonitoring of Perfluorinated Substances in Adults Exposed to Contaminated Drinking Water in Regione Veneto (Italy).

De Felip E.<sup>1</sup>, Abballe A.<sup>1</sup>, Dellatte E.<sup>1</sup>, De Luca S., Fulgenzi AR., Iacovella N.<sup>1</sup>, <u>Ingelido AM</u>.<sup>1</sup>, Marra V.<sup>1</sup>, Russo F.<sup>2</sup>, Valentini S.<sup>1</sup>, Vazzoler M.<sup>2</sup>, and Musmeci L<sup>3</sup>.

#### Introduction

In 2006, results of the analysis of perfluorinated substances (PFAS) carried out in water samples of some European rivers under the European Project PERFORCE ("Perfluorinated Organic Compounds in the European Environment") showed that, among the rivers investigated, the river Po (Northern Italy) was the one with the highest concentrations of PFOA. Following this finding, analysis of PFAS in water was extended to other rivers<sup>2</sup>, and an area heavily contaminated by PFAS was identified in the Brenta river basins, in Veneto Region. In this area, groundwater contamination had affected also drinking water supplies.

In 2013 the Regione Veneto decided to assess exposure to PFAS of the population living in contaminated areas versus a control group of subjects residing in neighboring, not contaminated areas. Assessment of exposure was carried out through a human biomonitoring study, funded by the Regione and carried out by ISS in collaboration with the local health units of the areas of interest.

#### Materials and methods

Study population. Serum sampling was performed between July 2015 and April 2016 and involved 507 subjects, 257 residents in the areas affected by water contamination (exposed), and 250 residents in neighboring areas at estimated background exposure (not exposed); participants from each area were selected by gender and age with the same stratification criteria. Sampling areas were defined on the basis of data available on contamination by PFAS of the water supply chain. Areas of two ULSS (Unità Locale Socio Sanitaria, Local Health Unit), namely ULSS 5 and part of the ULSS 6, were identified as contaminated areas; areas of other four ULSS (8, 9, 15 e 22) plus a part of ULSS 6 not interested by water contamination were identified as control areas. The study was approved by the local ethical committees and each participant sign an informed consent before blood withdrawal. A questionnaire was administered to each study participant to obtain information on anthropometric and socio-demographic characteristics, lifestyle, drinking-water consumption, and diet.

Analysis. Serum samples were analyzed for nine perfluorocarboxylic acids (PFBA, PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFDeA, PFUnA, PFDoA) and three perfluorosulfonates (PFBS, PFHxS, PFOS). About 250µL of serum were spiked with labelled internal standards, extracted with acetonitrile and prepared for instrumental analysis carried out by HPLC MS/MS<sup>3</sup>.

<sup>&</sup>lt;sup>1</sup> Reparto di Chimica Tossicologica, Dipartimento Ambiente e connessa Prevenzione Primaria, Istituto Superiore di Sanità, Roma.

<sup>&</sup>lt;sup>2</sup> Regione del Veneto, Area Sanità e Sociale, Sezione Attuazione Programmazione Sanitaria, Settore Promozione e Sviluppo Igiene e Sanità Pubblica, Venezia.

<sup>&</sup>lt;sup>3</sup> Dipartimento Ambiente e connessa Prevenzione Primaria, Istituto Superiore di Sanità, Roma.

## Results and discussion

PFOA and PFOS were above the limits of quantification (LOQs) in all the analyzed samples, PFHpA, PFDA, PFUdA, PFHxS, PFNA were above respective LOQs in more than a half of the samples. The other five PFAS were less frequently detected, with detection frequency ranging from 12% to 42%. For PFBA, PFPeA, PFBS, PFHxA, PFHpA, PFHxS, PFOA, PFOS and PFDoA concentrations in exposed subjects were significantly higher (Mann Whitney test, p <0.05) than those of the residents of the control area. The highest difference was observed for PFOA (median value of the exposed group was eight times higher than the median value of the not exposed group), and the lowest for PFHxS and PFOS.

In order to identify subgroups at higher exposure the Mann Whitney test was performed, also stratifying the exposed group by ULSS (Figure 1).

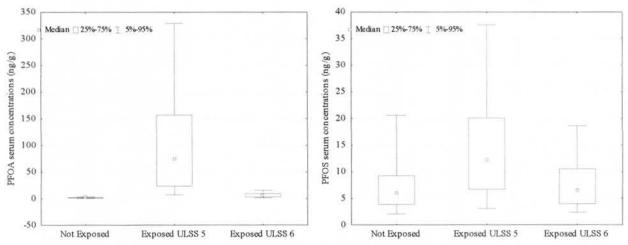


Figure 1. PFOA (left) and PFOS (right) serum concentrations (ng/g) in not exposed subjects and exposed subjects from ULSS 5 and 6.

Serum concentrations of all analytes, with the exception of PFDeA and PFUnA, resulted to be significantly higher in subjects residing in ULSS 5 than in subjects residing in contaminated areas from ULSS 6, as well as in subjects from not contaminated areas. Subjects from ULSS 5 had the highest levels of PFOA, with a median value 45 times higher than the not exposed group. Exposed subjects from ULSS 6 had serum concentrations significantly higher than the not exposed participants for PFBA, PFPeA, PFBS, PFHxA, PFHpA, PFOA and PFDoA.

The reasons of the remarkable difference in exposure between ULSS 5 and the contaminated areas of ULSS 6 are still under study and will be investigated through the analysis of the variables from questionnaires.

Figure 2 compares concentrations of PFOA and PFOS from this study with results from international studies carried out on populations exposed to water contaminated by PFAS<sup>4,5,6,7</sup>, and with results from national and international studies carried out on general population groups<sup>3,8,9</sup>. Serum concentrations of PFOA in the exposed group as a whole are comparable to concentrations assessed in populations exposed to PFAS, but if the comparison is restricted to the exposed subjects from ULSS 5, values observed are higher than those observed in other exposed populations. PFOS concentrations in exposed subjects are comparable with data from groups of exposed population in Germany and Sweden.

PFOA and PFOS concentrations in not exposed subjects are comparable to levels assessed in other studies carried out on general population.

For most of the other PFAS analyzed in this study, data available in literature are on the whole scarce. Within this limit, concentrations observed in this study result to be comparable to those observed in groups of the general population in other countries.

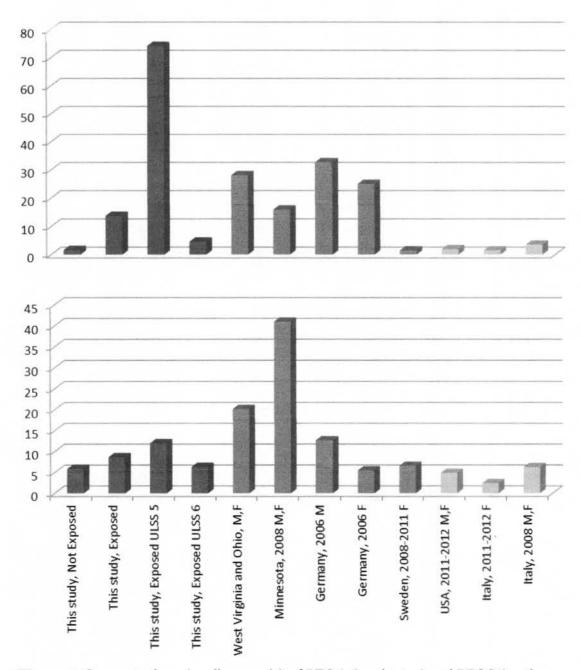


Figure 2. Concentrations (medians, ng/g) of PFOA (on the top) and PFOS (on the bottom) found in this study (in red), in other studies of populations exposed to contaminated water (in blue), and general population groups (in green)

## Acknowledgements

The authors are indebted to all medical doctors, nurses, and laboratory technicians from ULSS 5, 6, 8, 9, 15 and 22 who carried out enrolment and sampling. A special thank is due to Laura

Tagliapietra (Regione del Veneto, Area Sanità e Sociale, Sezione Attuazione Programmazione Sanitaria, Settore Promozione e Sviluppo Igiene e Sanità Pubblica), for her technical support in the enrolment planning.

This study was supported by a grant from Regione Veneto (Accordo di collaborazione tra Istituto Superiore di Sanità e Regione Veneto finalizzato al "Supporto tecnico scientifico, analitico e consultivo per l'analisi di rischio correlato alla contaminazione da PFAS di matrici ambientali e filiera idro-potabile in talune circostanze territoriali, e potenziale trasferimento di PFAS alla filiera alimentare e allo studio di biomonitoraggio" (2014-2017)).

## References

- de Voogt P., Berger U., de Coen W., de Wolf W., Heimstad E., McLachlan M., van Leeuwen S., van Roon A. (2006). <a href="http://www.science.uva.nl/perforce/Final%20reportA.pdf">http://www.science.uva.nl/perforce/Final%20reportA.pdf</a>.
- Polesello S., Pagnotta R., Marziali L., Patrolecco L., Rusconi M., Stefani F., Valsecchi S. (2013). <a href="http://www.minambiente.it/sites/default/files/archivio/allegati/reach/progettoPFAS\_ottobre2">http://www.minambiente.it/sites/default/files/archivio/allegati/reach/progettoPFAS\_ottobre2</a>
  013.pdf
- Ingelido A.M., Marra V., Abballe A.; Valentini S.; Iacovella N., Barbieri P.G., Porpora M.G., di Domenico A., De Felip E. (2010). Chemosphere 80, 1125-30.
- Frisbee S.J., Brooks P., Maher A., Flensborg P., Arnold S., Fletcher T., Steenland K., Shankar A., Knox S., Pollard C., Halverson J.A., Vieira V.M., Jin Leyden K.M., Ducatman A.M. (2009). EHP 117, 1873-1882.
- Brede E., Wilhelm M., Göen T., Müller J., Rauchfuss K., Kraft M., Hölzer J. (2010). Int J Hyg Environ Health 213, 217–223.
- Kari A., Johnson J., Williams A., Huset C. (2009). <a href="http://www.health.state.mn.us/divs/hpcd/tracking/biomonitoring/projects/pfcfinalrpt2009.pd">http://www.health.state.mn.us/divs/hpcd/tracking/biomonitoring/projects/pfcfinalrpt2009.pd</a>
- Gyllenhammar I., Berger U., Sundström M., McCleaf P., Eurén K., Eriksson S., Ahlgren S., Lignell S., Aune M., Kotova N., Glynn A. (2015). *Environ Res* 140 673–683.
- De Felip E., Abballe A., Albano F.L., Battista T., Carraro V., Conversano M., Franchini S., Giambanco L., Iacovella N., Ingelido A.M., Maiorana A., Maneschi F., Marra V., Mercurio A., Nale R., Nucci B., Panella V., Pirola F., Porpora M.G., Procopio E., Suma N., Valentini S., Valsenti L., Vecchiè V. (2015). Chemosphere 137, 1–8.
- Lewis R.C., Johns L.E., Meeker J.D. (2015). Int. J. Environ. Res. Public Health 12, 6098-6114.