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Impact of PFAS exposure on prevalence of immune-mediated diseases in adults in the Czech Republic

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ABSTRACT

Background: Per- and polyfluoroalkyl substances (PFASs) are emerging environmental contaminants with multiple hazardous properties including immunomodulation potency. Human exposure to PFASs has been associated with various immune-mediated diseases and outcomes. This study aimed to investigate the association between PFAS exposure and immune-mediated diseases such as allergies, eczemas, and autoimmune diseases in a population of adults in the Czech Republic.

Methods: This study included 309 adults from the Central European Longitudinal Study of Parents and Children: Young Adults (CELSPAC: YA). 12 PFASs were measured in participants' serum by HPLC-MS/MS, 3 PFASs were removed from the subsequent analyses due to low detection frequency. The associations of 9 PFASs with 9 immune-mediated diseases were assessed by logistic regression. Furthermore, Bayesian kernel machine regression (BKMR) was used to estimate the effect of the PFAS mixture on immune-mediated diseases. All analyses were adjusted for sex, age, BMI, smoking, education, and family history of immune-mediated diseases. In cases of a statistically significant interaction of PFASs and sex, stratified analyses were performed for men and women.

Results: Perfluorooctanoate (PFOA) and perfluorooctane sulfonate (PFOS) were negatively associated with both atopic eczema (OR per IQR increase 0.58 (95% CI 0.37–0.90) for PFOA and 0.56 (0.32–0.95) for PFOS) and contact dermatitis (0.37 (0.16–0.85) for PFOA and 0.33 (0.11–0.94) for PFOS). Perfluoroundecanoate (PFUnDA) was negatively associated with pollen, dust, and mite allergy (0.62 (0.43–0.89)). BKMR modelling showed a negative tendency in the overall effect of PFAS mixture on immune-health outcomes. Based on the stratified analysis, sex was suggested to be an effect modifier in the association of PFOS and atopic eczema.

Conclusion: Our results contribute to the body of literature that observes the immunosuppressive effect of PFAS exposure during eczemas and allergies, both for PFASs individually and as a mixture.

1. Introduction

The immune system plays an essential role in protecting the human body against pathogens from the environment and against cancer (Corthay, 2014; Delves and Roitt, 2000). However, the immune system can be disrupted, which may lead to a range of disorders, including allergic and autoimmune diseases, whose prevalence has been steeply increasing worldwide (Akdis, 2021). Approximately 20%–30% of the world's population suffer from some type of allergic disease (World Allergy Organization, 2013), more than 3.6% suffer from asthma (Mattiuzzi and Lippi, 2020), and between 3% and 5% suffer from autoimmune diseases (Wang et al., 2015). Disruptions of the immune system are influenced by both heritable factors and, to a greater extent, by non-heritable factors, i.e., the environment (Brodin et al., 2015). Among the numerous environmental factors, exposure to chemical substances was suggested to play one of the key roles (Winans et al., 2011).

Per- and polyfluoroalkyl substances (PFASs) are a large group of synthetic compounds that have broad applications in the industrial and consumer sector. They have been used since the 1940s as emulsifiers for firefighting foams and fluoropolymer manufacturing due to their surfactant properties, as well as chemically resistant coatings and flame

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retardants due to their resistance to oxidative degradation (Dean et al., 2020). Some of PFASs are currently regulated, nevertheless, many of PFASs are abundantly present in the human environment and human matrices due to their slow degradation rate under natural conditions. In a body, PFASs tend to accumulate mainly in plasma, then in the kidney and liver (Verreault et al., 2005). PFASs in the human blood are predominantly bound to albumin and can be thus transported to organs of the body (Alesio et al., 2022; Forsthuber et al., 2020; Jian et al., 2018). Studies describe the association of PFAS exposure with kidney and testicular cancer, endocrine disruption, liver damage, and lower birth weight among other adverse effects (Fenton et al., 2021). Importantly, as PFASs are frequently found in the blood, they can affect immune cells and the entire immune system.

Research of the immunomodulatory potency of PFASs in cohort studies found PFASs associated with both the deregulation of mostly adaptive immunity and apical adverse outcomes such as immune-mediated diseases. Strong evidence is reported for an association of PFAS exposure with suppressed antibody responses after vaccination (Abraham et al., 2020; Grandjean et al., 2017a; Kielsen et al., 2016; Looker et al., 2014; Timmermann et al., 2020) that has been also reported from studies of prenatal PFAS exposure in mother-child cohorts (Grandjean et al., 2017b; Granum et al., 2013). In addition, PFAS exposure has been found to contribute to the severity of COVID-19 disease (Catelan et al., 2021; Grandjean et al., 2020) and to increase the risk of other infectious diseases (Bulka et al., 2021; Kvalem et al., 2020; Timmermann et al., 2020). Higher susceptibility to infectious diseases based on prenatal PFAS exposure was also reported in mother-child cohorts (Ait Bamai et al., 2020; Dalsager et al., 2021; Granum et al., 2013; Wang et al., 2022). Further, asthma has been positively associated with PFAS exposure (Averina et al., 2019; Dong et al., 2013; Kvalem et al., 2020; Timmermann et al., 2017; Zhu et al., 2016). Allergic outcomes have been associated with PFAS exposure (Ait Bamai et al., 2020; Averina et al., 2019; Buser and Scinicariello, 2016; Kvalem et al., 2020), nevertheless in both directions, positive and negative associations. Autoimmune diseases associated with PFAS exposure have been reported within a population exposed to extremely high environmental concentrations (Steenland et al., 2015; Kyle et al., 2013). Taken together, PFASs were shown to be a chemical stressor that is convincingly connected with immune changes that affect the progression and/or severity of immune-mediated diseases.

Most epidemiological studies investigated the association between PFAS exposure and immune-mediated diseases in child populations or mother-child pairs, however, studies investigating PFAS-related effects on the immune system among adult populations are scarce. Therefore, our study was specifically focused on the adult population to investigate the impact of PFAS exposure on immune-mediated diseases such as asthma, allergies, and autoimmune diseases. We employed the Central European Longitudinal Studies of Parents and Children: Young Adults (CELSPAC: YA) study representing the adult population in the Czech Republic.

2. Materials and methods

2.1. Study population

Data for this study originate from the CELSPAC: YA study. Young Adults (YA) cohort is an ongoing follow-up study of the Czech part of the ELSPAC birth cohort (European Longitudinal Study of Pregnancy and Childhood) that was initiated in 1991–1992 in the Czech Republic, namely in Brno and Znojmo region. Detailed information about ELSPAC-CZ study is provided in Piler et al. (2017). For our research goal, we examined available participants (309 individuals) from YA cohort re-assessed in 2019. All these participants were involved in the study voluntarily. The CELSPAC: YA study was approved by the ELSPAC Ethics Committee (Ref. No: ELSPAC/EK/2/2019, dated March 13, 2019). In summary, participants underwent examinations including blood collection, measurement of height and weight, and filling questionnaires. Questionnaires including both the health condition and the health history were filled with health practitioner assistance. Other questionnaires (i.e., family status, education, smoking, among others) were filled out by the participants themselves. Among YA cohort participants, 12 immune-mediated diseases were identified, namely: asthma; pollen, dust, and mite allergy; food allergy; drug allergy; insect stings allergy; contact dermatitis; atopic eczema; psoriasis; rheumatoid arthritis; lupus (SLE); celiac disease; inflammatory bowel disease. Rheumatoid arthritis, lupus (SLE), and inflammatory bowel disease were omitted from subsequent analyses due to low occurrence among participants (less than 5 cases, i.e., 1.6% of participants).

2.2. Analysis of PFASs in blood samples

PFAS concentrations in participants' serum were measured by high-performance liquid chromatography coupled with tandem mass spectrometry (HPLC-MS/MS). Analyses were performed with the HPLC apparatus Agilent 1290 series (Agilent Technologies, Waldbronn, Germany) consisting of a vacuum degasser, a binary pump, a thermostated autosampler (10 °C), and a thermostatted column compartment kept at 30 °C. The column was SYNERGI 4μ Fusion MAX-RP 80Ä 100 mm × 2 mm i. d., equipped with Phenomenex SecurityGuard C18 4 × 2 mm. The mobile phase consisted of Methanol: 5 Mm ammonium fluoride in ratio 55:45 (A) and Methanol (B). The binary pump gradient was non-linear. A gradient program was used for elution: 0% B (initial), 0–70% B (from 0.1 to 4 min), 70–100% B (from 4 to 6 min). After 1.5 min, the ratio was decreased to 0% B and held for 3.5 min for column equilibration before next injection. The flow rate of the mobile phase was 0.40 mL/min 10 μL of individual sample was injected for the analyses. The mass spectrometer was an AB Sciex Qtrap 5500+ (AB Sciex, Concord, ON, Canada) with electrospray ionization (ESI). Ions were detected in the negative mode. The ionization parameters were as follows: capillary voltage −4.5 kV; desolvation temperature 450 °C; Curtain gas 15 psi, Ion Source Gas1 50 psi, Ion Source Gas2 30 psi. Quantitative analysis was performed using the Analyst software.

12 different PFASs were analysed in serum samples: perfluoropentanoate (PFPA), perfluorohexanoate (PFHxA), perfluoroheptanoate (PFHpA), perfluorooctanoate (PFOA), perfluorononanoate (PFNA), perfluorodecanoate (PFDA), perfluoroundecanoate (PFUnDA), pefluorododecanoate (PFDoDA), perfluorobutane sulfonate (PFBS), pefluorohexane sulfonate (PFHxS), pefluoroheptane sulfonate (PFHpS), perfluorooctane sulfonate (PFOS)). Three PFASs (PFHxA, PFHpA, PFDoDA) had more than 60% values below the limit of quantification (LOQ) and thus were omitted from the subsequent analysis. Values of LOQ and limit of detection (LOD) for individual PFASs can be seen in Supplementary information (SI, Table S1).

2.3. Data processing and statistical analysis

Data were processed in R programming software (v4.1.0; R Core Team, 2021). To visualize our a priori beliefs in the causal structure in the data, a directed acyclic graph (DAG) was created (Textor et al., 2016). Based on DAG (SI, Fig. S1) and together with information from the literature (von Holst et al., 2021), a minimal sufficient set of confounders was identified and used in the subsequent analysis: sex, age, body mass index (BMI), smoking, education, and family history of immune-mediated diseases. Due to the potential U-shape relationship between BMI and a health outcome (Jørgensen et al., 2016), BMI was used as a categorical variable of 4 levels: underweight (<18.5 kg/m^2), normal weight (18.5–24.9 kg/m^2), overweight (25.0–29.9 k kg/m^2), and obesity (≥ 30 kg/m^2) according to the World Health Organization (WHO) and the National Institute of Health (NIH) classification (Weir and Jan 2022). Smoking was assessed as a binary variable: smoker and non-smoker based on an answer to the question: "Have you smoked

more than 100 cigarettes during your life?". Education is representative of socioeconomic status and entered the analysis in three categories following the International Standard Classification of Education: primary education, secondary education, and university education (Eurostat, 2022). Family history of immune-mediated diseases (further referred to as "family history") was determined based on the answer to the question: "Do any of your parents or siblings have or have had an allergy and/or asthma and/or atopic eczema?" with possible answers "yes" or "no". PFAS concentration values were log-transformed to reduce the potential impact of outliers in the exposure distribution. To further moderate the potential impact of outliers on our results, "Winsorize" function from the "DecsTools" package (v0.99.44; Signorell et al., 2022) was used. 90% winsorizing was used, this method set all outliers below the 5th percentile and above the 95th percentile to the value of the 5th percentile, resp. 95th percentile. Data below LOD or between the LOD and the LOQ were imputed using the maximum likelihood method for fitting statistical distributions from the "mle" function of the "stats4" package (v4.1.0). Based on the PFASs with 100% detection, a log-normal distribution was chosen to be fitted for each PFAS using all data above LOQ. Once this distribution was known, new values to replace the left-censored values were randomly generated. The highest generated values were used to impute left-censored values $<$ LOQ but $>$ LOD, the rest were used to impute left-censored values $<$ LOD. Missing data in confounding variables (smoking, n = 5; family history, n = 8; education, n = 3) were imputed by the Multiple Imputation Chained Equation (MICE) method from the "mice" package (v3.14.0; Buuren and Groothuis-Oudshoorn, 2011) using other confounding variables (i.e., sex, age, BMI, smoking, education, and family history, except the one that was just imputed) and all exposure variables as an input. In case of missing values of health outcome, the participant was omitted from the analysis for the particular disease. Differences in prevalence of immune-mediated diseases between sexes were tested by Chi-square test and Fisher's exact test (if the expected values $<$ 5). Spearman correlation analysis was performed to obtain a correlation relationship between individual serum PFAS concentrations. Differences in PFAS serum concentration between women and men were tested by Mann-Whitney-Wilcoxon test.

Logistic regression was used to express the association between serum PFAS concentrations and the prevalence of diseases employing the "glm" function from the "stats" package (v4.1.0). Beta coefficients sourced from logistic regression, that correspond to log odds ratio were converted to odds ratio (OR) and expressed as OR per interquartile range (IQR) increase together with 95% confidence intervals (CI). Potential influential outliers identified during regression diagnostic were removed from the dataset and the analysis was run again as a sensitivity analysis. There have been described recommendations to use risk ratio (RR) for cohort studies instead of OR due to its better interpretability (Knol et al., 2012; Ranganathan et al., 2015). Therefore, RRs were calculated as a part of the sensitivity analysis. To obtain RR, Poisson regression with robust standard errors was used. Poisson regression is commonly used for the outcome of type "count", however, it can be also used for a binary outcome, when standard errors are adjusted by the "sandwich" function from the "sandwich" package. The resulting values then correspond to log risk ratio with respective standard errors, from which confidence intervals can be calculated (W. Chen et al., 2018b; Zou, 2004). The effect was then expressed as RR per IQR increase with 95% CI.

To assess the effect of the PFAS mixture, Bayesian kernel machine regression (BKMR) was used. It was employed for the outcomes identified in statistically significant observations in logistic regression analysis. BKMR model estimates multidimensional predictor-response function h(z) and can employ exposures, that are correlated with each other. A detailed description of the function and its application can be found in Bobb et al. (2018). For the purpose of our study with a binary outcome, probit regression was employed (Bobb, 2018). Models were run with 10,000 iterations and were adjusted for the same set of confounders as previous analyses (sex, age, BMI, smoking, education, and family history). The BKMR model produces Posterior Inclusion Probability (PIP) that describes the estimated importance of the individual PFAS towards the final effect (i.e., health outcome). PIPs are ranging from 0 to 1, and the greater the PIP is, the greater contribution the individual component has to the mixture effect. The overall effect of the mixture on the particular health outcome was expressed graphically as a quantile graph. The quantile graph shows the estimated change in effect (e.g., immune-mediated disease prevalence) when all the exposures at a particular percentile are compared to when all the exposures are at their 50th percentile. Moreover, a univariate exposure-response function with 95% credible intervals for each PFAS in mixture was visualized. The univariate exposure-response function shows the relationship between one exposure and the outcome while other exposures are fixed on their median values.

The statistically significant observations identified in the logistic regression analysis were tested for interaction between PFAS and sex as different effect for males and females has been reported in the literature (Q. Chen et al., 2018a; Impinen et al., 2019; Kvalem et al., 2020; Okada et al., 2014). For observations with statistically significant interaction, a stratified analysis was performed separately for women and men; this include i) logistic regression to discover if the effect in the stratified population will be different from the whole-population effect and ii) BKMR modelling to explore how the effect of PFAS mixture differs in women and men.

3. Results and discussion

3.1. Population characteristics

A general characteristic of the study population is summarized in Table 1. The population consists of a comparable number of men and women, a total of 309 participants (sex-stratified general characteristics are presented in SI, Table S2). The vast majority of the population was

Table 1
Main characteristics of the study population (sub-set of the YA cohort study, n = 309); values represent median with 1st quartile and 3rd quartile for age, and the number of participants with the corresponding percentage in brackets for other variables.

Characteristic		
Sex		
	women	158 (51.1%)
	men	151 (49.9%)
Age (years)	all participants	27 (26; 27)
BMI		
	underweight	12 (3.9%)
	normal weight	197 (63.8%)
	overweight	79 (25.6%)
	obesity	21 (6.8%)
Smoking		
	yes	96 (31.1%)
	no	208 (67.3%)
	missing information	5 (1.6%)
Education		
	primary education	3 (1.0%)
	secondary education	75 (24.3%)
	university education	228 (73.8%)
	missing information	3 (1.0%)
Family history		
	yes	171 (55.3%)
	no	130 (42.1%)
	missing information	8 (2.6%)

BMI = body mass index, underweight: $<$18.5 kg/m^2, normal weight: 18.5–24.9 kg/m^2, overweight: 25.0–29.9 k kg/m^2, obesity: $\geq$30 kg/m^2.
Smoking = smoking status. Was determined based on the answer to the question: "Have you smoked more than 100 cigarettes during your life?".
Family history = family history of immune-mediated diseases. Was determined based on the answer to the question: "Do any of your parents or siblings have or have had an allergy and/or asthma and/or atopic eczema?".

investigated around 27 years old and generally of normal weight, non-smokers, with high education. Prevalence of the immune-mediated diseases is shown in Table 2. The most prevalent outcome was pollen, dust, and mite allergy (41%). In the case of the stratified population by sex, pollen, dust, and mite allergy remained the most prevalent in both sex-specific groups (47% in men, 37% in women). Although the global prevalence of allergic diseases is estimated to be 20–30% (World Allergy Organization, 2013), the prevalence of sensitization to inhalant allergens (including pollen, dust, and mite) in the populations of Europe and the United States reaches more than 40% (Wheatley and Togias, 2015). Thus, the prevalence in our study population corresponded to the European population.

3.2. Serum PFAS concentrations in the study population

PFOA and PFOS reached higher concentrations than other PFASs with a median of 1.11 ng/mL and 1.93 ng/mL respectively (Table 3). Correlations between all 9 PFASs were assessed, and most of them were observed to be correlated with each other (SI, Fig. S2). The serum PFAS concentrations observed in our study are among the lower concentrations reported in the European context (EU HBM Dashboard, 2023). For example, medians of PFASs in the plasma of pregnant women in the MoBa cohort (n = 1943) were 2.54 ng/mL for PFOA and 12.87 ng/mL for PFOS (Impinen et al., 2019). A study conducted in Greenland, Poland (Warsaw), and Ukraine (Kharkiv) with a total of 307 men observed means of 4.0 ng/mL for PFOA and 27.2 ng/mL for PFOS (Leter et al., 2014). Even some of the children cohorts have relatively higher concentrations than observed in our study, e.g., a Norwegian study observed medians of 4.62 ng/mL for PFOA and 20.9 ng/mL for PFOS in serum of 10-year-old children (n = 378) (Kvalem et al., 2020). A serum concentration of 6.7 ng/mL for PFOS and 2.0 ng/mL for PFOA was observed in 13-year-old individuals (n = 74) within a population study from Faroe Island (Timmermann et al., 2017). Thus, we can consider our population as moderately exposed to PFAS in the context of European countries.

The distribution of PFAS concentrations among men and women in our study indicates sex-specific differences with lower concentrations in females, this difference was statistically significant for PFOA, PFNA, PFHxS, PFHpS and PFOS (Table 3). Statistically significant differences of PFAS concentration between men and women have been previously observed also in other population studies (Grandjean et al., 2020; Nyström et al., 2022). Based on pharmacokinetic modelling, it is suggested that menstruation, and thus regular loss of blood, can significantly contribute to reduced PFAS levels in blood among women (Lorber et al., 2015; Wong et al., 2014). This assumption is also supported by a birth cohort study from Faroe Island, where children were followed up until their 28 years (the number of participants varies from 122 to 399) (Shih et al., 2021). Concentrations of PFASs were measured in cord blood, then in serum at 7, 14, 22, and 28 years of age. The difference between concentrations in men and women was increasing with age, whereas women aged 28 years had significantly lower concentrations than men aged 28 years for all 5 measured PFASs (Shih et al., 2021). Interestingly, higher concentrations for males than females were also observed among 10-year-old children (Kvalem et al., 2020). Although, the sex-specific differences were observed in our study as well as in published studies, the detailed sex-specific pharmacokinetic mechanism of the PFAS behaviour in the human body is not fully understood.

3.3. Individual PFAS exposures and immune-mediated diseases

Estimates of the effect from logistic regression are shown in Table 4, expressed as OR (95% CI) per IQR increase; results including p-values can be found in SI, Table S3. Five observations were found to be statistically significant, all of them in inverse trend. Both eight-carbon PFASs showed a negative association with atopic eczema which

Table 2
Prevalence of the immune-mediated diseases in the whole study population, and separately for women and men.

	All participants (n = 309)				Women (n = 158)				Men (n = 151)				
	yes	no	NA	prev	yes	no	NA	prev	yes	no	NA	prev	p-value
Asthma	43	263	3	14%	24	134	0	15%	19	129	3	13%	0.67
Atopic eczema	47	258	4	15%	29	127	2	19%	18	131	2	12%	0.16
Celiac disease	6	302	1	2%	4	154	0	3%	2	148	1	1%	0.69
Contact dermatitis	12	296	1	4%	9	149	0	6%	3	147	1	2%	0.17
Drug allergy	44	264	1	14%	29	129	0	18%	15	135	1	10%	0.05
Food allergy	28	277	4	9%	23	134	1	15%	5	143	3	3%	**<0.01***
Insect stings allergy	21	285	3	7%	18	140	0	11%	3	145	3	2%	**<0.01***
Pollen, dust, and mite allergy	126	180	3	41%	58	99	1	37%	68	81	2	46%	0.15
Psoriasis	7	301	1	2%	2	156	0	1%	5	145	1	3%	0.27

*statistically significant difference (p-value <0.05) between women and men. NA = missing values; prev = prevalence, was calculated omitting the missing values.

Table 3
PFAS concentrations in serum (ng/mL) of all participants and of women and men separately.

	PFPA	PFOA	PFNA	PFDA	PFUnDA	PFBS	PFHxS	PFHpS	PFOS
All participants									
Median	0.13	1.11	0.31	0.13	0.06	0.06	0.32	0.04	1.93
1st quartile	0.08	0.86	0.22	0.10	0.04	0.02	0.21	0.02	1.33
3rd quartile	0.19	1.43	0.44	0.19	0.09	0.20	0.44	0.06	2.81
LOQ	0.04	0.07	0.012	0.01	0.012	0.04	0.014	0.017	0.09
>LOQ n (%)	300 (97%)	309 (100%)	309 (100%)	309 (100%)	303 (98%)	200 (65%)	309 (100%)	259 (84%)	309 (100%)
Women									
Median	0.13	1.03	0.29	0.13	0.06	0.07	0.24	0.03	1.58
1st quartile	0.08	0.78	0.19	0.09	0.04	0.03	0.16	0.02	1.09
3rd quartile	0.19	1.36	0.39	0.19	0.09	0.20	0.33	0.05	2.31
Men									
Median	0.12	1.17	0.34	0.13	0.06	0.05	0.41	0.05	2.34
1st quartile	0.08	0.97	0.25	0.11	0.04	0.02	0.32	0.03	1.66
3rd quartile	0.18	1.48	0.50	0.21	0.10	0.25	0.51	0.07	3.19
p-value	0.717	**0.002***	**<0.001***	0.233	0.636	0.324	**<0.001***	**<0.001***	**<0.001***

*statistically significant difference (p-value <0.05) between women and men. LOQ = limit of quantification.

Table 4
The association between individual PFASs and immune-mediated diseases; results are expressed as OR (95% CI) per IQR increase.

	PFPA	PFOA	PFNA	PFDA	PFUnDA	PFBS	PFHxS	PFHpS	PFOS
Asthma	1.30 (0.75–2.24)	1.29 (0.82–2.02)	1.22 (0.72–2.08)	1.00 (0.64–1.57)	0.86 (0.53–1.42)	1.27 (0.75–2.15)	1.41 (0.79–2.52)	0.92 (0.63–1.35)	1.15 (0.67–1.98)
Atopic eczema	0.90 (0.53–1.54)	**0.58* (0.37–0.90)**	0.58 (0.34–1.00)	0.72 (0.46–1.13)	0.74 (0.46–1.19)	1.02 (0.61–1.69)	0.69 (0.40–1.19)	0.70 (0.49–1.00)	**0.56* (0.32–0.95)**
Celiac disease	1.17 (0.29–4.78)	1.02 (0.34–3.06)	1.11 (0.28–4.33)	1.01 (0.30–3.41)	1.23 (0.31–4.94)	0.76 (0.20–2.88)	2.97 (0.67–13.12)	1.99 (0.47–8.41)	1.32 (0.32–5.40)
Contact dermatitis	1.77 (0.69–4.57)	**0.37* (0.16–0.85)**	0.36 (0.13–1.02)	0.52 (0.23–1.18)	0.56 (0.24–1.31)	0.93 (0.37–2.32)	0.50 (0.19–1.34)	0.68 (0.37–1.26)	**0.33* (0.11–0.94)**
Drug allergy	1.01 (0.59–1.73)	0.96 (0.62–1.47)	0.75 (0.44–1.26)	0.90 (0.58–1.39)	0.79 (0.49–1.27)	0.63 (0.38–1.07)	0.66 (0.38–1.15)	0.72 (0.50–1.03)	0.65 (0.38–1.11)
Food allergy	0.99 (0.50–1.94)	0.90 (0.52–1.54)	0.94 (0.49–1.79)	1.22 (0.70–2.11)	1.27 (0.69–2.36)	0.76 (0.39–1.47)	0.69 (0.34–1.44)	1.08 (0.65–1.77)	0.79 (0.41–1.56)
Insect stings allergy	1.48 (0.71–3.07)	0.80 (0.43–1.47)	0.51 (0.23–1.15)	0.77 (0.40–1.50)	0.69 (0.35–1.36)	0.71 (0.34–1.49)	1.83 (0.85–3.96)	1.07 (0.61–1.87)	0.94 (0.45–1.98)
Pollen, dust, and mite allergy	0.87 (0.59–1.30)	1.28 (0.92–1.76)	1.13 (0.77–1.66)	0.99 (0.72–1.38)	**0.62* (0.43–0.89)**	0.98 (0.68–1.43)	0.83 (0.55–1.26)	0.92 (0.69–1.23)	0.79 (0.53–1.16)
Psoriasis	0.50 (0.13–1.85)	0.38 (0.12–1.23)	0.72 (0.19–2.76)	0.73 (0.24–2.26)	1.21 (0.37–3.94)	0.71 (0.21–2.35)	1.13 (0.31–4.14)	1.95 (0.53–7.15)	0.52 (0.13–2.02)

The model was adjusted for sex, age, BMI, smoking, education, and family history. Statistically significant results are marked in bold with * (p-value <0.05). OR = odds ratio, CI = confidence interval, IQR = interquartile range.

corresponds to OR (95% CI) per IQR increase of 0.58 (0.37–0.90) for PFOA and 0.56 (0.32–0.95) for PFOS. Similarly, an inverse association was found for contact dermatitis: 0.37 (0.16–0.85) for PFOA and 0.33 (0.11–0.94) for PFOS. Furthermore, a negative association was also observed between pollen, dust, and mite allergy and PFUnDA with 0.62 (0.43–0.89).

During the diagnostics of logistic regression, potential influential outliers were identified in 4 observations. Logistic regression was run again without these potential influential outliers as a sensitivity analysis to see their impact on the analysis result. The sensitivity analysis showed that potential influential outliers did not significantly influence the analysis results (SI, Table S4) and thus were kept in the analysis. As a next part of sensitivity analysis, Poisson regression with robust standard errors was used to calculate RR (SI, Table S5). Results showed the same trend and similar strength of the effect. Nine of the observations were significant. Among others, a significant negative association was observed in atopic eczema and contact dermatitis for both PFOA and PFOS, together with a negative association in pollen dust, and mite allergy for PFUnDA, thus the same observations as were observed in results from logistic regression.

Evidence of the immunomodulatory effect of PFASs has been repeatedly described as an effect of decreased antibody levels after vaccination (Abraham et al., 2020; Grandjean et al., 2017a; Kielsen et al., 2016; Looker et al., 2014; Timmermann et al., 2020), nevertheless the evidence about other immune-mediated health outcomes is not so strong nor consistent. Our results contribute to the body of literature that observes the immunosuppressive effect of PFASs on eczemas and allergies.

The inverse effect of PFAS exposure to eczemas has recently been described. More specifically, PFUnDA measured in maternal blood was negatively associated with ever-diagnosed atopic eczema in 7-year-old children within the MoBa cohort (Impinen et al., 2019). Furthermore, levels of PFUnDA measured in cord blood were negatively associated with both atopic and non-atopic eczema in 1-year-old children (n = 60) within the Melbourne Atopy Cohort (Lowe et al., 2019). Inverse association of PFHxS measured in maternal blood and atopic eczema in children was also observed in the Hokkaido study and the effect remained consistent during 0–2-year-old children (Okada et al., 2014), 4-year-old children (Goudarzi et al., 2016) and 7-year-old children (Ait Bamai et al., 2020). Kvalem et al. (2020) found a negative association between PFHxS blood levels in 10-year-old children and atopic dermatitis in the age of 10–16 years.

Even if many PFASs have been inversely associated with eczema, the inverse association with specifically PFOA and PFOS, which was shown in our study, is not frequently reported in the literature. In consent with our study, maternal PFOS concentrations were negatively associated with eczema in children between 1.5 and 7 years within the INMA birth cohort (Manzano-Salgado et al., 2019). However, there is number of longitudinal studies showing no association between eczemas and exposure to PFOA and/or PFOS assessing both prenatal (Impinen et al., 2018, 2019; Lowe et al., 2019; Okada et al., 2014) and childhood exposure (Averina et al., 2019; Kvalem et al., 2020). Additionally, Kvalem et al. (2020) observed no association between eczemas and exposure to PFOA and/or PFOS also in the cross-sectional design of study (in the 10-year-old children). On the other hand, Wen et al. (2019) observed a positive association between PFOA measured in cord blood and atopic dermatitis in 5-year-old children.

Pollen, dust, and mite allergy is one of the most common allergies and usually manifests as rhinitis or rhinoconjunctivitis (Brożek et al., 2017). These outcomes were associated with PFAS exposure in previous studies, however, the evidence of effect is inconsistent. Specifically, PFNA measured in maternal plasma was negatively associated with allergic rhinoconjunctivitis in 4-year-old children within the Hokkaido study, but this finding was observed in a population with a small number of participants with rhinoconjunctivitis (Goudarzi et al., 2016). Further, serum PFNA concentration was found to be negatively associated with

allergic rhinoconjunctivitis in 13-year-old children in the cross-sectional design of the study, but not in the longitudinal design of the same study (Timmermann et al., 2017). Averina et al. (2019) investigating PFAS exposure in adolescents in The Tromsø study found a negative association of serum PFOS, PFNA, and ΣPFAS concentration with sensitization for inhalant allergens in the longitudinal design of the study, however, this association was not linear and thus considered weak. The same study identified no association with self-reported rhinitis and pollen allergy in the cross-sectional design (around 16-year-old), nor with self-reported rhinitis in the longitudinal design (3 years follow-up) (Averina et al., 2019). Interestingly, a cross-sectional study of US adolescents observed PFOS exposure negatively associated with sensitization to plants, involving pollen allergens (Stein et al., 2016). On the contrary, the same study found a positive association between PFOA exposure and rhinitis, together with no observed association between PFAS exposure and sensitization to dust mites (Stein et al., 2016). Further, Kvalem et al. (2020) reported PFNA and PFHpS exposure at 10 years positively associated with rhinitis at 16 years in girls, whereas no associations were found in cross-sectional design (at age of 10 years). Further, Impinen et al. (2018) did not observe any association between prenatal PFAS exposure and rhinitis in 10-year-old children. Finally, a meta-analysis, that pooled results from 4 studies identified a positive association between PFOA concentration and allergic rhinitis (Luo et al., 2020).

Our study observed a negative association between pollen, dust, and mite allergy and PFUnDA exposure. In a consent with our study, a few weak associations were reported. Specifically, rhinitis in 10-year-old children was negatively associated with cord blood PFUnDA concentrations, but these results were no longer statistically significant after multiple testing correction (Impinen et al., 2018). Similarly, maternal plasma PFUnDA concentrations were negatively associated with morbidities in 4-year-old children: total allergic diseases and rhinoconjunctivitis, the latter only for boys (Goudarzi et al., 2016). Nevertheless, Goudarzi et al. (2016) stated that the results regarding rhinoconjunctivitis must be interpreted with caution as there was a small number of participants with this outcome in the population. Furthermore, the evidence of the effect of PFUnDA exposure is limited because this PFAS has not been studied as extensively as other PFASs.

3.4. Exposure to PFAS mixture and immune-mediated diseases

BKMR modelling was performed for outcomes that were significantly associated with PFASs in the main analysis, i.e., atopic eczema, contact dermatitis, and pollen, dust, and mite allergy. Table 5 shows PIPs corresponding to the estimated importance of the individual PFAS within the mixture towards the particular health outcome. PFOA was shown to be the most influential component of the PFAS mixture in atopic eczema (PIP = 0.44) as well as in contact dermatitis (PIP = 0.52). The most influential exposure in pollen, dust, and mite allergy within the PFAS mixture was PFUnDA (PIP = 0.92). Quantile graphs, representing the overall effect of the mixture, showed decreasing dependencies with increasing concentration of PFAS mixture for all three immune-mediated diseases (SI, Fig. S3), however, none of these three dependencies were statistically significant. Univariate exposure-response functions display the effect of each individual PFAS exposure conditional on the other PFAS exposures in the mixture on atopic eczema (Fig. 1), contact dermatitis (Fig. 2), and pollen dust mite allergy (Fig. 3). Both PFOA and PFOS showed a negative association with both atopic eczema (Fig. 1) and contact dermatitis (Fig. 2). PFUnDA was negatively associated with pollen, dust, and mite allergy, while the PFNA together with PFOA were positively associated with this outcome (Fig. 2).

Table 5
Posterior Inclusion Probabilities (PIPs) for 9 PFASs for atopic eczema, contact dermatitis, and pollen, dust, and mite allergy by BKMR model.

	Atopic eczema	Contact dermatitis	Pollen, dust, and mite allergy
PFPA	0.23	0.39	0.47
PFOA	**0.44***	**0.52***	0.66
PFNA	0.21	0.43	0.71
PFDA	0.17	0.36	0.59
PFUnDA	0.07	0.31	**0.92***
PFBS	0.22	0.23	0.28
PFHxS	0.31	0.37	0.48
PFHpS	0.35	0.25	0.38
PFOS	0.38	0.44	0.60

The model was adjusted for sex, age, BMI, smoking, education, and family history. Observations that were statistically significant in the main analysis (Table 4) are coloured in pale blue. PFASs with the highest PIP for each outcome in the mixture are marked in bold with *.

Results from BMKR modelling showed that the effect of PFAS mixture is driven by the individual PFAS with the highest PIPs. These identified drivers corresponded to the individual PFAS exposures identified in statistically significant observations during the main analysis (Table 4). In the case of atopic eczema and contact dermatitis, PFOA and PFOS as drivers of the mixture effect could be expected as their serum concentrations were the highest when compared to serum concentrations of other PFAS in our study. In the case of pollen dust mite allergy, PFUnDA affected most the effect of the mixture with very high PIP (0.92). This observation is interesting as PFUnDA had one of the lowest medians of PFAS mixture components in the population.

Although the assessment of PFAS exposure as a mixture has high informative value, only a limited number of studies investigating the association of PFAS mixtures and immune-mediated outcomes have been published so far (Ojo et al., 2021). Using BKMR modelling, an increased prevalence of common cold associated with the overall effect of PFAS mixture (4 PFASs) was observed in cross-sectional study of US children, but not in the adolescents (Zhang et al., 2022). Similarly, an incidence of urticaria among adults was found to be positively associated with exposure to a mixture of 8 different PFASs in a nested case-control study (Shen et al., 2022). In contrast to the BKMR approach, where individual contribution toward a studied effect is investigated, several studies assessing PFAS exposure as a mixture exist. For example, the quantile g-computation method revealed association between PFAS mixture (4 PFASs) and increased susceptibility to persistent infections in adolescents and adults in cross-sectional study (Bulka et al., 2021). Based on the principal component analysis, a mixture of chemicals with predominant PFAS content in maternal serum was negatively associated with wheezing in children aged 5–9 years, but not with asthma and eczema (Smit et al., 2015).

3.5. PFAS exposure and immune-mediated diseases stratified by sex

All 5 statistically significant observations identified in the main analysis (Table 4) were tested for the interaction between PFAS and sex. The interaction was statistically significant only for the association of PFOS and atopic eczema (data not shown), and the results of the stratified logistic regression for this association are presented in Table 6. For women, we observed a negative association (OR (95% CI) per IQR increase 0.23 (0.10–0.53)), which was stronger than for the whole population (Table 4). The OR for men indicates a positive tendency, although this finding was not statistically significant.

Based on identified interaction, BKMR modelling was performed for atopic eczema separately for women and men to observe the role of the individual PFAS within the mixture, especially the effect of PFOS. For both, men and women, PFOS had the highest PIP and therefore the biggest influence on the effect of the whole mixture (Table 7). A univariate exposure-response function showed an inverse direction of the PFOS effect on atopic eczema: a negative effect in women (Fig. 4A) and a positive effect in men (Fig. 4B). The inverse effect by sex was shown also in quantile graphs: although these did not provide significant results, we can see a decreasing tendency for women (Fig. 5A) and an increasing tendency for men (Fig. 5B). Interestingly, in the case of men population,

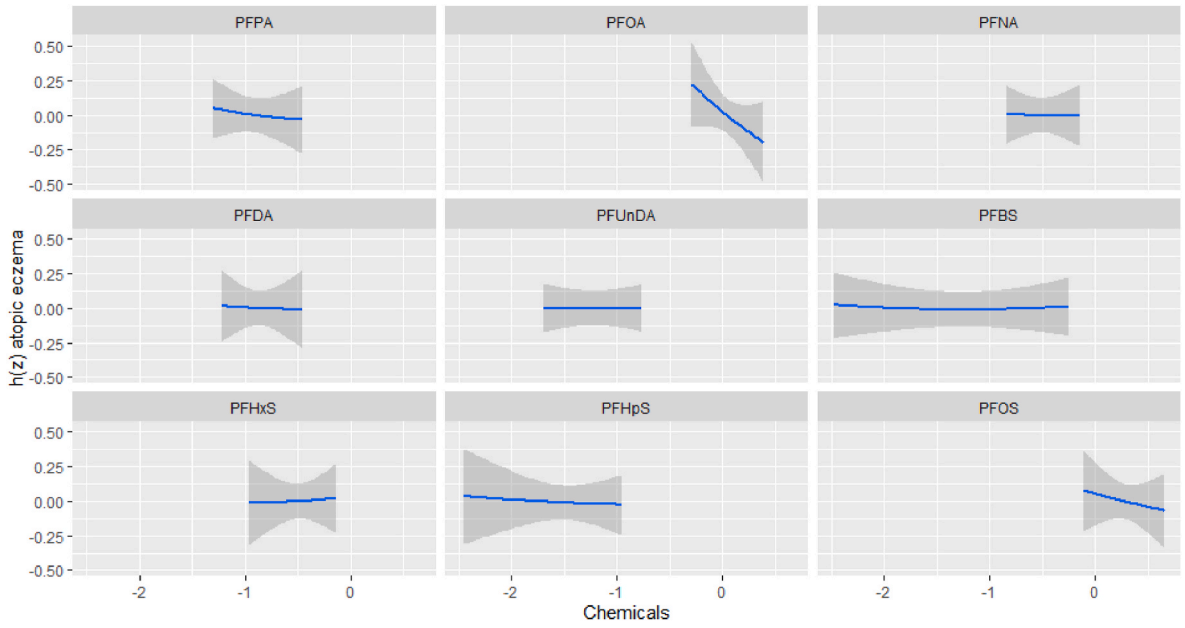

Fig. 1. Univariate exposure-response function with 95% credible intervals for each of 9 PFASs and atopic eczema by BKMR model. Graphs display the effect of the single PFAS exposure conditional on the other PFAS exposures (other PFASs are fixed at their median). The model was adjusted for sex, age, BMI, smoking, education, and family history.

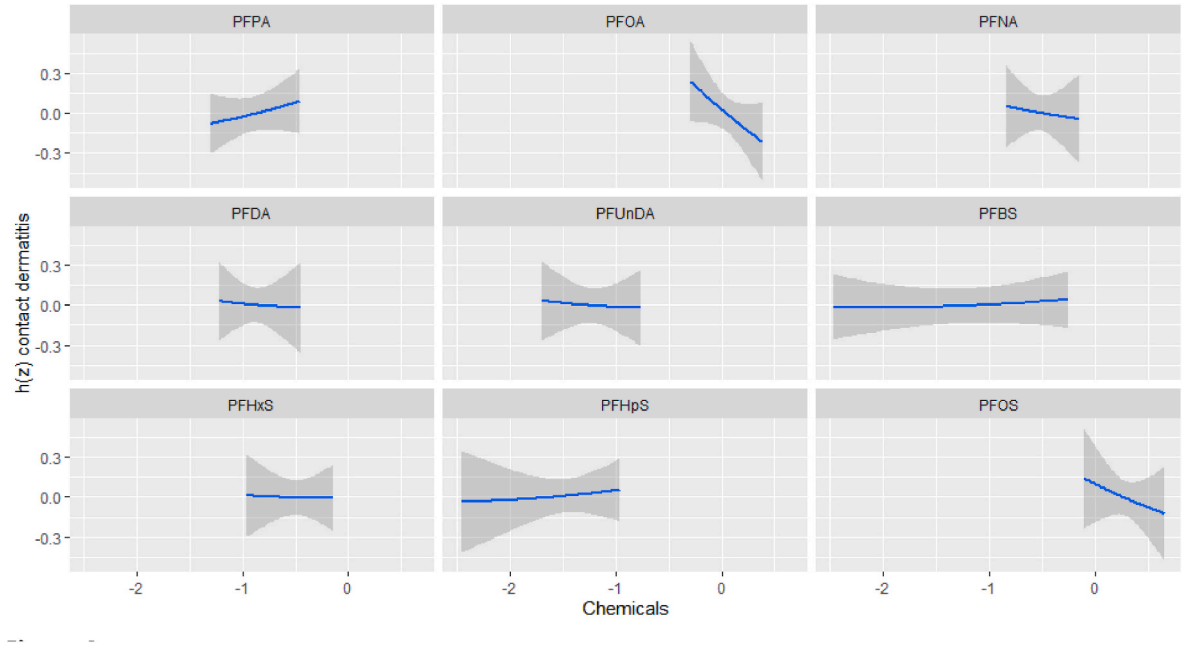

Fig. 2. Univariate exposure-response function with 95% credible intervals for each of 9 PFASs and contact dermatitis by BKMR model. Graphs display the effect of the single PFAS exposure conditional on the other PFAS exposures (other PFASs are fixed at their median). The model was adjusted for sex, age, BMI, smoking, education, and family history.

the strength of PFOS influence (PIP = 0.634) was tightly followed by PFOA (PIP = 0.632) but with the opposite effect than the PFOS had (Fig. 5B). This strong influence of PFOA in the mixture could explain the weak and therefore not significant effect observed by logistic regression analysis (Table 6, column regarding men) as well as the moderate trend in the overall effect in the mixture (Fig. 5B).

Our results showed that sex can play the role of effect modifier in the relationship between PFAS exposure and immune-mediated diseases. Sex-specific interaction of PFASs and immune-mediated health outcomes has been already described in the literature. Some studies observed inverse sex-specific associations in the similar direction as our study showed, i.e., a negative effect for women and a positive effect for men. Specifically, the levels of IgE and concentrations of both PFOS and PFOA in cord blood were positively associated only in boys, not in girls (Wang et al., 2011). In the Hokkaido study, levels of maternal PFOA were associated with decreased IgE levels in cord blood among girls, no association was observed for boys (Okada et al., 2012). In the same study, decreased risk of eczema among girls (2 years old) was associated with PFUnDA and perfluorotridecanoic acid (PFTrDA) levels measured in maternal blood (Okada et al., 2014). The same study observed a

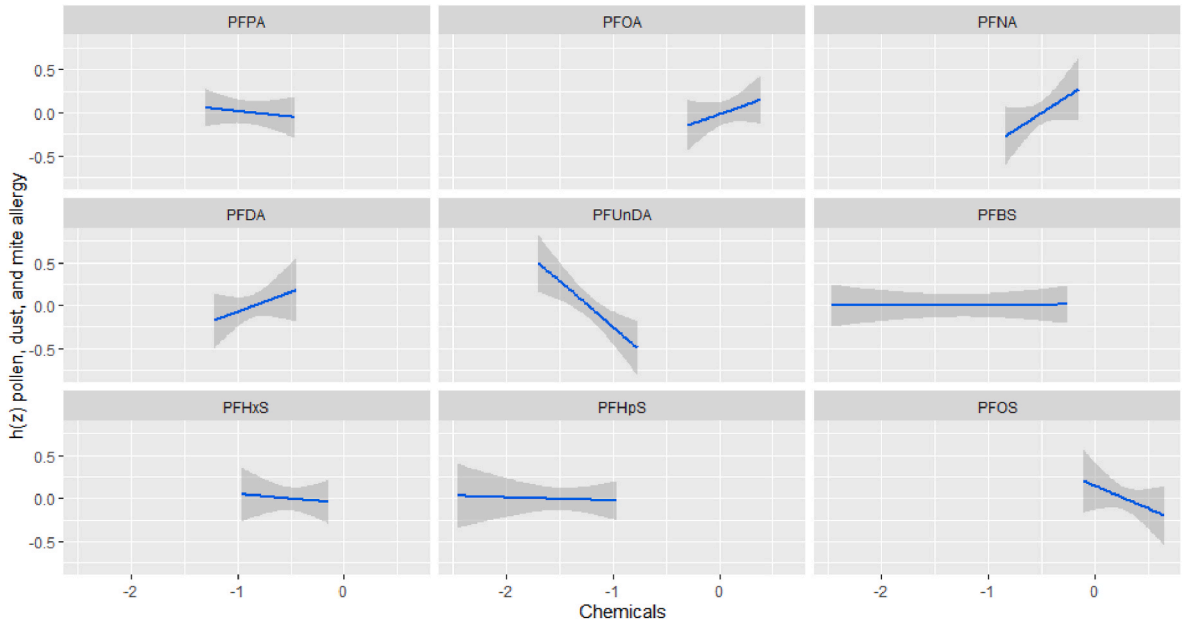

Fig. 3. Univariate exposure-response function with 95% credible intervals for each of 9 PFASs and pollen, dust, and mite allergy by BKMR model. Graphs display the effect of the single PFAS exposure conditional on the other PFAS exposures (other PFASs are fixed at their median). The model was adjusted for sex, age, BMI, smoking, education, and family history.

Table 6
The association between PFOS and atopic eczema stratified by sex; results are expressed as OR (95% CI) per IQR increase.

Women	Men
0.23 (0.10–0.53)	2.15 (0.80–5.76)

The model was adjusted for age, BMI, smoking, education, and family history. Statistically significant results are marked in bold (p-value <0.05). OR = odds ratio, CI = confidence interval, IQR = interquartile range.

Table 7
Posterior Inclusion Probabilities (PIPs) for the mixture of 9 PFASs for atopic eczema separately for women and men by BKMR model.

	Women	Men
PFPA	0.40	0.57
PFOA	0.37	0.63
PFNA	0.40	0.54
PFDA	0.43	0.53
PFUnDA	0.37	0.51
PFBS	0.33	0.43
PFHxS	0.41	0.49
PFHpS	0.36	0.49
PFOS	**0.84***	**0.63***

The model was adjusted for age, BMI, smoking, education, and family history. PFAS with the highest PIP in the mixture is marked with *.

decreased risk of total allergies associated with PFOA, PFNA, PFUnDA, PFDoDA, PFTrDA only for girls, not for boys (Okada et al., 2014). Prenatal exposure to PFUnDA was inversely associated with atopic eczema in 7-year-old girls in the MoBa cohort (Impinen et al., 2019). Kvalem et al. (2020) observed a decreased risk of atopic dermatitis associated with PFNA, PFDA, and PFUnDA levels in 10–16 years old girls and interestingly with PFHxS in boys of the same age. On the contrary, Q. Chen et al., 2018a observed an increased risk of atopic dermatitis in 2 years old girls associated with PFOA and PFNA cord blood concentration, no effect was observed among boys.

Even if the PFAS-related sex-specific differences in the health effects have been frequently reported in the literature, the cause is still unknown. As one of the possible causes can be considered differences in PFAS blood concentrations, that is often observed between men and women populations (Grandjean et al., 2020; Kvalem et al., 2020; Nyström et al., 2022). Another possible cause can be an interplay between PFAS and sex hormones, as a more substantial health effect was observed in individuals with higher estradiol levels (Zhoug et al., 2016). Nevertheless, probably also other sex-specific differences in metabolism and detoxification processes can influence the effect of PFAS exposure. To fully elucidate the mechanism behind the contradictory associations, a detailed mechanistic study with individual chemicals would be needed.

3.6. The putative underlying mechanism linking PFAS exposure and immune-mediated adverse health outcomes

Our results together with evidence from the literature indicate that PFASs have an immunomodulating effect, impacting specifically T-cells. In both atopic eczema and contact dermatitis, T-cells play a crucial role. Contact dermatitis is driven by T cytotoxic cells while T helper (Th) cells also play an important role. Th1 cells contribute to inflammation by the production of cytokines e.g., interferon (IFN)-γ, and are more abundant than Th2 cells, which are suggested to have a regulatory function (Girolomoni et al., 2001). Atopic eczema, on the other hand, is controlled by Th2 cells and is often accompanied by increased production of IgE, therefore humoral response also play an important role in aetiology of this disease (Girolomoni et al., 2001). Similarly, an increased occurrence of allergen-specific Th2 cells accompanied by IgE production is also typical for pollen, dust, and mite allergy (Bousquet et al., 2020).

Several epidemiological studies found evidence for PFAS-mediated Th1/Th2 disbalance reported as increased production of Th2 cytokines (Zhu et al., 2016) and decreased production of Th1 cytokines (Abraham et al., 2020; Zhu et al., 2016). Disbalance of Th1/Th2 towards Th2 increase induced by PFASs has been reported also *in vitro* (Li et al., 2020) as well as in vivo (Zheng et al., 2011; Zhong et al., 2016). However, in vivo experiments showed also the Th1/Th2 disbalance in opposite direction, i.e., an increase of Th1 cytokines and a decrease of Th2 cytokines when exposed to PFASs (De Guise and Levin, 2021).

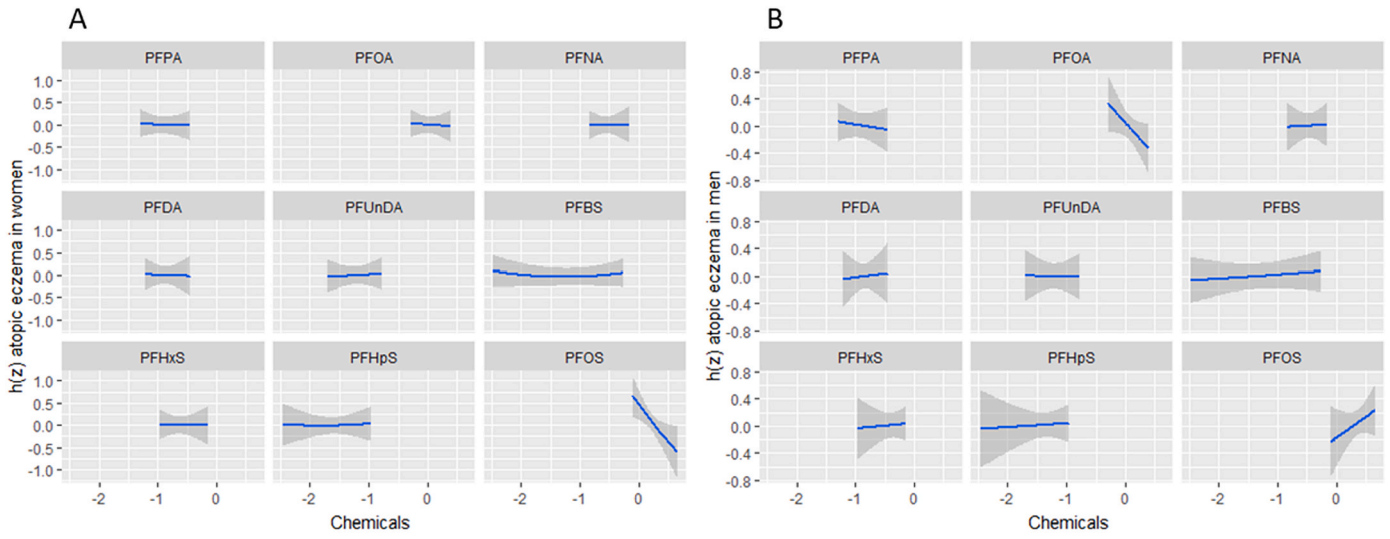

Fig. 4. Univariate exposure-response function with 95% credible intervals for each of 9 PFASs and atopic eczema separately on women (5A) and men (5B) by BKMR model. Graphs display the effect of the single PFAS exposure conditional on the other PFAS exposures (other PFASs are fixed at their median). The model was adjusted for age, BMI, smoking, education, and family history.

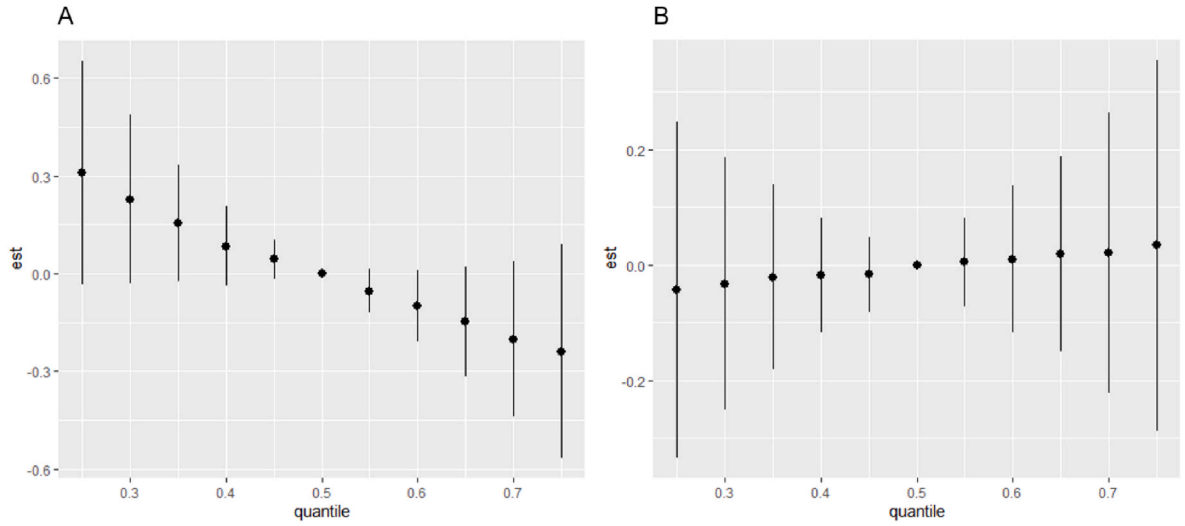

Fig. 5. The overall effect of the PFAS mixture on atopic eczema separately in women (6A) and men (6B) depicted in quantile graphs by BKMR model. The quantile graph shows the estimated change in effect when all the exposures at a particular percentile are compared to when all the exposures are at their 50th percentile. The estimate is accompanied by 95% CI.

Zhang et al. (2021) observed both directions of Th1/Th2 disbalance dependent on the duration of the PFAS exposure (zebrafish). In summary, the available data from the mechanistic studies show that Th1/Th2 balance and downstream signalling can be affected by PFAS exposure. Nevertheless, the underlying mechanism is likely not limited to the Th1/Th2 disbalance. Probably, more specific processes within the immune system (e.g., calcium signalling) as well as other mechanisms within the whole organism (e.g., lipid metabolism, oxidative stress) can be involved in the immunotoxicity of PFAS (Ehrlich et al., 2023). Anyway, the mechanism underlying the PFAS exposure effect on immunity is complex and is still not fully understood. Other important aspect is that PFASs, as a group of chemicals, differ in their length of carbon chain, functional group, and isomer forms. This can naturally influence their properties and subsequently also the apical effect (Sheng et al., 2018). Thus, the effect of each individual PFAS can differ from other PFASs as well as from effect of the whole PFAS mixture. Our findings, together with the published studies support the building of the adverse outcome pathway (AOP) that is a current concept in epidemiology and toxicology (Bajard et al., 2023). Such AOP allows to explain the chain of events leading from exposure through deregulation of adaptive immunity to higher apical adverse health outcomes (Neagu et al., 2021).

3.7. Strengths and limitations

Our study has several strengths. Diseases in our study were reported with the assistance of health practitioners, thus the information about the health condition is more accurate in comparison with e.g., self-reported questionnaires. Moreover, our study contributes to a scarce number of studies researching the impact of PFASs on immune-mediated diseases in adult populations. The present study presents both, odds ratios, and risk ratios to better describe the risk, which is currently not a common practice. Further, BKMR modelling brought novel information about the PFAS mixture effect and the role of its components on immune-mediated diseases. Our study has also several limitations. Our study participants are not a representative sample of the Czech population as the recruitment of participants was based on a volunteer basis.

Therefore, people with higher interest in their health, that are usually of higher education, are involved. Moreover, 309 people is a rather limited sample size. Due to the cross-sectional design of the study, there could be a potential risk of reverse causation. Nevertheless, many prospective studies (Ait Bamai et al., 2020; Q. Chen et al., 2018a; Grandjean et al., 2017a, 2017b; Kvalem et al., 2020; Wang et al., 2011), as well as results from in vivo experiments (De Guise and Levin, 2021; Zhang et al., 2021; Zheng et al., 2011) described immunomodulation potency of PFASs. Thus, reverse causation in our study is less likely. Lastly, as our study is explanatory, we chose not to correct for multiple testing as we want to be as sensitive as possible accepting the risk of false positive findings, nevertheless the FDR-corrected significance levels are available in SI, Table S3, Table S5.

4. Conclusion

Our study investigated the association of serum PFAS concentrations and immune-mediated diseases in the population of adults living in the Czech Republic. Employing methods such as logistic regression and Poisson regression with robust standard errors, we found consistent results of immunomodulatory effects of PFASs, specifically negative association with atopic eczema, contact dermatitis, and pollen, dust, and mite allergy. By employing BKMR we described the overall mixture effect as well as the role of individual substances of the PFAS mixture in immune-mediated diseases and these PFASs were identified to be the main drivers: PFOA, PFOS and PFUnDA. Moreover, we identified a sex-specific association between PFOS and atopic eczema, namely a negative effect in women and a positive effect in men. Our study is contributing to the understanding of the impact of environmental chemicals on human health and represents a solid base for future meta-analysis regarding the impact of PFASs on immune-mediated diseases.

Credit author statement

Barbora Rudzanová: Conceptualization, Methodology, Investigation, Formal analysis, Writing—original draft, Writing—review & editing; **Jelle Vlaanderen:** Conceptualization, Supervision, Writing—review & editing; **Jiří Kalina:** Methodology, Investigation, Formal analysis, Writing—review & editing; **Pavel Piler:** Resources, Data curation, Writing—review & editing; **Martin Zvonař:** Conceptualization, Coordination, Writing—review & editing; **Jana Klánová:** Conceptualization, Coordination, Resources, Writing—review & editing; **Luděk Bláha:** Conceptualization, Supervision, Writing—review & editing; **Ondřej Adamovský:** Conceptualization, Supervision, Writing—review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

The authors do not have permission to share data.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envres.2023.115969.

References

Abraham, K., Mielke, H., Fromme, H., Völkel, W., Menzel, J., Peiser, M., Zepp, F., Willich, S.N., Weikert, C., 2020. Internal exposure to perfluoroalkyl substances (PFASs) and biological markers in 101 healthy 1-year-old children: associations between levels of perfluorooctanoic acid (PFOA) and vaccine response. Arch. Toxicol. 94, 2131–2147. https://doi.org/10.1007/s00204-020-02715-4.

Ait Bamai, Y., Goudarzi, H., Araki, A., Okada, E., Kashino, I., Miyashita, C., Kishi, R., 2020. Effect of prenatal exposure to per- and polyfluoroalkyl substances on childhood allergies and common infectious diseases in children up to age 7 years: the Hokkaido study on environment and children's health. Environ. Int. 143, 105979 https://doi.org/10.1016/j.envint.2020.105979.

Akdis, C.A., 2021. Does the epithelial barrier hypothesis explain the increase in allergy, autoimmunity and other chronic conditions? Nat. Rev. Immunol. 21, 739–751. https://doi.org/10.1038/s41577-021-00538-7.

Alesio, J.L., Slitt, A., Bothun, G.D., 2022. Critical new insights into the binding of poly- and perfluoroalkyl substances (PFAS) to albumin protein. Chemosphere 287, 131979. https://doi.org/10.1016/j.chemosphere.2021.131979.

Averina, M., Brox, J., Huber, S., Furberg, A.-S., Sørensen, M., 2019. Serum perfluoroalkyl substances (PFAS) and risk of asthma and various allergies in adolescents. The Tromsø study Fit Futures in Northern Norway. Environ. Res. 169, 114–121. https://doi.org/10.1016/j.envres.2018.11.005.

Bajard, L., Adamovsky, O., Audouze, K., Baken, K., Barouki, R., Beltman, J.B., Beronius, A., Bonefeld-Jørgensen, E.C., Cano-Sancho, G., de Baat, M.L., Di Tillio, F., Fernández, M.F., FitzGerald, R.E., Gundacker, C., Hernández, A.F., Hilscherova, K., Karakitsios, S., Kuchovska, E., Long, M., Luijten, M., Majid, S., Marx-Stoelting, P., Mustieles, V., Negi, C.K., Sarigiannis, D., Scholz, S., Sovadinova, I., Stierum, R., Tanabe, S., Tollefsen, K.E., van den Brand, A.D., Vogs, C., Wielsøe, M., Wittwehr, C., Blaha, L., 2023. Application of AOPs to assist regulatory assessment of chemical risks – case studies, needs and recommendations. Environ. Res. 217, 114650 https://doi.org/10.1016/j.envres.2022.114650.

Bobb, J.F., 2018. Example using the bkmr R package for probit regression with simulated data [WWW Document]accessed 11.November.22. https://jenfb.github.io/bkmr/ProbitEx.html.

Bobb, J.F., Claus Henn, B., Valeri, L., Coull, B.A., 2018. Statistical software for analyzing the health effects of multiple concurrent exposures via Bayesian kernel machine regression. Environ. Health 17, 67. https://doi.org/10.1186/s12940-018-0413-y.

Bousquet, J., Anto, J.M., Bachert, C., Baiardini, I., Bosnic-Anticevich, S., Walter Canonica, G., Melén, E., Palomares, O., Scadding, G.K., Togias, A., Toppila-Salmi, S., 2020. Allergic rhinitis. Nat. Rev. Dis. Prim. 6, 1–17. https://doi.org/10.1038/s41572-020-00227-0.

Brodin, P., Jojic, V., Gao, T., Bhattacharya, S., Angel, C.J.L., Furman, D., Shen-Orr, S., Dekker, C.L., Swan, G.E., Butte, A.J., Maecker, H.T., Davis, M.M., 2015. Variation in the human immune system is largely driven by non-heritable influences. Cell 160, 37–47. https://doi.org/10.1016/j.cell.2014.12.020.

Brożek, J.L., Bousquet, J., Agache, I., Agarwal, A., Bachert, C., Bosnic-Anticevich, S., Brignardello-Petersen, R., Canonica, G.W., Casale, T., Chavannes, N.H., Correia de Sousa, J., Cruz, A.A., Cuello-Garcia, C.A., Demoly, P., Dykewicz, M., Etxeandia-Ikobaltzeta, I., Florez, I.D., Fokkens, W., Fonseca, J., Hellings, P.W., Klimek, L., Kowalski, S., Kuna, P., Laisaar, K.-T., Larenas-Linnemann, D.E., Lødrup Carlsen, K.C., Manning, P.J., Meltzer, E., Mullol, J., Muraro, A., O'Hehir, R., Ohta, K., Panzner, P., Papadopoulos, N., Park, H.-S., Passalacqua, G., Pawankar, R., Price, D., Riva, J.J., Roldán, Y., Ryan, D., Sadeghirad, B., Samolinski, B., Schmid-Grendelmeier, P., Sheikh, A., Togias, A., Valero, A., Valiulis, A., Valovirta, E., Ventresca, M., Wallace, D., Waserman, S., Wickman, M., Wiercioch, W., Yepes-Nuñez, J.J., Zhang, L., Zhang, Y., Zidarn, M., Zuberbier, T., Schünemann, H.J., 2017. Allergic rhinitis and its impact on asthma (ARIA) guidelines-2016 revision. J. Allergy Clin. Immunol. 140, 950–958. https://doi.org/10.1016/j.jaci.2017.03.050.

Bulka, C.M., Avula, V., Fry, R.C., 2021. Associations of exposure to perfluoroalkyl substances individually and in mixtures with persistent infections: recent findings from NHANES 1999-2016. Environ. Pollut. 275, 116619 https://doi.org/10.1016/j.envpol.2021.116619.

Buser, M.C., Scinicariello, F., 2016. Perfluoroalkyl substances and food allergies in adolescents. Environ. Int. 88, 74–79. https://doi.org/10.1016/j.envint.2015.12.020.

Buuren, S. van, Groothuis-Oudshoorn, K., 2011. Mice: multivariate imputation by chained equations in R. J. Stat. Software 45, 1–67. https://doi.org/10.18637/jss.v045.i03.

Catelan, D., Biggeri, A., Russo, F., Gregori, D., Pitter, G., Da Re, F., Fletcher, T., Canova, C., 2021. Exposure to perfluoroalkyl substances and mortality for COVID-19: a spatial ecological analysis in the veneto region (Italy). Int. J. Environ. Res. Publ. Health 18. https://doi.org/10.3390/ijerph18052734.

Chen, Q., Huang, R., Hua, L., Guo, Y., Huang, L., Zhao, Y., Wang, X., Zhang, J., 2018a. Prenatal exposure to perfluoroalkyl and polyfluoroalkyl substances and childhood atopic dermatitis: a prospective birth cohort study. Environ. Health 17, 8. https://doi.org/10.1186/s12940-018-0352-7.

Chen, W., Qian, L., Shi, J., Franklin, M., 2018b. Comparing performance between log-binomial and robust Poisson regression models for estimating risk ratios under model misspecification. BMC Med. Res. Methodol. 18, 63. https://doi.org/10.1186/s12874-018-0519-5.

Corthay, A., 2014. Does the immune system naturally protect against cancer? Front. Immunol. 5.

Dalsager, L., Christensen, N., Halekoh, U., Timmermann, C.A.G., Nielsen, F., Kyhl, H.B., Husby, S., Grandjean, P., Jensen, T.K., Andersen, H.R., 2021. Exposure to perfluoroalkyl substances during fetal life and hospitalization for infectious disease in childhood: a study among 1,503 children from the Odense Child Cohort. Environ. Int. 149, 106395 https://doi.org/10.1016/j.envint.2021.106395.

De Guise, S., Levin, M., 2021. Suppression of Th2 cytokines as a potential mechanism for reduced antibody response following PFOA exposure in female B6C3F1 mice. Toxicol. Lett. 351, 155–162. https://doi.org/10.1016/j.toxlet.2021.09.002.

Dean, W.S., Adejumo, H.A., Caiati, A., Garay, P.M., Harmata, A.S., Li, L., Rodriguez, E.E., Sundar, S., 2020. A framework for regulation of new and existing PFAS by EPA. Policy Anal 16, 14.

Delves, P.J., Roitt, I.M., 2000. The immune system. N. Engl. J. Med. 343, 37–49. https://doi.org/10.1056/NEJM200007063430107.

Dong, G.-H., Tung, K.-Y., Tsai, C.-H., Liu, M.-M., Wang, D., Liu, W., Jin, Y.-H., Hsieh, W.-S., Lee, Y.L., Chen, P.-C., 2013. Serum polyfluoroalkyl concentrations, asthma outcomes, and immunological markers in a case-control study of Taiwanese children. Environ. Health Perspect. 121, 507–513. https://doi.org/10.1289/ehp.1205351.

Ehrlich, V., Bil, W., Vandebriel, R., Granum, B., Luijten, M., Lindeman, B., Grandjean, P., Kaiser, A.-M., Hauzenberger, I., Hartmann, C., Gundacker, C., Uhl, M., 2023. Consideration of pathways for immunotoxicity of per- and polyfluoroalkyl substances (PFAS). Environ. Health 22, 19. https://doi.org/10.1186/s12940-022-00958-5.

EU HBM Dashboard, 2023. HBM4EU accessed 1.September.23. https://www.hbm4eu.eu/what-we-do/european-hbm-platform/eu-hbm-dashboard/. accessed 1. September.23.

Eurostat, 2022. International standard classification of education (ISCED) [WWW Document]accessed 11.November.22. https://ec.europa.eu/eurostat/statistics-explained/index.php?title=International_Standard_Classification_of_Education_(ISCED.

Fenton, S.E., Ducatman, A., Boobis, A., DeWitt, J.C., Lau, C., Ng, C., Smith, J.S., Roberts, S.M., 2021. Per- and polyfluoroalkyl substance toxicity and human health review: current state of knowledge and strategies for informing future research. Environ. Toxicol. Chem. 40, 606–630. https://doi.org/10.1002/etc.4890.

Forsthuber, M., Kaiser, A.M., Granitzer, S., Hassl, I., Hengstschläger, M., Stangl, H., Gundacker, C., 2020. Albumin is the major carrier protein for PFOS, PFOA, PFHxS, PFNA and PFDA in human plasma. Environ. Int. 137, 105324 https://doi.org/10.1016/j.envint.2019.105324.

Girolomoni, G., Sebastiani, S., Albanesi, C., Cavani, A., 2001. T-cell subpopulations in the development of atopic and contact allergy. Curr. Opin. Immunol. 13, 733–737. https://doi.org/10.1016/S0952-7915(01)00287-4.

Goudarzi, H., Miyashita, C., Okada, E., Kashino, I., Kobayashi, S., Chen, C.-J., Ito, S., Araki, A., Matsuura, H., Ito, Y.M., Kishi, R., 2016. Effects of prenatal exposure to perfluoroalkyl acids on prevalence ofallergic diseases among 4-year-old children. Environ. Int. 94, 124–132. https://doi.org/10.1016/j.envint.2016.05.020.

Grandjean, P., Heilmann, C., Weihe, P., Nielsen, F., Mogensen, U.B., Budtz-Jørgensen, E., 2017a. Serum vaccine antibody concentrations in adolescents exposed to perfluorinated compounds. Environ. Health Perspect. 125, 077018 https://doi.org/10.1289/EHP275.

Grandjean, P., Heilmann, C., Weihe, P., Nielsen, F., Mogensen, U.B., Timmermann, A., Budtz-Jørgensen, E., 2017b. Estimated exposures to perfluorinated compounds in infancy predict attenuated vaccine antibody concentrations at age 5-years. J. Immunot. 14, 188–195. https://doi.org/10.1080/1547691X.2017.1360968.

Grandjean, P., Timmermann, C.A.G., Kruse, M., Nielsen, F., Vinholt, P.J., Boding, L., Heilmann, C., Mølbak, K., 2020. Severity of COVID-19 at elevated exposure to perfluorinated alkylates. PLoS One 15, e0244815. https://doi.org/10.1371/journal.pone.0244815.

Granum, B., Haug, L.S., Namork, E., Stølevik, S.B., Thomsen, C., Aaberge, I.S., van Loveren, H., Løvik, M., Nygaard, U.C., 2013. Pre-natal exposure to perfluoroalkyl substances may be associated with altered vaccine antibody levels and immune-related health outcomes in early childhood. J. Immunot. 10, 373–379. https://doi.org/10.3109/1547691X.2012.755580.

Impinen, A., Nygaard, U.C., Lødrup Carlsen, K.C., Mowinckel, P., Carlsen, K.H., Haug, L. S., Granum, B., 2018. Prenatal exposure to perfluoralkyl substances (PFASs) associated with respiratory tract infections but not allergy- and asthma-related health outcomes in childhood. Environ. Res. 160, 518–523. https://doi.org/10.1016/j.envres.2017.10.012.

Impinen, A., Longnecker, M.P., Nygaard, U.C., London, S.J., Ferguson, K.K., Haug, L.S., Granum, B., 2019. Maternal levels of perfluoroalkyl substances (PFASs) during pregnancy and childhood allergy and asthma related outcomes and infections in the Norwegian Mother and Child (MoBa) cohort. Environ. Int. 124, 462–472. https://doi.org/10.1016/j.envint.2018.12.041.

Jian, J.-M., Chen, D., Han, F.-J., Guo, Y., Zeng, L., Lu, X., Wang, F., 2018. A short review on human exposure to and tissue distribution of per- and polyfluoroalkyl substances (PFASs). Sci. Total Environ. 636, 1058–1069. https://doi.org/10.1016/j.scitotenv.2018.04.380.

Jørgensen, T.S.H., Osler, M., Ängquist, L.H., Zimmermann, E., Christensen, G.T., Sørensen, T.I.A., 2016. The U-shaped association of body mass index with mortality: influence of the traits height, intelligence, and education. Obesity 24, 2240–2247. https://doi.org/10.1002/oby.21615.

Kielsen, K., Shamim, Z., Ryder, L.P., Nielsen, F., Grandjean, P., Budtz-Jørgensen, E., Heilmann, C., 2016. Antibody response to booster vaccination with tetanus and diphtheria in adults exposed to perfluorinated alkylates. J. Immunot. 13, 270–273. https://doi.org/10.3109/1547691X.2015.1067259.

Knol, M.J., Le Cessie, S., Algra, A., Vandenbroucke, J.P., Groenwold, R.H.H., 2012. Overestimation of risk ratios by odds ratios in trials and cohort studies: alternatives to logistic regression. CMAJ Can. Med. Assoc. J. 184, 895–899. https://doi.org/10.1503/cmaj.101715.

Kvalem, H.E., Nygaard, U.C., Lødrup Carlsen, K.C., Carlsen, K.H., Haug, L.S., Granum, B., 2020. Perfluoroalkyl substances, airways infections, allergy and asthma related health outcomes – implications of gender, exposure period and study design. Environ. Int. 134, 105259 https://doi.org/10.1016/j.envint.2019.105259.

Kyle, Steenland, Zhao, Liping, Andrea, Winquist, Parks, Christine, 2013. Ulcerative colitis and perfluorooctanoic acid (PFOA) in a highly exposed population of community residents and workers in the mid-Ohio valley. Environ. Health Perspect. 121, 900–905. https://doi.org/10.1289/ehp.1206449.

Leter, G., Consales, C., Eleuteri, P., Uccelli, R., Specht, I.O., Toft, G., Moccia, T., Budillon, A., Jönsson, B.A.G., Lindh, C.H., Giwercman, A., Pedersen, H.S., Ludwicki, J.K., Zviezdai, V., Heederik, D., Bonde, J.P.E., Spanò, M., 2014. Exposure to perfluoroalkyl substances and sperm DNA global methylation in arctic and European populations. Environ. Mol. Mutagen. 55, 591–600. https://doi.org/10.1002/em.21874.

Li, R., Guo, C., Lin, X., Chan, T.F., Lai, K.P., Chen, J., 2020. Integrative omics analyses uncover the mechanism underlying the immunotoxicity of perfluorooctanesulfonate in human lymphocytes. Chemosphere 256, 127062. https://doi.org/10.1016/j.chemosphere.2020.127062.

Looker, C., Luster, M.I., Calafat, A.M., Johnson, V.J., Burleson, G.R., Burleson, F.G., Fletcher, T., 2014. Influenza vaccine response in adults exposed to perfluorooctanoate and perfluorooctanesulfonate. Toxicol. Sci. Off. J. Soc. Toxicol. 138, 76–88. https://doi.org/10.1093/toxsci/kft269.

Lorber, M., Eaglesham, G.E., Hobson, P., Toms, L.-M.L., Mueller, J.F., Thompson, J.S., 2015. The effect of ongoing blood loss on human serum concentrations of perfluorinated acids. Chemosphere 118, 170–177. https://doi.org/10.1016/j.chemosphere.2014.07.093.

Lowe, A.J., Dharmage, S.C., Abramson, M.J., Vijayasarathy, S., Erbas, B., Mueller, J.F., Lodge, C.J., 2019. Cord-serum per- and poly-fluoroalkyl substances and atopy and eczema at 12-months. Allergy 74, 812–815. https://doi.org/10.1111/all.13669.

Luo, Y., Deji, Z., Huang, Z., 2020. Exposure to perfluoroalkyl substances and allergic outcomes in children: a systematic review and meta-analysis. Environ. Res. 191, 110145 https://doi.org/10.1016/j.envres.2020.110145.

Manzano-Salgado, C.B., Granum, B., Lopez-Espinosa, M.-J., Ballester, F., Iñiguez, C., Gascón, M., Martínez, D., Guxens, M., Basterretxea, M., Zabaleta, C., Schettgen, T., Sunyer, J., Vrijheid, M., Casas, M., 2019. Prenatal exposure to perfluoroalkyl substances, immune-related outcomes, and lung function in children from a Spanish birth cohort study. Int. J. Hyg Environ. Health 222, 945–954. https://doi.org/10.1016/j.ijheh.2019.06.005.

Mattiuzzi, C., Lippi, G., 2020. Worldwide asthma epidemiology: insights from the global health data exchange database. Int. Forum Allergy Rhinol. 10, 75–80. https://doi.org/10.1002/alr.22464.

Neagu, M., Constantin, C., Bardi, G., Duraes, L., 2021. Adverse outcome pathway in immunotoxicity of perfluoroalkyls. Curr. Opin. Toxicol. 25, 23–29. https://doi.org/10.1016/j.cotox.2021.02.001.

Nyström, J., Benskin, J.P., Plassmann, M., Sandblom, O., Glynn, A., Lampa, E., Gyllenhammar, I., Moraeus, L., Lignell, S., 2022. Demographic, life-style and physiological determinants of serum per- and polyfluoroalkyl substance (PFAS) concentrations in a national cross-sectional survey of Swedish adolescents. Environ. Res. 208, 112674 https://doi.org/10.1016/j.envres.2022.112674.

Ojo, A.F., Peng, C., Ng, J.C., 2021. Assessing the human health risks of per- and polyfluoroalkyl substances: a need for greater focus on their interactions as mixtures. J. Hazard Mater. 407, 124863 https://doi.org/10.1016/j.jhazmat.2020.124863.

Okada, E., Sasaki, S., Saijo, Y., Washino, N., Miyashita, C., Kobayashi, S., Konishi, K., Ito, Y.M., Ito, R., Nakata, A., Iwasaki, Y., Saito, K., Nakazawa, H., Kishi, R., 2012. Prenatal exposure to perfluorinated chemicals and relationship with allergies and infectious diseases in infants. Environ. Res. 112, 118–125. https://doi.org/10.1016/j.envres.2011.10.003.

Okada, E., Sasaki, S., Kashino, I., Matsuura, H., Miyashita, C., Kobayashi, S., Itoh, K., Ikeno, T., Tamakoshi, A., Kishi, R., 2014. Prenatal exposure to perfluoroalkyl acids and allergic diseases in early childhood. Environ. Int. 65, 127–134. https://doi.org/10.1016/j.envint.2014.01.007.

Piler, P., Kandrnal, V., Kukla, L., Andrýsková, L., Švancara, J., Jarkovský, J., Dušek, L., Pikhart, H., Bobák, M., Klánová, J., 2017. Cohort profile: the European longitudinal study of pregnancy and childhood (ELSPAC) in the Czech republic. Int. J. Epidemiol. 46, 1379. https://doi.org/10.1093/ije/dyw091, 1379f.

Ranganathan, P., Aggarwal, R., Pramesh, C.S., 2015. Common pitfalls in statistical analysis: odds versus risk. Perspect. Clin. Res. 6, 222. https://doi.org/10.4103/2229-3485.167092.

Shen, M., Xiao, Y., Huang, Y., Jing, D., Su, J., Luo, D., Duan, Y., Xiao, S., Li, J., Chen, X., 2022. Perfluoroalkyl substances are linked to incident chronic spontaneous urticaria: a nested case-control study. Chemosphere 287, 132358. https://doi.org/10.1016/j.chemosphere.2021.132358.

Sheng, N., Cui, R., Wang, Jinghua, Guo, Y., Wang, Jianshe, Dai, J., 2018. Cytotoxicity of novel fluorinated alternatives to long-chain perfluoroalkyl substances to human liver cell line and their binding capacity to human liver fatty acid binding protein. Arch. Toxicol. 92, 359–369. https://doi.org/10.1007/s00204-017-2055-1.

Shih, Y.-H., Blomberg, A.J., Bind, M.-A., Holm, D., Nielsen, F., Heilmann, C., Weihe, P., Grandjean, P., 2021. Serum vaccine antibody concentrations in adults exposed to

per- and polyfluoroalkyl substances: a birth cohort in the Faroe Islands. J. Immunot. 18, 85–92. https://doi.org/10.1080/1547691X.2021.1922957.

Signorell, A., Aho, K., Alfons, A., Anderegg, N., Aragon, T., Arachchige, C., Arppe, A., Baddeley, A., Barton, K., Bolker, B., Borchers, H.W., Caeiro, F., Champely, S., Chessel, D., Chhay, L., Cooper, N., Cummins, C., Dewey, M., Doran, H.C., Dray, S., Dupont, C., Eddelbuettel, D., Ekstrom, C., Elff, M., Enos, J., Farebrother, R.W., Fox, J., Francois, R., Friendly, M., Galili, T., Gamer, M., Gastwirth, J.L., Gegzna, V., Gel, Y.R., Graber, S., Gross, J., Grothendieck, G., Jr, F.E.H., Heiberger, R., Hoehle, M., Hoffmann, C.W., Hojsgaard, S., Hothorn, T., Huerzeler, M., Hui, W.W., Hurd, P., Hyndman, R.J., Jackson, C., Kohl, M., Korpela, M., Kuhn, M., Labes, D., Leisch, F., Lemon, J., Li, D., Maechler, M., Magnusson, A., Mainwaring, B., Malter, D., Marsaglia, G., Marsaglia, J., Matei, A., Meyer, D., Miao, W., Millo, G., Min, Y., Mitchell, D., Mueller, F., Naepflin, M., Navarro, D., Nilsson, H., Nordhausen, K., Ogle, D., Ooi, H., Parsons, N., Pavoine, S., Plate, T., Prendergast, L., Rapold, R., Revelle, W., Rinker, T., Ripley, B.D., Rodriguez, C., Russell, N., Sabbe, N., Scherer, R., Seshan, V.E., Smithson, M., Snow, G., Soetaert, K., Stahel, W.A., Stephenson, A., Stevenson, M., Stubner, R., Templ, M., Lang, D.T., Therneau, T., Tille, Y., Torgo, L., Trapletti, A., Ulrich, J., Ushey, K., VanDerWal, J., Venables, B., Verzani, J., Iglesias, P.J.V., Warnes, G.R., Wellek, S., Wickham, H., Wilcox, R.R., Wolf, P., Wollschlaeger, D., Wood, J., Wu, Y., Yee, T., Zeileis, A., 2022. DescTools: Tools for Descriptive Statistics.

Smit, L.a.M., Lenters, V., Høyer, B.B., Lindh, C.H., Pedersen, H.S., Liermontova, I., Jönsson, B.a.G., Piersma, A.H., Bonde, J.P., Toft, G., Vermeulen, R., Heederik, D., 2015. Prenatal exposure to environmental chemical contaminants and asthma and eczema in school-age children. Allergy 70, 653–660. https://doi.org/10.1111/all.12605.

Steenland, K., Zhao, L., Winquist, A., 2015. A cohort incidence study of workers exposed to perfluorooctanoic acid (PFOA). Occup. Environ. Med. 72, 373–380. https://doi.org/10.1136/oemed-2014-102364.

Stein, C.R., McGovern, K.J., Pajak, A.M., Maglione, P.J., Wolff, M.S., 2016. Perfluoroalkyl and polyfluoroalkyl substances and indicators of immune function in children aged 12–19 y: National Health and Nutrition Examination Survey. Pediatr. Res. 79, 348–357. https://doi.org/10.1038/pr.2015.213.

Textor, J., van der Zander, B., Gilthorpe, M.S., Liśkiewicz, M., Ellison, G.T., 2016. Robust causal inference using directed acyclic graphs: the R package 'dagitty. Int. J. Epidemiol. 45, 1887–1894. https://doi.org/10.1093/ije/dyw341.

Timmermann, C.A.G., Budtz-Jørgensen, E., Kold Jensen, T., Osuna, C.E., Skaalum Petersen, M., Steuerwald, U., Nielsen, F., Poulsen, L.K., Weihe, P., Grandjean, P., 2017. Association between perfluoroalkyl substance exposure and asthma and allergic disease in children as modified by MMR vaccination. J. Immunot. 14, 39–49. https://doi.org/10.1080/1547691X.2016.1254306.

Timmermann, C.A.G., Jensen, K.J., Nielsen, F., Budtz-Jørgensen, E., van der Klis, F., Benn, C.S., Grandjean, P., Fisker, A.B., 2020. Serum perfluoroalkyl substances, vaccine responses, and morbidity in a cohort of Guinea-bissau children. Environ. Health Perspect. 128 https://doi.org/10.1289/EHP6517.

Verreault, J., Houde, M., Gabrielsen, G.W., Berger, U., Haukås, M., Letcher, R.J., Muir, D. C.G., 2005. Perfluorinated alkyl substances in plasma, liver, brain, and eggs of glaucous gulls (Larus hyperboreus) from the Norwegian arctic. Environ. Sci. Technol. 39, 7439–7445. https://doi.org/10.1021/es051097y.

von Holst, H., Nayak, P., Dembek, Z., Buehler, S., Echeverria, D., Fallacara, D., John, L., 2021. Perfluoroalkyl substances exposure and immunity, allergic response, infection, and asthma in children: review of epidemiologic studies. Heliyon 7, e08160. https://doi.org/10.1016/j.heliyon.2021.e08160.

Wang, I.-J., Hsieh, W.-S., Chen, C.-Y., Fletcher, T., Lien, G.-W., Chiang, H.-L., Chiang, C.-F., Wu, T.-N., Chen, P.-C., 2011. The effect of prenatal perfluorinated chemicals exposures on pediatric atopy. Environ. Res. 111, 785–791. https://doi.org/10.1016/j.envres.2011.04.006.

Wang, L., Wang, F.-S., Gershwin, M.E., 2015. Human autoimmune diseases: a comprehensive update. J. Intern. Med. 278, 369–395. https://doi.org/10.1111/joim.12395.

Wang, Z., Shi, R., Ding, G., Yao, Q., Pan, C., Gao, Y., Tian, Y., 2022. Association between maternal serum concentration of perfluoroalkyl substances (PFASs) at delivery and acute infectious diseases in infancy. Chemosphere 289, 133235. https://doi.org/10.1016/j.chemosphere.2021.133235.

Weir, C.B., Jan, A., 2022. BMI Classification Percentile and Cut Off Points. StatPearls Publishing.

Wen, H.-J., Wang, S.-L., Chuang, Y.-C., Chen, P.-C., Guo, Y.L., 2019. Prenatal perfluorooctanoic acid exposure is associated with early onset atopic dermatitis in 5-year-old children. Chemosphere 231, 25–31. https://doi.org/10.1016/j.chemosphere.2019.05.100.

Wheatley, L.M., Togias, A., 2015. Allergic Rhinitis. N. Engl. J. Med. 372, 456–463. https://doi.org/10.1056/NEJMcp1412282.

Winans, B., Humble, M.C., Lawrence, B.P., 2011. Environmental toxicants and the developing immune system: a missing link in the global battle against infectious disease? Reprod. Toxicol., Prenatal Programming and Toxicity II (PPTOX II). Role of Environ. Stressors Dev. Origins of Disease 31, 327–336. https://doi.org/10.1016/j.reprotox.2010.09.004.

Wong, F., MacLeod, M., Mueller, J.F., Cousins, I.T., 2014. Enhanced elimination of perfluorooctane sulfonic acid by menstruating women: evidence from population-based pharmacokinetic modeling. Environ. Sci. Technol. 48, 8807–8814. https://doi.org/10.1021/es500796y.

World Allergy Organization, 2013. WAO White Book on Allergy: Update 2013. World Allergy Organization, Milwaukee.

Zhang, H., Shen, L., Fang, W., Zhang, X., Zhong, Y., 2021. Perfluorooctanoic acid-induced immunotoxicity via NF-kappa B pathway in zebrafish (Danio rerio) kidney. Fish Shellfish Immunol. 113, 9–19. https://doi.org/10.1016/j.fsi.2021.03.004.

Zhang, Y., Mustieles, V., Sun, Y., Oulhote, Y., Wang, Y.-X., Messerlian, C., 2022. Association between serum per- and polyfluoroalkyl substances concentrations and common cold among children and adolescents in the United States. Environ. Int. 164, 107239 https://doi.org/10.1016/j.envint.2022.107239.

Zheng, L., Dong, G.-H., Zhang, Y.-H., Liang, Z.-F., Jin, Y.-H., He, Q.-C., 2011. Type 1 and Type 2 cytokines imbalance in adult male C57BL/6 mice following a 7-day oral exposure to perfluorooctanesulfonate (PFOS). J. Immunot. 8, 30–38. https://doi.org/10.3109/1547691X.2010.537287.

Zhong, S.-Q., Chen, Z.-X., Kong, M.-L., Xie, Y.-Q., Zhou, Y., Qin, X.-D., Paul, G., Zeng, X.-W., Dong, G.-H., 2016. Testosterone-mediated endocrine function and TH1/TH2 cytokine balance after prenatal exposure to perfluorooctane sulfonate: by sex status. Int. J. Mol. Sci. 17, 1509. https://doi.org/10.3390/ijms17091509.

Zhu, Y., Qin, X.-D., Zeng, X.-W., Paul, G., Morawska, L., Su, M.-W., Tsai, C.-H., Wang, S.-Q., Lee, Y.L., Dong, G.-H., 2016. Associations of serum perfluoroalkyl acid levels with T-helper cell-specific cytokines in children: by gender and asthma status. Sci. Total Environ. 559, 166–173. https://doi.org/10.1016/j.scitotenv.2016.03.187.

Zou, G., 2004. A modified Poisson regression approach to prospective studies with binary data. Am. J. Epidemiol. 159, 702–706. https://doi.org/10.1093/aje/kwh090.