Received: 20 December 2017 | Accepted: 25 February 2018

DOI: 10.1111/ina.12458

ORIGINAL ARTICLE

WILEY

Acute health effects of desktop 3D printing (fused deposition modeling) using acrylonitrile butadiene styrene and polylactic acid materials: An experimental exposure study in human volunteers

I. Gümperlein[1] | E. Fischer[1] | G. Dietrich-Gümperlein[1] | S. Karrasch[1,2,3] | D. Nowak[1,3] | R. A. Jörres[1,3] | R. Schierl[1]

[1]Institute and Outpatient Clinic for Occupational, Social and Environmental Medicine, Inner City Clinic, University Hospital of Munich, Ludwig-Maximilians-Universität, Munich, Germany

[2]Institute of Epidemiology I, Helmholtz Zentrum München - German Research Center for Environmental Health, Neuherberg, Germany

[3]Comprehensive Pneumology Center Munich (CPC-M), Member of the German Center for Lung Research, Munich/Neuherberg, Germany

Correspondence
Ina Gümperlein, Institute and Outpatient Clinic for Occupational, Social and Environmental Medicine, Inner City Clinic, University Hospital of Munich, Ludwig-Maximilians-Universität, Munich, Germany.
Email: ina.guemperlein@med.uni-muenchen.de

Abstract

3D printers are increasingly run at home. Nanoparticle emissions from those printers have been reported, which raises the question whether adverse health effects from ultrafine particles (UFP) can be elicited by 3D printers. We exposed 26 healthy adults in a single-blinded, randomized, cross-over design to emissions of a desktop 3D printer using fused deposition modeling (FDM) for 1 hour (high UFP-emitting acrylonitrile butadiene styrene [ABS] vs low-emitting polylactic acid [PLA]). Before and after exposures, cytokines (IL-1β, IL-6, TNF-α, INF-γ) and ECP in nasal secretions, exhaled nitric oxide (FeNO), urinary 8-isoprostaglandin $F_{2\alpha}$ (8-iso $PGF_{2\alpha}$), and self-reported symptoms were assessed. The exposures had no significant differential effect on 8-iso $PGF_{2\alpha}$ and nasal biomarkers. However, there was a difference ($P < .05$) in the time course of FeNO, with higher levels after ABS exposure. Moreover, indisposition and odor nuisance were increased for ABS exposure. These data suggest that 1 hour of exposure to 3D printer emissions had no acute effect on inflammatory markers in nasal secretions and urine. The slight relative increase in FeNO after ABS printing compared to PLA might be due to eosinophilic inflammation from inhaled UFP particles. This possibility should be investigated in further studies using additional biomarkers and longer observation periods.

KEYWORDS

3D printer emissions, exposure study, indoor air, nanoparticles, oxidative stress, ultrafine particles

1 | BACKGROUND

Desktop 3D printers are getting more popular for both professional purposes and personal use.[1,2] They are advertised even for children as a means for creating toys,[3,4] although known to produce ultrafine particles (UFP).[5-7] For laser printer devices, potential health risks from UFP have already been extensively discussed, and the available studies are ranging from cell culture experiments[8,9] to human exposures.[10,11] The UFP produced by 3D printers are also generated from volatile organic compounds (VOCs) as primary emissions of these printers,[6,7,12-15] but with large differences between the types of plastic filaments used.[15-18] Therefore, in principle 3D printers also could pose a health risk, especially if they are used for personal purposes in the absence of professional protection measures. This is underlined by data showing the toxicity of 3D printed parts in sensitive biological testing systems.[19,20] As it is well recognized that cell

culture and animal experiments are of limited relevance to assess the health risk for human subjects, despite efforts to adapt them to real exposures,[21] exposure experiments in human subjects are indispensable. Although they have limitations, particularly with regard to the duration of exposure, they are informative to detect acute effects and to estimate potential long-term effects from short-term alterations.

We therefore performed a controlled exposure experiment with 26 young, healthy volunteers by comparing two materials that are in widespread use in 3D printing, namely acrylonitrile butadiene styrene (ABS) and polylactic acid (PLA). These compounds are also of interest since previous measurements had demonstrated a large difference regarding the amount of nanoparticles produced with common 3D printing devices.[5,14,16-18,22] These two materials are easily available[23] and can be used in most 3D printers for personal use. As outcome measures, we chose a panel of examinations covering biochemical responses from nasal secretions, exhaled air and urine, the subjects' symptoms and well-being, as well as spirometry after exposures of 1-hour duration. In order to maximize the statistical power, the study was designed as a randomized, cross-over protocol, with assessments prior to, immediately and 2-3 hours after exposures.

Practical Implications

- This study investigated acute effects of desktop 3D printer emissions. Subjects were exposed to emissions during ABS (high UFP-emitter) and PLA (low UFP-emitter) printing. Several sensitive biochemical measures were analyzed before and after the exposure. While most of the parameters did not show a significant relative change between exposures, exhaled NO and self-reported odor nuisance increased after ABS exposure.

TABLE 1 Characteristics of the study population

Characteristics	Female	Male
N	13	13
Age (years)	25.8 (±3.6)	25.0 (±3.2)
BMI (kg m^{-2})	21.6 (±2.3)	24.2 (±2.8)
ABS is first exposure	6	7

Mean values (±SD).

2 | MATERIALS AND METHODS

2.1 | Study group

We recruited 26 young (18-31 years) volunteers, most of them university students, by direct contact. All subjects were non-smokers and anamnestic healthy, including normal lung function. None of them reported a perennial allergy, and those reporting a seasonal allergy were studied outside their season. The characteristics of the participants are given in Table 1. The study was approved by the local Ethics Committee, and all subjects gave their written informed consent.

2.2 | Study protocol

The study was realized within a single-blinded, randomized, cross-over design, with assessments before and after exposures. The single-blinded design was chosen due to technical requirements in operating the equipment, but all measures were taken to ensure that the subjects were not aware of the type of exposure and the interaction between operator and subject was minimal during exposure.

The patient information was enclosed to a screening email which was send to each prospect (n = 49). All potential participants were invited for a screening visit (n = 36) at which the inclusion and exclusion criteria were assessed (n = 27). Additionally, a questionnaire regarding chemical sensitivity was administered (chemical part from Bailer et al[24]). Moreover, all procedures except the symptom questionnaire were performed by the subjects, with the aim to make them familiar with the assessments and to improve the quality of measurements at the exposure days. The values obtained were also used for comparison with the baseline values at the exposure days. The procedures included spirometry, the determination of exhaled nitric oxide (FeNO) and exhaled carbon monoxide (CO), and the sampling of nasal secretions and urine. The CO values were also used to verify the non-smoking status, using a cutoff value of 5 ppb.[25] The cutoff value used for FeNO was 50 ppb as this is indicative of acute allergic airway inflammation or allergy.[26-28]

Persons with lung function outside the normal limits (n = 2) or elevated values of FeNO (n = 4) were excluded, as well as subjects in whom the assertion of non-smoking was not confirmed by measurements of exhaled CO (n = 3).

The screening visit was followed by two exposure visits (n = 26), which were separated by an interval of 5-7 days and always started at 10 AM in order to minimize the potential effect of circadian variations. Assessments were performed prior to exposures, as well as immediately afterward and 2-3 hours after termination of the exposures. The details are given in Table 2. It should be noted that nasal secretions were obtained only before and 2-3 hours after exposure. The reasons were twofold: Firstly, the assessment is likely to affect subsequent samples that are taken within short time, due to irritation of the mucosa; secondly, biochemical responses due to cell activation and/or influx are likely to need some time to occur. Moreover, exhaled CO was measured only at the beginning, as it mainly served to exclude previous exposures, particularly from smoking or passive smoking. FeNO was assessed at all time points in order to detect both immediate and delayed responses. This measurement is very unlikely to exert effects on subsequent or other measurements.

TABLE 2 Order of assessments on exposure days

Time point	Parameter
Before exposures (10 AM)	Urine sample
	CO in exhaled breath
	Nasal secretion
	FeNO
	Spirometry
1-h exposure	
Immediately after exposure	Questionnaire
	FeNO
	Spirometry
	Urine sample
2-3 h after termination of exposure	Nasal secretion
	FeNO
	Urine sample

Spirometry was performed only before and immediately after the exposures, since any effects that could be reasonably expected would be acute effects. Measurements were done after the determination of FeNO, to avoid potential effects of forced expiration on FeNO. Symptom questionnaires were administered only once immediately after exposures, since the questions were related to the perception of the exposures. Moreover, to avoid potential effects from the observer in this single-blinded study, the questionnaire was shown, while the study subjects were still in the exposure chamber.

2.3 | Exposure

2.3.1 | Setup and 3D printing

The study took place in an exposure chamber of volume 32 m^3 that is in use for occupational and environmental exposures.[10,11] The air conditioning system was turned off during exposures in order to avoid a clearance of particles and to ensure a realistic setting, as most 3D printers are operated at home without ventilation, because of unwanted warping of the printed object if exposed to a temperature gradient. Only one volunteer was studied per exposure. Participants were sitting at a defined position in front of the printer, with their face about 40 cm from the printing head (Figure 1). During exposure, the participants were reading, studying, or watched the printer.

All experiments were carried out using a commercially available desktop 3D printer (Ultimaker 2, Ultimaker B.V., the Netherlands). This printer, which uses the fused deposition modeling (FDM) technique, is capable of using several materials, including ABS and PLA. Additionally, it is equipped with a heated printing bed, which might result in more uniform and reproducible emissions. The two materials which we compared are in widespread use and thus suitable to reflect real-world exposure conditions for FDM printing. They were produced by the same company (Formfutura BV, Netherlands, purchased from www.3dmensionals.de). Specifically, we used black ABS (EasyFil, diameter 2.85 mm, 750 g Premium Filament) and black PLA (EasyFil, diameter 2.85 mm, 750 g Premium Filament). All printer settings (eg, extruder temperature, heat bed temperature) were chosen as recommended for the printer and the filaments by the manufacturers. In order to standardize the exposures and to make them as realistic as possible, the printed object was of a size and complexity that it could be printed within 1 hour of exposure. The pattern chosen[29] is shown in Figure S4.

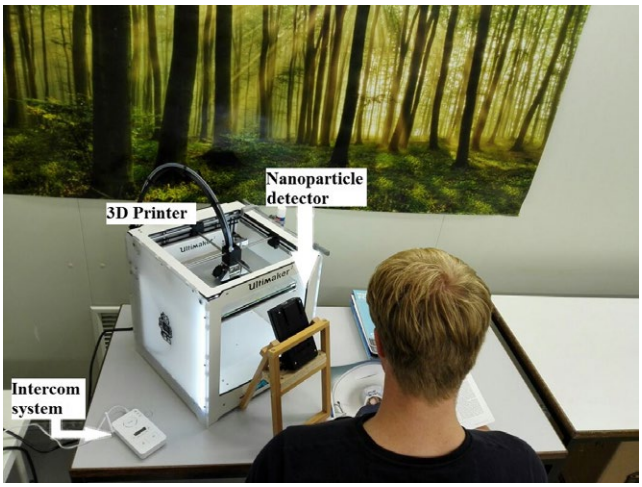

FIGURE 1 Study setup with subject

2.3.2 | Monitoring of ultrafine particles (UFP)

Prior to the start of the study, the UFP emissions of the 3D printer were characterized in detail. These assessments confirmed the finding that the filament material is a key determinant of UFP emission.[5,13] Three types of devices were used for this purpose: a condensation particle counter (CPC, model 3007, TSI Inc., USA), an engine exhaust particle sizer (EEPS, model 3090, TSI), and a partector (naneos particle solutions GmbH, Switzerland). These devices recorded particle number concentration (cm^{-3}), particle number size distribution, and lung-deposited surface area (LDSA) ($\mu m^2\ cm^{-3}$). When ABS and PLA filaments were characterized using these devices, it turned out that they were correlated to high degree and that the partector, as a handheld nanoparticle detector, was sufficient to reliably monitor the emissions during each exposure. Moreover, the device had the advantages of small size, low noise and no need for liquid supply, thereby minimizing potential disturbances of the participants through particle monitoring. Larger particles ($PM_{2.5}$ and PM_{10}) were monitored using an aerosol spectrometer (Model 1.108, GRIMM Aerosol Technik GmbH &Co. KG, Germany).

2.4 | Assessments

2.4.1 | Spirometry and exhaled biomarkers

Spirometry was performed following established recommendations[30] to determine forced expiratory volume in 1-second (FEV_1)

and forced vital capacity (FVC). Exhaled carbon monoxide (CO) was measured using a portable device according to the manufacturer's instructions (carbon monoxide monitor, BreathCO, Vitalograph Ltd, England). The fractional level of exhaled nitric oxide (FeNO) at a flow rate of 50 mL/s was recorded according to guidelines from the American Thoracic Society (ATS) and the European Respiratory Society (ERS)[31] using a chemiluminescence nitric oxide analyzer (NOA 280, Sievers Instruments Inc., USA) and a custom-made flow-control device.

2.4.2 | Nasal secretions

Samples of nasal secretion were obtained by the cotton wool method as described elsewhere.[32,33] Briefly, a small roll of cotton wool was gently inserted through each nostril and placed into the middle meatus. After 15 minutes, the wool was removed and immediately centrifuged for 10 minutes at 4°C. The obtained material was stored at −20°C until measurement. The following cytokines were determined by immunoassay-based microfluidic platform Ella using Simple Plex assays (both from Protein Simple, USA) and following the recommendations of the manufacturer: interleukins IL-1β and IL-6, as well as tumor necrosis factor alpha (TNF-α) and γ-interferon (IFN-γ). The limits of detection (LOD) were 0.064, 0.260, 0.278, and 0.490 pg/mL, respectively.

The level of eosinophil cationic protein (ECP) in nasal secretions was measured via enzyme-linked immunosorbent assay (ELISA) using the ECP Kit (ImmunoCAP® ECP) and the corresponding analyzing device (UniCAP®100; both from Phadia AB, Sweden) as recommended by the manufacturer. ECP analysis was performed only in samples with sufficient volume (50 μL) after the measurement of cytokines (n = 80).

2.4.3 | Urine samples

Urine samples were analyzed for 8-iso $PGF_{2\alpha}$, an established marker of oxidative stress,[34-37] via gas chromatography with tandem mass spectrometer (GC-MS/MS). The detailed measurement procedure can be found in the supporting information S1.

2.4.4 | Questionnaires

Prior to the experiments, the participants answered eight questions of the Chemical and General Environmental Sensitivity (CGES) questionnaire[24] (see supporting information S3). Immediately after exposures and still in the exposure chamber, the participants answered a standardized questionnaire covering a spectrum of symptoms and the perception of the exposure. For all items, a visual analog scale (VAS) ranging from 0 to 10 cm was used; depending on the type of question, the limits were labeled "not at all" and "very strong," or "very bad," and "very good." Assessed symptoms comprised dry cough, itchy/scratchy throat, difficulty swallowing, phlegmy/wet cough, wheezing/whistling sounds while breathing, chest tightness, shortage of breath, urge to sneeze, runny nose, nasal congestion (stuffy nose), burning sensation in nose, itchy nose, headache, feeling of dizziness, cardiac/circulation problems, nausea, burning sensation in the eyes, dry eyes, tired eyes, itchy eyes, itchy skin, skin rash/irritation. The assessment of overall experience covered the following questions: How was your overall well-being in the chamber? How strongly did you perceive the smell in the chamber? How strongly were you bothered by the smell in the chamber? How strongly were you bothered by the printing activity in the chamber overall?

2.5 | Separate assessment of VOC emission

In order to compare VOC emissions from the filaments used in the present study to observations reported in the literature,[6,7,12-14] an additional, semi-quantitative analysis outside the exposure chamber was performed in which the filaments were heated in a laboratory setting. The setup and method for heating and air sampling are available in the supporting information S2.1.

2.6 | Data analysis

For descriptive purposes, mean values and standard deviations (SD), geometric mean values and geometric SD (as a factor) or median values and quartiles are reported, depending on the distribution of the data. The values of spirometric parameters and logarithmically transformed FeNO were approximately normally distributed. Correspondingly, the paired *t* test and repeated-measures analysis of variance (ANOVA) were used to assess potential differences as well as differences in time course after exposures (interaction terms). For the parameters of nasal secretions and urine, the values before and after exposures were compared by the Wilcoxon matched-pairs signed-ranks test, since the data were not normally distributed. The same test was used for symptom data. To compare the two exposures as well as the cytokine responses with each other, repeated-measures ANOVA was employed. For this purpose, logarithmically transformed cytokine values were used, which were approximately normally distributed. Statistical significance was assumed for *P* values <.05. *P* values are given explicitly as far as possible, and no correction for multiple testing was applied. Statistical analysis was performed using the software SPSS Statistics (IBM Corporation, USA). For data analysis and graphics, the software Origin (OriginLab Corporation, USA) was used.

3 | RESULTS

3.1 | Exposures

In two cases of ABS printing, the printed object lost attachment to the printing bed; therefore, it had to be removed and the printing had to be started again but the total exposure time of 1 hour was maintained. The exposure levels in terms of LDSA for all exposures are shown in Figure 2. The initial peak with ABS printing (Figure 3) occurred in all exposures, whereby the two outliers (indicated in Figure 2) resulted in additional high emissions. When excluding

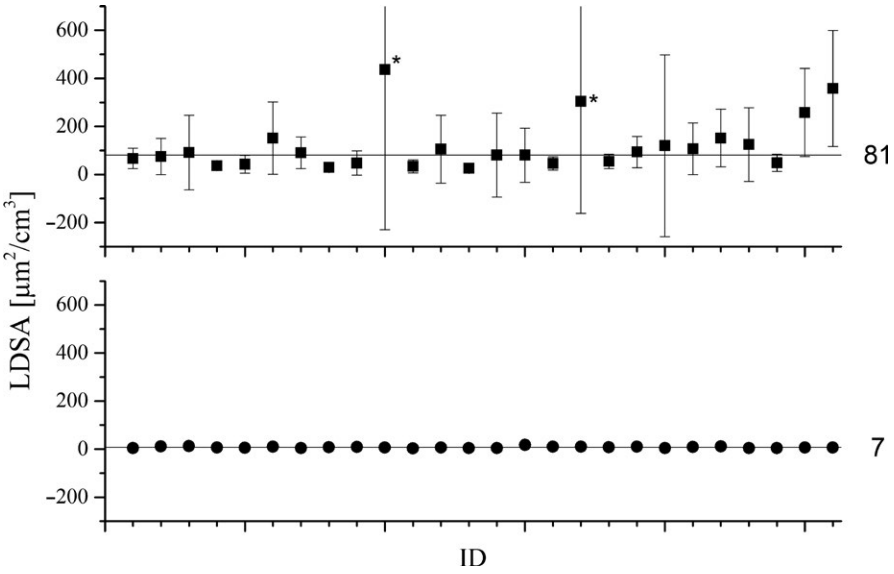

FIGURE 2 Average lung-deposited surface area (LDSA) ($\mu m^2\ cm^{-3}$) values over 1 h print. Squares: acrylonitrile butadiene styrene (ABS); circles: polylactic acid (PLA). Asterisks mark the two exposures with the printing failure, which were excluded in the analysis of the emissions

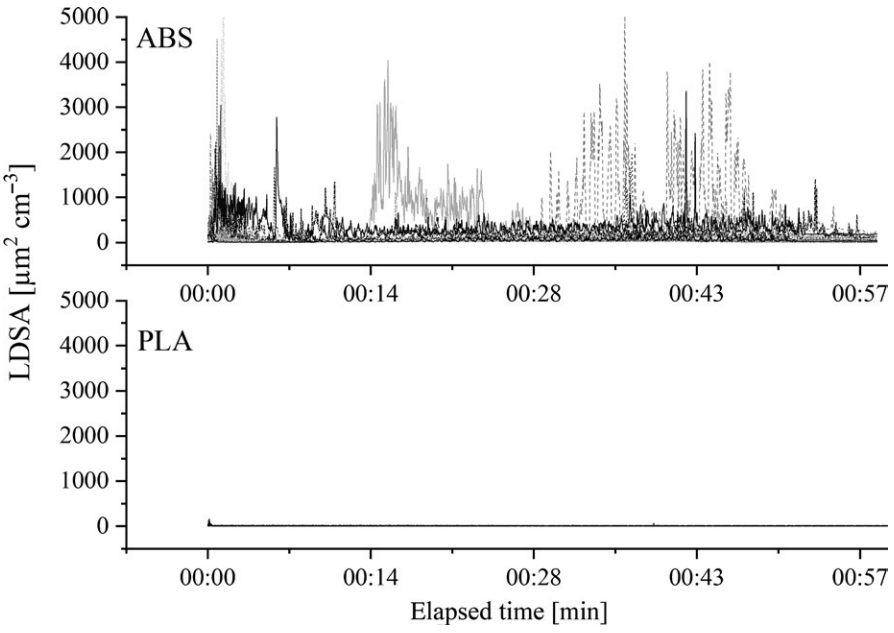

FIGURE 3 Particle emissions during acrylonitrile butadiene styrene (ABS) and polylactic acid (PLA) exposures, respectively. Each shade represents the exposure of one subject. The intermediate peaks in the upper panel correspond to the two printing failures, which occurred with ABS printing (see text)

these two exposures, the median ([25th; 75th percentiles]; min; max) values of LDSA ($\mu m^2\ cm^{-3}$) were 81.0 ([47.1; 113]; 25.7; 358) for ABS and 7.2 ([4.8; 10]; 2.9; 17) for PLA.

In the experiments used for the setup of the protocol, we also had measured the size distribution and particle number concentration for printing with both types of filaments. The numbers for PLA were negligible compared to background values, whereas those for ABS were well measurable. The changes of the size distribution over 1 hour for a representative ABS printing are shown in Figure 4 the corresponding mean UFP number concentration was 1 600 000 cm^{-3}. The concentrations of fine particulate matter were close to the background levels of ($PM_{2.5}$) or indistinguishable from background levels (PM_{10}); therefore, these values are not shown.

3.2 | Spirometry and exhaled biomarkers

Neither FEV_1 nor FVC showed statistically significant changes when comparing the values determined before and after exposures. The mean values are shown in Table 3. Exhaled CO was lower than 5 ppm in all participants, indicating not relevant prior exposure to cigarette smoke or heavy traffic.

Baseline FeNO values prior to the exposures were 18.2 ÷ 1.7 ppb for ABS and 18.0 ÷ 1.7 ppb for PLA (geometric mean ÷ geometric SD factor). One participant consistently showed FeNO values above 50 ppb, although at the screening visit, FeNO was below 50 ppb. However, excluding this participant from the analysis did not change the pattern of statistical significance therefore the participant was kept in the analysis. The parametric analyses of FeNO

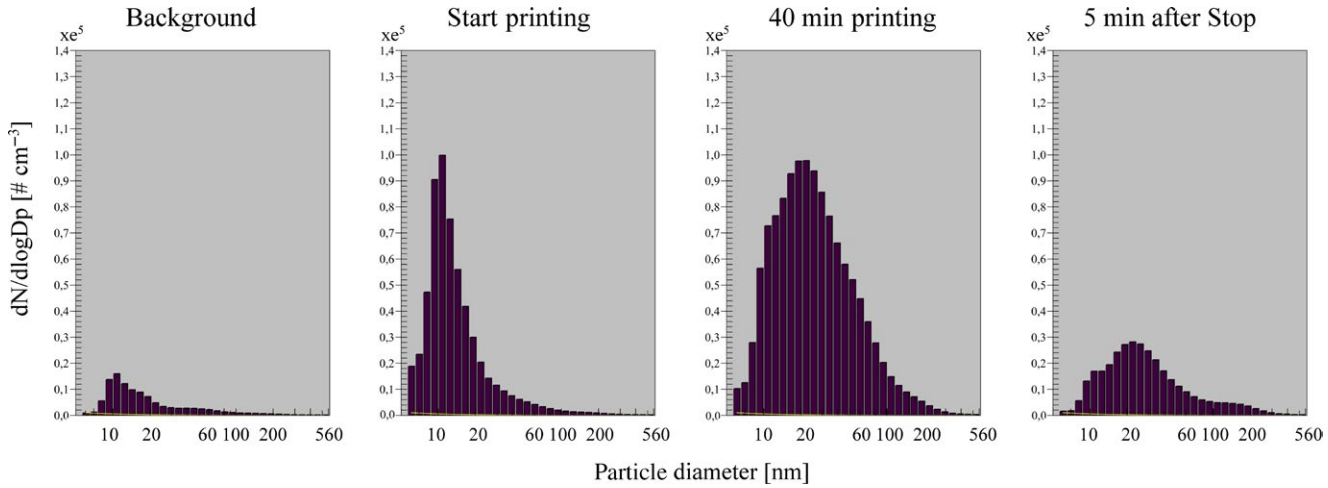

FIGURE 4 Size distribution at different times of printing using acrylonitrile butadiene styrene (ABS) and the object used in the exposure study. The data were obtained with an EEPS (TSI GmbH, Germany)

were performed using log values, because FeNO showed an approximately log-normal distribution.

The time course (Table 3 and Figure 5) was analyzed by repeated-measures ANOVA, using three time points and two exposures as categories. Overall, there was a borderline significant difference between time points ($P = .057$) and a borderline significant interaction between time and exposure ($P = .084$) in the linear model. When using a polynomial (up to quadratic terms) to describe the overall time course, the changes over time followed an overall quadratic (inverse parabolic) pattern ($P = .016$), with an additional linear term ($P = .028$) accounting for the different time course of ABS and PLA exposure (see Figure 5). Alternatively, when performing the comparisons in terms of pairwise contrasts, the difference between ABS and PLA observed 2-3 hours after exposures was significant compared to the baseline difference.

3.3 | Cytokines and ECP in nasal secretion

Median values and quartiles of the cytokine levels assessed in nasal secretions are shown in Table 3. For IL-1β, IL-6, and TNF-α, all values were within the respective calibration ranges of the assay used, whereas for IFN-γ, most values ranged below. Despite this, values for IFN-γ were computed and used in the data analysis. Pairwise comparisons (Wilcoxon test) did not indicate significant changes for IFN-γ after one of the two exposures. In contrast, there were significant changes for IL-1β, Il-6, and TNF-α after ABS exposure and for IL-6 after PLA exposure (Table 3).

3.3.1 | Comparison between exposures

These results raised the question whether these changes occurred in parallel. Therefore, we again used repeated-measures ANOVA after logarithmic transformation of values which led to approximate normal distributions. There were no significant differences for IFN-γ. While the values of IL-1β significantly ($P = .005$) increased over time, there was no significant difference (interaction) between exposures ($P = .588$). A similar pattern was observed for IL-6 ($P < .001$ and $P = .942$) and for TNF-α ($P = .006$ and $P = .299$).

These results indicated significant increases over time, which were, however, in parallel for ABS and PLA. The next question was whether the factors of increase were the same across cytokines in order to differentiate between (possibly different) active secretions and mere (homogeneous) dilutions due to the repeated sampling. Therefore, a repeated-measures ANOVA was performed again, using the pre-post differences of log-transformed values, with the two exposures and the four cytokines as categories. There was no significant difference between the two exposures ($P = .541$), but a highly significant ($P < .001$) difference between cytokines. The increases for IL-6 were higher than those of the other cytokines ($P < .001$ each) by a factor of about 1.6 or more (Figure 6). This differential response pointed toward a particularly strong response to repeated sampling for IL-6.

3.3.2 | Correlation between cytokines

To further elucidate the cytokine response, we performed a correlation analysis. In both exposures, the responses of IFN-γ and TNF-α were correlated with each other ($P \leq .01$ each). Similarly, the responses of IL-1β and IL-6 were correlated ($P < .001$ each). This correlation pattern was confirmed in a tentative factor analysis which we performed despite the fact that data from only 26 subjects were available. The result showed that the responses of IL-1β and IL-6 were correlated within each of the exposures, whereas the responses of IFN-γ and TNF-α were correlated not only within, but also across exposures. This could indicate a difference in responses between ABS and PLA despite the fact that the mean responses were similar and in parallel.

3.3.3 | Eosinophilic cationic protein

Median values and quartiles of ECP levels in nasal secretions are shown in Table 3. Samples from 17 participants were available for

TABLE 3 Results obtained before, immediately, and 2-3 h after exposures. Mean values (±SD), geometric mean (÷ geometric SD), medians (25th, 75th quartiles), and *P* values (Wilcoxon) are shown

Mean (±SD) geometric mean (÷geometric SD factor) median (25th; 75th percentiles)	ABS				PLA			
	Pre	Immediately after	2-3 h after	*P* value	Pre	Immediately after	2-3 h after	*P* value
FEV_1 [L]	4.20 (±0.82)	4.20 (±0.81)			4.23 (±0.83)	4.20 (±0.81)		
FVC [L]	5.20 (±1.0)	5.11 (±1.0)			5.17 (±1.0)	5.14 (±1.0)		
FeNO [ppb]	18.2 (÷1.73)	19.1 (÷1.74)	18.7 (÷1.74)		18.0 (÷1.71)	18.5 (÷1.72)	17.4 (÷1.67)	
Urine								
8-iso $PGF_{2\alpha}$ [ng/L]	232 (÷2.40)	137 (÷ 2.33)	174 (÷1.88)		327 (÷2.06)	146 (÷2.28)	190 (÷2.36)	
8-iso $PGF_{2\alpha}$ [ng/g creat]	209 (÷2.00)	257 (÷ 2.15)	262 (÷1.82)		235 (÷1.94)	302 (÷1.94)	271 (÷2.06)	
Nasal secretion								
IFN-γ [pg/mL]	0.63 (0.44; 1.1)		0.75 (0.47; 1.3)	.310	0.71 (0.53; 0.86)		0.72 (0.47; 1.2)	.162
IL-1β [pg/mL]	15.8 (7.92; 46.7)		24.1 (17.5; 99.1)	.049	20.8 (9.70; 49.4)		32.5 (19.0; 6.0)	.269
IL-6 [pg/mL]	38.9 (12.5; 74.0)		68.0 (39.3; 150)	.000	27.7 (17.9;74.3)		89.6 (46.6; 127)	.000
TNF-α [pg/mL]	6.8 (5.8; 8.8)		7.8 (6.5; 13)	.012	7.0 (5.9; 12)		9.6 (7.5; 12)	.055
ECP [µg/L]	19.7 (1.00; 47.7)		25.4 (14.8; 134)	.015	18.4 (1.00; 40.1)		38.7 (14.8; 105)	.006

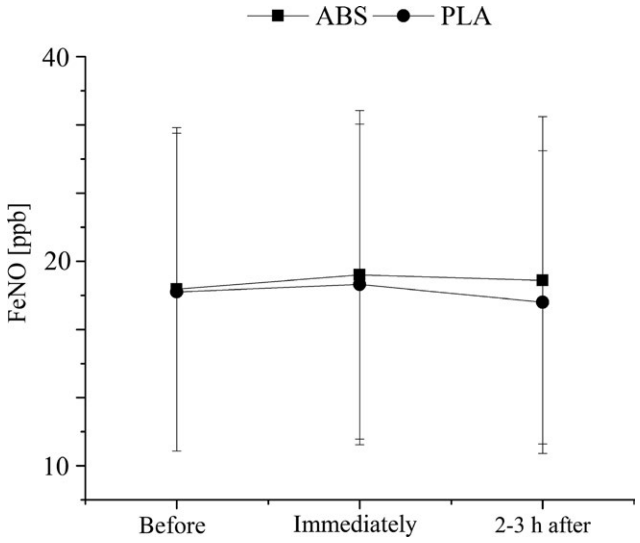

FIGURE 5 Mean values (±SD) of logarithmically transformed FeNO levels at the three time points

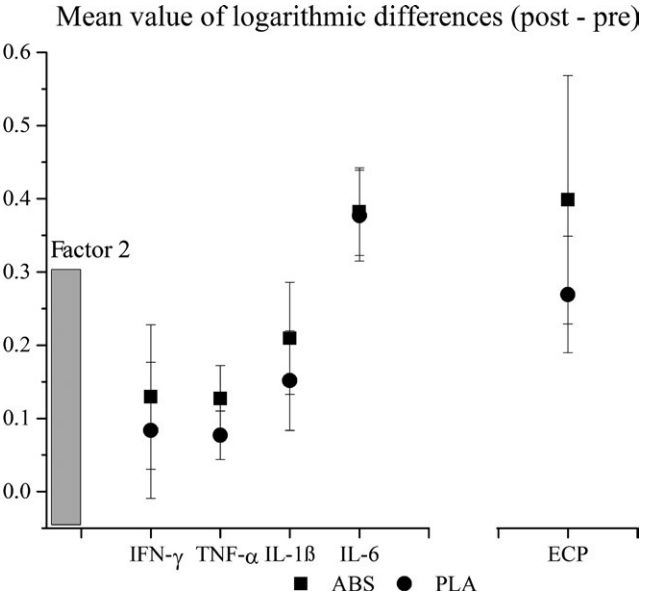

FIGURE 6 Mean values of logarithmic differences (post-pre) with standard error of the mean (±SEM) as measure of its precision are shown for the four cytokines (IFN-γ, TNF-α, IL-1β, and IL-6) and for eosinophil cationic protein (ECP). The size of logarithmic change corresponding to factor 2 is indicated. Samples are of 26 and 17 participants for cytokines and ECP, respectively

this assessment before and after both exposures. ECP levels before exposures were not significantly different from each other, as well as the two values assessed 2-3 hours after exposures. There were, however, significant increases after both exposures ($P < .05$ each) but without significant difference between these changes. Overall, ECP levels were correlated with FeNO (Figure 7), as underlined by a significant regression coefficient in an analysis of covariance ($P < .001$).

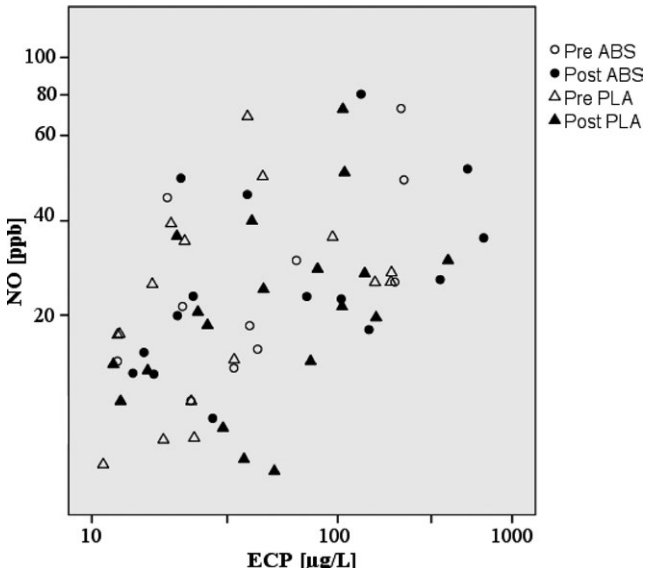

FIGURE 7 Relationship between FeNO and eosinophil cationic protein (ECP) levels. Measurements in which the ECP levels where below the detection limit (<2 µg/L) are omitted

3.4 | Urine biomarker of oxidative stress

In the analysis of urinary isoprostanes, it is common to adjust for creatinine levels, although the use of creatinine for this purpose has been questioned.[34] Indeed, the creatinine levels of the subjects varied widely between before and after exposure values. Twenty-five of the 182 urine samples showed a creatinine level out of the accepted range (30-250 mg/dL). However, no fundamental changes in associations were observed when excluding them from the analysis. Therefore, the analysis of urinary isoprostane concentration was performed with and without adjustment for creatinine levels (Table 3) for all subjects. When analyzing logarithmically transformed 8-iso $PGF_{2\alpha}$ values by repeated-measures ANOVA, there was no significant difference between exposures (P = .093), but there was a significant (P = .003) overall change over time (Figure 8). When using 8-iso $PGF_{2\alpha}$ values normalized for creatinine, there were no significant overall differences between exposures (P = .188) and also no significant changes over time (P = .154). These data did not indicate differential effects of exposures irrespective of the way in which data were analyzed.

3.5 | Questionnaires

All study participants were characterized by eight questions of the CGES questionnaire[24] regarding their self-reported chemical sensitivity (see supporting information S3). The median values (upper quartiles) for all questions ranged up to 2.5 (3.0) within the possible range of 1-5. There were three participants reporting values of up to 4, but these did not show markedly different responses in measurements or questionnaires compared to the other subjects.

The responses to the single questions regarding symptoms are shown in Figure 9. When summarizing these answers into symptom groups regarding either nose, or eyes, or neck/throat, or respiratory, or circulation, none of the sum scores differed between ABS and PLA exposure (Wilcoxon test). However, there were significant differences regarding well-being (P = .007) and odor nuisance (P = .015). The perceived overall interference (P = .098) and the smell perception (P = .726) were not significantly different between the two exposures.

3.6 | VOC emission

The specific mixtures of compounds in the filaments for 3D printing are usually not declared in detail. Thus, when purchasing the same material from different sources, great variations may be observed. In the present study, it was not possible to determine VOC emissions during the exposures. In order to compare the emissions from

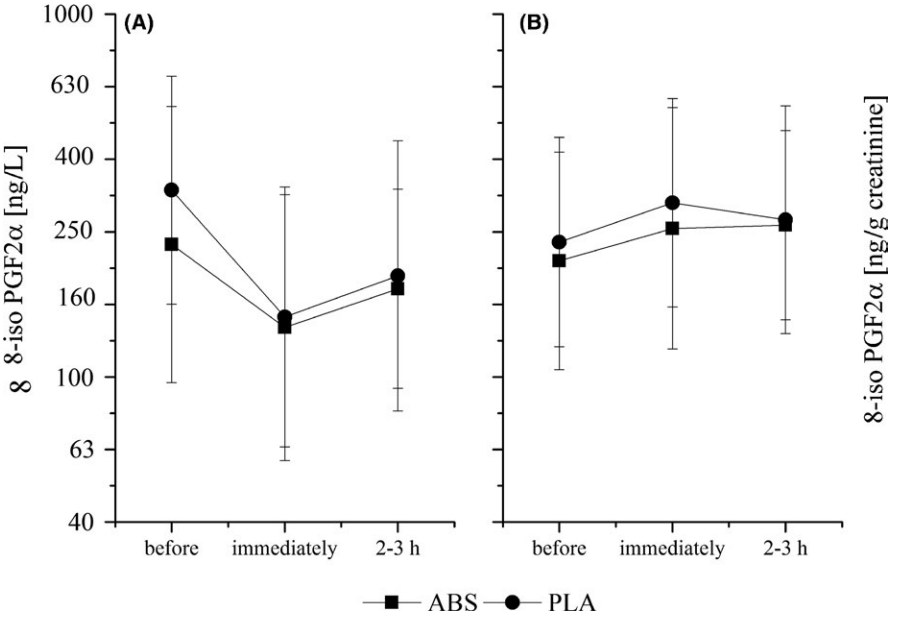

FIGURE 8 Mean values (±SD) of logarithmic transformed urinary 8-iso $PGF_{2\alpha}$ levels. Values in B are normalized for creatinine

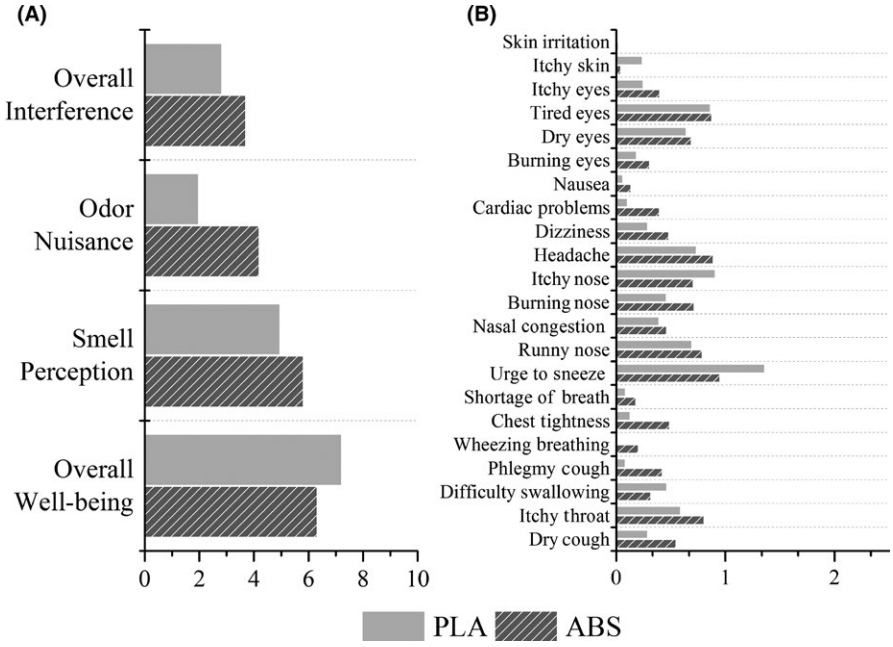

FIGURE 9 Results of questionnaires after 1-h exposure to 3D printing emissions of acrylonitrile butadiene styrene (ABS) and polylactic acid (PLA). Mean values of 26 subjects are shown. A, Questions regarding the overall experience. B, Symptoms

the filaments used in the present study with values reported in the literature,[6,7,12-14] we performed a separate VOC analysis in a laboratory setting. The result is shown in the supporting information S2.2.

4 | DISCUSSION

The present study investigated biochemical and psychological responses to controlled exposures using 3D printer emissions of healthy volunteers. Each participant was exposed to two materials, corresponding either to a high-level exposure (particle number concentration > 10^6 cm^{-3}, ABS) to nanoparticles, or a low-level exposure (<6.10^{-3} cm^{-3}, PLA). Despite the large difference in these levels, we observed no or only weak effects. There were no responses in spirometric lung function. The changes observed in nasal cytokine levels occurred in parallel for both exposures and were most likely due to mucosal irritation resulting from repeated sampling. However, the relative increases were not the same across cytokines, and IL-6 showed the strongest response. 8-iso $PGF_{2\alpha}$ in urine, a marker of oxidative stress, also showed parallel changes over time, which again could be attributed to repeated sampling. Moreover, the levels of exhaled NO showed changes over time for both exposures, but there was a significant although small difference between FeNO levels 2-3 hours after ABS and PLA exposure. This difference was mainly due to a decrease of FeNO after PLA exposure. The two exposures were perceived differently by the subjects, as reflected in less well-being and higher odor nuisance during ABS printing. Overall, the results of this controlled, 1-hour experimental exposure to 3D printer emissions did not indicate clinically relevant acute inflammatory effects of ABS and PLA. The observed changes in FeNO are difficult to explain but could be worthwhile of further exploration, because they might indicate subtle changes elicited by the materials used.

4.1 | Inflammatory markers in nasal secretion

Cytokine levels in nasal secretions were assessed using the same method as employed in our study on the effects of laser printer emissions.[10] In the present study, there were some responses but these occurred in parallel for ABS and PLA exposures. These findings were similar to the previous result comparing high and low laser printer emissions. Probably they have to be attributed to the repeated sampling which caused mucosal irritation by the cotton wool pads. The response to the irritation was not homogeneous as indicated by the differences between the changes of cytokine levels; compared to TNF-α, IL-1β, and IFN-γ, the response of IL-6 to repeated sampling was stronger. Such methodological factors are impossible to avoid in pre-post comparisons performed within finite time, and we cannot exclude that they could have masked a differential response to the materials. Indeed, the pre-post changes for TNF-α, IL-1β, and IFN-γ (as well as ECP, see below) appeared to be slightly, although not significantly, larger for ABS than PLA. This was not the case for IL-6, which might indicate a ceiling effect. Despite these tendencies, one should note that all changes and differences observed were small. They probably should not be considered as signs of physiologically or clinically relevant effects on the nasal mucosa, even for the high levels of ABS exposure.

We do not have information on inflammatory cells which could have been involved in the changes of nasal cytokine levels. Sampling of cells would have required nasal lavages,[38] which have an even stronger effect on subsequent measurements than the nasal pads which we used. Besides neutrophils, macrophages are important players. During phagocytosis of nanoparticles, macrophages release cytokines which can be used to determine the inflammatory potential of the stimulus.[39] Similarly, eosinophils can release granules containing compounds such as ECP that elicit further responses of the nasal mucosa.[40]

Since eosinophils numbers and ECP levels correlate well, ECP is often used as a marker of eosinophil activation and tissue eosinophilia.[32] Its levels could be measured in 17 subjects in whom, after the assessment of cytokines, enough nasal fluid was available at all four time points (pre and post, both materials). There was a small, non-significant increase after both exposures, without significant difference between them. These observations are similar to those described for serum ECP in our study on laser printer emissions.[10]

4.2 | FeNO

Eosinophilia is often related to the levels of nitric oxide; thus, ECP levels may be compared with FeNO despite the fact that they originate from different compartments. For this analysis, FeNO and ECP levels at the same time points were taken. Indeed, we found a significant correlation between both parameters across subjects, underlining the validity of both the assessment of nasal ECP and of bronchial NO. We do not have ECP data for comparison with the FeNO measurement performed immediately after exposure, but there is no reason to assume a lower validity of FeNO data at this time point.

While for ECP no difference in response between the materials was observed, there were statistically significant differences in the time course of FeNO after ABS compared to PLA exposure. Both exposures tended to show slightly elevated values immediately afterward but 2-3 hours later FeNO levels after PLA exposure was lower than those after ABS exposure and lower than baseline values. Thus, there was a relative increase in FeNO after ABS exposure, or conversely a relative decrease after PLA exposure. These effects are difficult to explain, and there are different factors that might have contributed to these responses.

Changes in FeNO can be due to alterations in its biochemical production or physical release, or due to scavenging in terms of chemical reactions with other compounds, especially oxidants. There is a multitude of compounds that can act as oxidants. For example, it is well known that cigarette smoking is associated with reduced FeNO values.[41] Marini et al[42] observed decreased FeNO levels not only after inhalation of tobacco smoke but also e-cigarette smoke, which is known to contain UFP and VOCs. Both, scavenging by oxidants and downregulation of NO production may have played a role in these findings. In contrast, Zhang et al[43] found a positive relationship between FeNO and the burden of air pollutants, for example, ultrafine particles which might point toward upregulation. Another factor could be a change in the diffusion barrier of NO into the airway lumen, in accordance with reductions of FeNO after inhalation of hypertonic saline.[44,45] It is not known which factors during PLA exposure could have caused such a change. We observed differences in VOCs emitted during ABS and PLA printing, in accordance with literature data,[12,17] but such an effect is unlikely, as both particle and VOC emissions from PLA were very low. Conversely, it is very unlikely that circadian changes made a significant contribution over a time period of 4-5 hours. We also performed FeNO measurements prior to spirometry thereby avoiding potential effects of forced maneuvers on FeNO that have been described.[28] However, FeNO measurements were performed after the sampling of nasal secretions. Indirect effects of nasal sampling on FeNO are not known until now. Therefore, our observation on a different response of FeNO after exposure to emissions from ABS and PLA printing remains unexplained at present.

4.3 | Urinary 8-iso $PGF_{2\alpha}$

As oxidative stress elicited by air pollutants is considered as one factor contributing to responses, we assessed the levels of 8-iso $PGF_{2\alpha}$ in urine before and after exposures. There were no significant differences between the changes in 8-iso $PGF_{2\alpha}$ levels over time that were observed for ABS and PLA. The repeated sampling took place over a time period of a few hours and thus was associated with changes in urine composition, as indicated by the decrease of creatinine levels. Normalization of 8-iso $PGF_{2\alpha}$ levels to creatinine did not indicate a difference between exposures. It resulted in a reversal of the time course of 8-iso $PGF_{2\alpha}$ compared to the results obtained without normalization.

We performed the statistical analysis with both metrics (ng/L and ng/g creatinine), but no differences between exposures were detectable. There is also the issue of time delay between exposure and potential changes in urine composition. Future studies should consider expanding the sampling period until the next day, because the formation and excretion of isoprostanes may be rather slow. Moreover, other compounds and metabolites could be measured in urine, as many potentially relevant substances are emitted in 3D printing, among them styrene, toluene, benzene and ethylbenzene.[46-48]

4.4 | Spirometry

Spirometry was included into the panel of assessments to ensure a comparable status of participants at both exposure days. No changes were observed. This indicates that potential irritating effects of 3D printer emissions, if they exist, were below the threshold to elicit a measurable effect. Acute effects of inhaled irritants on lung function in the sub-ppm range can be seen with, for example, toluene diisocyanate, which, however, appears to be a much stronger irritant than 3D printer emissions.[49]

4.5 | Symptoms

There were no differences in the self-reported symptoms between the exposures, indicating that despite the differences in exposure levels, the total burden from the UFP and VOC was too low to elicit differential effects regarding acute symptoms. Whether subjects with airway hyperresponsiveness, particularly patients with asthma, would have shown a different result is not known; in our previous study on laser printer emissions, we did not observe any difference related to the diagnosis of asthma or the presence of non-specific airway hyperresponsiveness. In contrast to general symptoms, the self-reported odor nuisance and overall well-being differ between ABS and PLA exposure, underlining that the emissions associated

with ABS printing were perceived as annoying. This appears to be in accordance with a general impression by most users.

4.6 | Chemical and general environmental sensitivity (CGES)

The general CGES questionnaire was limited to eight questions referring to "chemical odor sensitivity"; it was used for describing the characteristics of the subject population at baseline. Almost all participants were identified in the low range of the scale. When excluding the two subjects with the highest CGES responses, statistical analyses were not qualitatively different; therefore, these subjects were included in all analysis. The results of the CGES questionnaire indicate that the participants studied did not show a higher than average chemical sensitivity.

4.7 | Exposures

Participants were placed directly in front of the printer, with their head next to the printing head. This position was chosen to maximize the exposure levels but also since experience indicates that many users continuously monitor the progress of printing in order to intervene whether problems occur. Therefore, many users of desktop 3D printers at least stay near to the printer. Some researchers described even higher UFP concentration some meters away from the printer, if printing was performed in a clean room without disturbance by users.[50] As a result, the chosen exposure scenario, which might seem unrealistic at the first view, was probably close to reality, at least for printing small objects that can be manufactured within about 1 hour. It would be of interest to study potential effects of longer exposures associated with a printing of larger and more complex objects, particularly regarding the time course of FeNO. It might also be that with prolonged, multihour exposures, the sampling of nasal secretions and urine can be performed without significant carry-over effects. Possibly it is a technical challenge to maintain constant UFP emissions as high as those used by us (particle number concentration > 10^6 cm^{-3} for ABS exposure) over a longer time but as far as realistic scenarios are targeted, a close monitoring of UFP concentrations might be sufficient to estimate total and peak exposures at the subjects' positions.

In separate laboratory experiments, we analyzed ABS and PLA materials and verified the emission of several VOCs that had been identified in the emissions of 3D printers.[6,7,12,14,17] Among these were substances with high irritation potential, such as styrene, ethylbenzene, acrolein, and formaldehyde (see Figures S2 and S3 in the supporting information S2). We were not able to quantify these VOCs in the exposure chamber, due to their low concentrations, instead we measured them in the head space of vials in which the materials had been heated to temperatures comparable to those achieved during 3D printing. One might consider it alarming that these substances are found in emissions from filaments, which are sold for domestic use. As long as regulations are not established, at least the additives and most important emitted VOCs should probably be declared by the manufacturers.

4.8 | Strengths and limitations

The strength of our study is based on the fact that it was performed as a controlled human exposure study comparing two widely used materials with grossly different emissions. Moreover, we used a panel of outcome measures ranging from functional indices over biomarkers to questionnaires, for which previous experience existed. The exposure scenario mimicked a short-term printing scenario typical for the manufacture of a small object closely supervised by the user. The sample size of 26 subjects was similar to the numbers used in many other experimental exposure studies in which significant effects have been observed. The size was sufficient to detect small differences in the time course of FeNO as well as to verify differences in nuisance and odor perception. All participants were healthy subjects without conditions that could have increased the variability of results. This selection, however, corresponds to the limitation that we cannot infer potential responses in subjects who might be prone to such responses, for example, due to asthma or other respiratory conditions. Moreover, this study, being the first human exposure study in the field, was limited to unprotected short-time printing that, however, is likely to cover a substantial fraction of domestic 3D printer use. At the same time, the measurements were limited to the detection of more or less acute effects after exposures and did not comprise a longer follow-up. Prolonged or repeated exposures and long-term follow-up would be the topic of future studies and certainly require a much more elaborate design than the present study. These studies would also have to deal with the problem that even non- or low-invasive assessments, such as the sampling of urine or the placement of nasal pads, may have carry-over effects. This is even more relevant for powerful procedures such as sputum induction, bronchoalveolar lavage, or bronchial challenges. This trade-off between feasibility and sensitivity may benefit from data pointing toward differential effects that are measurable with acceptable methods, such as FeNO.

5 | CONCLUSION

It is to be expected that additive manufacturing by 3D printing will be increasingly used in both professional and private settings. The emissions of ultrafine particles and volatile organic compounds are widely varying but can be high and include irritants, thereby indicating a need for the assessment of potential health effects. Within a short-time experimental scenario mimicking the printing of a small object, we found no acute changes in healthy subjects that could be evaluated as clinically significant. There was, however, a small differential effect in the level of exhaled nitric oxide which might be worth of further attention. Irrespective of this, the odor nuisance per se, particularly with ABS printing, should indicate that sufficient ventilation during and after 3D printing at home is a reasonable provision. This seems to be particularly justified if long-term exposures in special work places or the exposure

of children and adolescents is involved. These measures may benefit from the detailed description of reduction strategies available in the literature.[6,51]

ACKNOWLEDGEMENT

The authors thank J. Reith from TSI GmbH for the generous allocation of the EEPS 3090 and M. Stein from Protein Simple for the supply of the device and his expertise, as well as J. Haidl for the development of the software for the data readout analysis. Finally, we gratefully acknowledge the commitment of all participants.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest. This study was realized by funding from the institute's sources without external support.

ORCID

I. Gümperlein http://orcid.org/0000-0003-4860-0702

S. Karrasch http://orcid.org/0000-0001-9807-2915

REFERENCES

1. Forbes.de. Wohlers report 2016: 3D printing industry surpassed $5.1 billion. 2016. https://www.forbes.com/sites/tjmccue/2016/04/25/wohlers-report-2016-3d-printer-industry-surpassed-5-1-billion/#484fc9ed19a0. Accessed October 23, 2017.
2. Canalys Inc. 3D printing market to be worth US$22.4 billion in 2020. Media Alert. 2016.
3. Bitkom Research. 3D-Druck im Hausgebrauch. 2017. https://www.bitkom.org/Presse/Anhaenge-an-PIs/2017/03-Maerz/Bitkom-Pressekonferenz-3D-Druck-Verbraucher-01-03-2017.pdf. Accessed October 23, 2017.
4. Mattel GmbH. Mattel entwickelt ThingMaker und ermöglicht Familien durch den... - Mattel GmbH. 2016. http://news.mattel.de/pressreleases/thingmaker-3d-drucker-1402363. Accessed October 23, 2017.
5. Stephens B, Azimi P, El Orch Z, Ramos T. Ultrafine particle emissions from desktop 3D printers. *Atmos Environ*. 2013;79:334-339.
6. Azimi P, Zhao D, Pouzet C, Crain NE, Stephens B. Emissions of ultrafine particles and volatile organic compounds from commercially available desktop three-dimensional printers with multiple filaments. *Environ Sci Technol*. 2016;50:1260-1268.
7. Afshar-Mohajer N, Wu CY, Ladun T, Rajon DA, Huang Y. Characterization of particulate matters and total VOC emissions from a binder jetting 3D printer. *Build Environ*. 2015;93:293-301.
8. Tang T, Gminski R, Könczöl M, Modest C, Armbruster B, Mersch-Sundermann V. Investigations on cytotoxic and genotoxic effects of laser printer emissions in human epithelial A549 lung cells using an air/liquid exposure system. *Environ Mol Mutagen*. 2012;53:125-135.
9. Sisler JD, Pirela SV, Friend S, et al. Small airway epithelial cells exposure to printer-emitted engineered nanoparticles induces cellular effects on human microvascular endothelial cells in an alveolar-capillary co-culture model. *Nanotoxicology*. 2015;9:769-779.
10. Karrasch S, Simon M, Herbig B, et al. Health effects of laser printer emissions: a controlled exposure study. *Indoor Air*. 2017;27:753-765.
11. Herbig B, Jörres RA, Schierl R, et al. Psychological and cognitive effects of laser printer emissions: a controlled exposure study. *Indoor Air*. 2017;28:112-124.
12. Kim Y, Yoon C, Ham S, et al. Emissions of nanoparticles and gaseous material from 3D printer operation. *Environ Sci Technol*. 2015;49:12044-12053.
13. Stefaniak AB, LeBouf RF, Yi J, et al. Characterization of chemical contaminants generated by a desktop fused deposition modeling 3-dimensional printer. *J Occup Environ Hyg*. 2017;14:540-550.
14. Steinle P. Characterization of emissions from a desktop 3D printer and indoor air measurements in office settings. *J Occup Environ Hyg*. 2016;13:121-132.
15. Mendes L, Kangas A, Kukko K, et al. Characterization of emissions from a desktop 3D printer. *J Ind Ecol*. 2017;21:94-106.
16. Vance ME, Pegues V, Van Montfrans S, Leng W, Marr LC. Aerosol emissions from fuse-deposition modeling 3D printers in a chamber and in real indoor environments. *Environ Sci Technol*. 2017;51:9516-9523.
17. Wojtyła S, Klama P, Baran T. Is 3D printing safe? Analysis of the thermal treatment of thermoplastics: ABS, PLA, PET, and nylon. *J Occup Environ Hyg*. 2017;14:D80-D85.
18. Yi J, LeBouf RF, Duling MG, et al. Emission of particulate matter from a desktop three-dimensional (3D) printer. *J Toxicol Environ Health A*. 2016;79:453-465.
19. Oskui SM, Diamante G, Liao C, et al. Assessing and reducing the toxicity of 3D-printed parts. *Environ Sci Technol Lett*. 2016;3:1-6.
20. Macdonald NP, Zhu F, Hall CJ, et al. Assessment of biocompatibility of 3D printed photopolymers using zebrafish embryo toxicity assays. *Lab Chip*. 2016;16:291-297.
21. Pirela SV, Martin J, Bello D, Demokritou P. Nanoparticle exposures from nano-enabled toner-based printing equipment and human health: state of science and future research needs. *Crit Rev Toxicol*. 2017;47:678-704.
22. Deng Y, Cao S-J, Chen A, Guo Y. The impact of manufacturing parameters on submicron particle emissions from a desktop 3D printer in the perspective of emission reduction. *Build Environ*. 2016;104:311-319.
23. Wong KV, Hernandez A. A review of additive manufacturing. *ISRN Mech Eng*. 2012;2012:1-10.
24. Bailer J, Rist F, Witthöft M, Paul C. Validierung eines Screeninginstruments zur Identifizierung von Multiple Chemical Sensitivity (MCS): Die Chemische Geruchssensitivitätsskala (CGSS) Validation of a Screening Instrument for Multiple Chemical Sensitivity (MCS): the Chemical Odor Sensitivit. *Bibliogr Psychother Psych Med*. 2004;54:396-404.
25. Javors MA, Hatch JP, Lamb RJ. Cut-off levels for breath carbon monoxide as a marker for cigarette smoking. *Addiction*. 2005;100:159-167.
26. Karrasch S, Linde K, Rücker G, et al. Accuracy of FeNO for diagnosing asthma: a systematic review. *Thorax*. 2017;72:109-116.
27. Pedrosa M, Cancelliere N, Barranco P, López-Carrasco V, Quirce S. Usefulness of exhaled nitric oxide for diagnosing asthma. *J Asthma*. 2010;47:817-821.
28. Dweik RA, Boggs PB, Erzurum SC, et al. An official ATS clinical practice guideline: interpretation of exhaled nitric oxide levels (FeNO) for clinical applications. *Am J Respir Crit Care Med*. 2011;184:602-615.
29. Muzz64. Fish fossilz. 2016. www.thingiverse.com/thing:1276095. Accessed July 20, 2016.
30. Miller MR, Hankinson JATS, Brusasco V, et al. Standardisation of spirometry. *Eur Respir J*. 2015;26:319-338.
31. American Thoracic Society & European Respiratory Society. ATS/ERS recommendations for standardized procedures for the online and offline measurement of exhaled lower respiratory nitric oxide and nasal nitric oxide, 2005. *Am J Respir Crit Care Med*. 2005;171:912-930.

32. Klimek L, Rasp G. Norm values for eosinophil cationic protein in nasal secretions: influence of specimen collection. *Clin Exp Allergy*. 1999;29:367-374.
33. Kramer MF, Burow G, Pfrogner E, Rasp G. In vitro diagnosis of chronic nasal inflammation. *Clin Exp Allergy*. 2004;34:1086-1092.
34. Sen C, Morimoto Y, Heak S, Cooney RV, Franke AA, Maskarinec G. Soy foods and urinary isoprostanes: results from a randomized study in premenopausal women. *Food Funct*. 2012;3:517-521.
35. Helmersson J, Larsson A, Vessby B, Basu S. Active smoking and a history of smoking are associated with enhanced prostaglandin F2α, interleukin-6 and F2-isoprostane formation in elderly men. *Atherosclerosis*. 2005;181:201-207.
36. Voynow JA, Kummarapurugu A. Isoprostanes and asthma. *Biochim Biophys Acta*. 2011;1810:1091-1095.
37. Seet RCS, Lee CY, Loke WM, et al. Biomarkers of oxidative damage in cigarette smokers: which biomarkers might reflect acute versus chronic oxidative stress? *Free Radic Biol Med*. 2011;50:1787-1793.
38. Nowak D, Jörres R, Martinez-Müller L, et al. Effect of 3 hours of passive smoke exposure in the evening on inflammatory markers in bronchoalveolar and nasal lavage fluid in subjects with mild asthma. *Int Arch Occup Environ Health*. 1997;70:85-93.
39. Oberdörster G, Maynard A, Donaldson K, et al. Principles for characterizing the potential human health effects from exposure to nanomaterials: elements of a screening strategy. *Part Fibre Toxicol*. 2005;2:8.
40. Kramer MF, Jordan TR, Klemens C, et al. Factors contributing to nasal allergic late phase eosinophilia. *Am J Otolaryngol* 2006;27:190-199.
41. Dressel H, de la Motte D, Reichert J, et al. Exhaled nitric oxide: independent effects of atopy, smoking, respiratory tract infection, gender and height. *Respir Med*. 2008;102:962-969.
42. Marini S, Buonanno G, Stabile L, Ficco G. Short-term effects of electronic and tobacco cigarettes on exhaled nitric oxide. *Toxicol Appl Pharmacol*. 2014;278:9-15.
43. Zhang X, Staimer N, Gillen DL, et al. Associations of oxidative stress and inflammatory biomarkers with chemically-characterized air pollutant exposures in an elderly cohort. *Environ Res*. 2016;150:306-319.
44. Beier J, Beeh KM, Kornmann O, Buhl R. Sputum induction leads to a decrease of exhaled nitric oxide unrelated to airflow. *Eur Respir J*. 2003;22:354-357.
45. Zuiker RGJA, Boot JD, Calderon C, et al. Sputum induction with hypertonic saline reduces fractional exhaled nitric oxide in chronic smokers and non-smokers. *Respir Med*. 2010;104:917-920.
46. Ogata M, Taguchi T. Quantitation of urinary metabolites of toluene, xylene, styrene, ethylbenzene, benzene and phenol by automated high performance liquid chromatography. *Int Arch Occup Environ Health*. 1987;59:263-272.
47. Shi CY, Chua SC, Lee BL, Ong HY, Jeyaratnam J, Ong CN. Kinetics of styrene urinary metabolites: a study in a low-level occupational exposure setting in Singapore. *Int Arch Occup Environ Health*. 1994;65:319-323.
48. Weisel CP. Benzene exposure: an overview of monitoring methods and their findings. *Chem Biol Interact*. 2010;19:58-66.
49. Allport DC, Gilbert DS, Outterside SM. *MDI and TDI: Safety, Health and the Environment : A Source Book and Practical Guide*. New York, NY: J. Wiley; 2003.
50. Zhou Y, Kong X, Chen A, Cao S. Investigation of ultrafine particle emissions of desktop 3D printers in the clean room. *Procedia Eng*. 2015;121:506-512.
51. Kwon O, Yoon C, Ham S, et al. Characterization and control of nanoparticle emission during 3D printing. *Environ Sci Technol*. 2017;51:10357-10368.

SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.

How to cite this article: Gümperlein I, Fischer E, Dietrich-Gümperlein G, et al. Acute health effects of desktop 3D printing (fused deposition modeling) using acrylonitrile butadiene styrene and polylactic acid materials: An experimental exposure study in human volunteers. *Indoor Air*. 2018;00:1–13. https://doi.org/10.1111/ina.12458