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Performance of a whole-body human dust inhalation challenge exposure chamber



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ARTICLE INFO	A B S T R A C T	
<i>Keywords:</i> Exposure chamber Human Inhalation challenge Dust	<i>Background:</i> Evaluation of the performance of a whole-body human dust exposure chamber is presented in this report. <i>Methods:</i> The volume of the chamber is 2.13 m ³ and it is operated at a flow rate of 1.0 m^3 /min. Makeup and exhaust air were filtered. A Wright Dust Feeder was used to generate fly ash, the testing agent. An elutriator was used to maintain particles in the respirable range. A Rupprecht and Patashnick PM-10 TEOM, a direct reading instrument, was used to monitor particle concentration. Particle size distributions were determined by a QCM cascade impactor. The evenness of dust concentrations in the chamber was determined gravimetrically. <i>Results:</i> Dust concentrations measured at different points within the chamber were associated with variability less than 10%. Dust concentrations measured by the TEOM, in $\mu g/m^3$, at 0.2, 0.4, 0.8 and 1.6 RPMs of the Wright Dust Feeder, were 110 ± 2.8 , 173 ± 8.5 , 398 ± 20 and 550 ± 17 , respectively. Particle size distributions (MMD and GSD) were 1.27μ m and 2.35 , 1.39 and 2.22 , 1.46 and 2.08 , 1.15 and 2.2 , respectively. Total dust concentrations measured gravimetrically in $\mu g/m^3$, were 135 ± 21 , 200 ± 35 , 333 ± 18 and 891 ± 27 , respectively. <i>Conclusion:</i> The whole-body human exposure chamber offers several advantages and has better performance than most of the inhalation challenge systems previously described.	

1. Introduction

Description of inhalation challenge systems had been previously discussed in the literature [1–3].

Lidén et al. [4] developed an inhalation system for investigating baker's asthma, dermatitis and urticaria. The test material used for the study was wheat flour and it was dispersed using a rotating brush. The investigators reported an average concentration of 5 mg per cubic meter (mg/m³) with the possibility of reaching concentrations up to 12 mg/m³. Using Casella cyclones, it was determined that 6% to 12% of the total dust concentration was in the respirable range. Spatial variation was reported to be 15% and the temporal variation was in the range of 7% to 11%. Median particle size ranged from 6 µm to 10 µm for fine particles and 50 µm for coarse particles.

In 2006, the same inhalation challenge system was used by Lundgren et al. [5] with wheat flour, pinewood dust and glove powder.

Direct readings were made with light scattering instruments. Institute of Occupational Medicine (IOM) samplers and Casella cyclones were used for determination of inhalable and respirable fractions. Open face cassettes 37 mm in diameter were used for the determination of total dust concentration. The investigators reported achieving total dust concentrations of 5 mg/m³ for wheat flour, 6 mg/m³ for pinewood and glove powder. The coefficient of variation ranged from 6% to 10%. Respirable dust fraction concentrations were 0.5 mg/m³ for one type of wheat flour and 0.3 mg/m³ for two other types of wheat flour. Respirable fraction concentration for pinewood was 1 mg/m³ and 0.6 mg/m³ for glove powder. The coefficient of variation for pinewood was 9% and 10% for glove powder. Wheat flour variation of concentration ranged from 21% to 36%.

Taylor et al. [6] developed a whole-body human exposure chamber for endotoxin exposure. For the creation of the endotoxin aerosol, the bacterium *Enterobacter agglomerans* was adhered to microcrystalline

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Abbreviations: TEOM, Tapered element oscillating microbalance; IOM, Institute of Occupational Medicine; mg/m³, milligrams per cubic meter; µm, micrometer; mm, millimeter; µg/m³, micrograms per cubic meter; PM_{2.5}, particulate matter 2.5 µm or smaller; PM₁, particulate matter 1 µm or smaller; m³, cubic meter; m³/min, cubic meter per minute; HEPA, high efficiency particulate air; °C, celsius; RPM, revolutions per minute; l/min, liters per minute; cm, centimeters; QCM, quartz crystal microbalance; PVC, polyvinyl chloride; MMD, mass median diameter; GSD, geometric standard deviation

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lattice particles. They reported that aerosol concentrations ranged from $250 \,\mu\text{g}$ per cubic meter ($\mu\text{g/m}^3$) to $400 \,\mu\text{g/m}^3$.

Sällsten et al. [7] described an inhalation challenge system for wood smoke exposures. The smoke was generated by burning hardwood and softwood. Subjects were exposed for four hours. Continuous monitoring of the smoke inside the chamber was made with a tapered element oscillating microbalance (TEOM 1400). Particulate matter 2.5 µm or smaller (PM2.5) and particulate matter 1 µm or smaller (PM1) measurements were obtained with stationary and personal air sampling equipment. Particle size distributions were determined with an electric low-pressure impactor. The investigators noted that there was no difference in concentrations between PM2.5 and PM1 data. Data from the electric low pressure impactor indicated that almost all particles sampled had a diameter less than one micrometer. During two sessions, the particle size distribution of the first session had a geometric mean diameter of 0.042 µm with a geometric standard deviation of 1.7. For the second session, the geometric mean diameter was 0.112 µm with a geometric standard deviation of 1.4. Average particle concentrations were in the range of 240 μ g/m³ to 280 μ g/m³.

In 2008; Eduard et al. [8], developed a whole-body exposure chamber for inhalation challenge studies using aerosols. An air lock was used to avoid any disturbance of the concentration when entering into the test atmosphere, and subjects would enter only when the test atmosphere had reached equilibrium. The aerosols were dispersed using a fluidized bed generator. Large particles were removed using a cyclone with a cut-off diameter of 3.5 µm. The test material used in this study was fused aluminum oxide. The investigators reported that equilibrium concentration was reached between 30 and 60 min after the generation was started, depending on target concentration. The concentrations of the test material measured at different positions inside of the exposure chamber were statistically different. Concentrations at the center of the chamber were higher than the concentrations in the periphery. For concentrations lower than the 1 mg/m³, the coefficient of variation of aerosol concentration was 10%-19%. When concentrations in the chamber were higher than 1 mg/m³, the researchers reported a coefficient of variation of 4%-6%. Concentrations could be maintained for more than 1 h after reaching a stable concentration. Particle size distribution determined by an optical particle counter showed a median diameter of 5.7 µm without the cyclone. With the cyclone attached to the generator, the median diameter was 2.9 µm. This inhalation challenge system was also used by Sikkeland et al. [9].

Isaxon et al. [10] used a whole body human exposure chamber for the study of nanosized particles produced during welding activities. The 21.6 m³ chamber room had its interiors made out of stainless steel, with the exception of its window (0.8 m²). The chamber which is entered via an antechamber has the capacity of testing three volunteers at the same time and it was maintained at a slight positive pressure. Generation of particles was performed by using MAG welding on stainless steel. For the generation of the fume, a welding pulse of 3 min was generated followed by 20 min of no welding activities. This generation took place in a 1.33 m³ adjacent chamber. Once generated, the welding fume was moved to the exposure chamber with pressurized air. Before entering the exposure chamber, the fume passed through a cyclone to eliminate non-desired particle sizes. The fume was then pre mixed with the air entering the exposure chamber. Spatial concentrations gradients were estimated to be less than 15%. Monitoring of particles was performed by using a PM_{2.5} TEOM. Measurement of particle number concentration was performed using a scanning mobility particle system. The researchers were able to obtain an average concentration of $1000 \pm 70 \,\mu\text{g/m}^3$. Particle geometric mean was around $160 \pm 10 \,\text{nm}$.

In 2016, Chen et al. described an exposure chamber for the generation of sulfate and traffic soot particles [11]. The 30 m³ chamber was constructed with stainless steel interiors. The sulfate aerosol was generated using an atomizer aerosol generator connected to a diffusion dryer. Soot particles were generated using a real soot generator with propane gas and nitrogen. Once the soot particles were generated, they were diluted with purified air and cuprous oxide was used to control carbon monoxide. Concentration inside the chamber was measured by using a Dust-Trak. Sulfate aerosols were introduced into the chamber until a target concentration of $80 \,\mu g/m^3$ was reached. Soot particles were produced through 20 s combustions until the target concentration of $13 \,\mu g/m^3$ was reached. The measurement of the number and area concentrations of particles within the size range of 6 nm and 560 nm was performed with a Fast Mobility Particle Sizer. The mean mass concentration of sulfate generated during the 2 h exposure studies were 74.19 ± 3.46 $\mu g/m^3$ for sulfate aerosols and $11.54 \pm 0.57 \,\mu g/m^3$ for soot. The researchers acknowledge that due to instrument' limitations, particles within PM_{2.5} range were not measured.

Other reports about utilization and performance of inhalation challenge systems were also reported by Hammad et al. [12], Sandström et al. [13], Jönsson et al. [14], Rudell et al. [15], Sundblad et al. [16], Schiffman et al. [17], Suarez et al. [18], and Tuomainen et al. [19]. It should be noted that the extent and effect of dust deposition on the walls of the exposure chambers was not investigated in any of the studies mentioned above.

The purpose of this study was to develop a human inhalation challenge system that can deliver a controlled low dose of fly ash dust in the respirable range to one or two persons for up to six hours. This activity is part of the research program of the Breath Laboratory at the College of Public Health, University of South Florida in Tampa, FL [20].

2. Materials and methods

The whole-body human exposure chamber is located at the USF College of Public Health and can be used for generation of gases and particulates. This report is focused on the generation of particulates. Performance with gases has been reported previously [21]. The Plexiglas chamber has a volume of 2.13 cubic meter (m³) (1.25 m × 0.8 m × 2.0 m) and it is operated at negative pressure of 10 cm of water (0.98 kPa) to avoid any leakages of the test agent into the laboratory. The flowrate of the system is 1 cubic meter per minute (m³/min). A schematic of the system is shown in Fig. 1.

Two high efficiency particulate air (HEPA) filters (AstroCel HCX*) made by American Air Filters International, Louisville, KY; are used for air filtration. One filter is located at the intake of the system to avoid particles entering the system and mixing with the test agent. A second filter is located after the exposure chamber to prevent particles from reaching the air blower.

Air flow measurements and control of the exposure chamber are performed with orifice meters. A Magnehelic gauge is connected to the orifice meter to measure the differential pressure before and after the orifice [22]. The difference in pressure before and after the orifice is related to the air flow rate. The orifice meters were calibrated with a Micro-Pitot tube [23].

A Spiral TM SL4P2 air blower manufactured by Ametek Industrial Products, Harleysville, PA; is used for moving air in the system. It has the capacity to move up to 2 cubic meters of air per minute. An air bypass before the air blower is installed to regulate the flow rate of air in the system. Filtered air from the blower is exhausted through a laboratory fume hood.

The fly ash used in this investigation was donated by a local power plant. Fly ash was oven dried at 200 Celsius (°C) for 12 h before using it for dust generation. This drying process minimizes the caking effects of moisture in the fly ash and enhances the generation process. The heating procedure will not affect the integrity of the fly ash because it is actually produced at much higher temperatures.

A Wright Dust Feeder (BGI Incorporated, Waltham, Massachusetts), was used for the generation of the fly ash dust cloud. The principle of this generator is that the dust is packed in the dust generator's chamber and then scraped with a rotating blade [24]. For the characterization of the exposure chamber, the Wright Dust Feeder was used at the following revolutions per minute (RPM) settings: 0.2, 0.4, 0.8 and 1.6. Dry



Fig. 1. Schematic Diagram of Whole-Body Human Exposure Chamber for Particulates.

nitrogen from a compressed cylinder, at a flow rate of 8.4 liters per minute (l/min), was used to carry the fly ash from the dust generator to the inhalation challenge system through a 5 liters dust trap installed after the generator.

A vertical elutriator was used for the separation of large and agglomerated fly ash particles [25]. It is constructed from a Plexiglas pipe with an inside diameter of 18.5 cm. The aerosol dust cloud passing through the elutriator is directed towards the chamber. The diameter of the largest particles of fly ash passing through the elutriator and entering the exposure chamber is theoretically 8 μ m, however in reality it is about 10 μ m [25].

A TEOM 1400ab was used for the continuous measurement of particle concentration inside of the exposure chamber. The concentration displayed by the instrument is an average of 10 min that updates every 2 s. The results are reported in $\mu g/m^3$ [26].

Determination of particle size distributions at different rates of generation was obtained using a quartz crystal microbalance (QCM) cascade impactor, Model PC-2 manufactured by California Measurements. The cascade impactor has 10 stages and the cut-off aerodynamic diameters of the instrument range from 0.1 μm to $>35\,\mu m$. Since the first stage of the impactor doesn't have pre-impactor, the cut- off point of the first stage is $>35\,\mu m$.

PVC filters 37 mm and 25 mm in diameter and 5 μ m pore size were used for determination of total dust concentrations, inhalable and respirable fraction concentrations inside the chamber. Determination of total dust concentration was performed by using 37 mm open-face cassettes. SKC aluminum cyclones were used for the determination of particulate concentration in the respirable fraction. The inhalable fraction concentration was determined using a SKC Button Aerosol Sampler with 25 mm PVC filters.

2.1. Particle size distribution

Particle size distributions were determined using the QCM cascade impactor. Five consecutive particle size distributions were obtained at each RPM setting in order to determine an average particle size distribution for each RPM setting.

2.2. Evenness of concentration in the chamber

Twelve PVC filters 37 mm in diameter placed in open face cassettes were used for the determination of dust concentration across the exposure chamber. The filters were placed facing down 135 cm above the floor of the chamber, the height of the breathing zone of a person sitting inside the chamber. Five consecutive runs of fly ash were made. Gravimetric analysis was made following the NIOSH Analytical Method 0500 [27]. For the purpose of comparison, the results from the twelve dust filters were divided into three patterns. The first pattern was for comparison between four rows of the filters. The second pattern was for comparison of two rows at the front and two rows at the back the chamber. The third pattern was for comparison between three groups that represent left, middle and right side of the exposure chamber. Representations of these patterns are shown in Fig. 2.

To investigate the effect of the presence of a person on the evenness of the dust concentration in the chamber, a full size mannequin (Allen Display, Midlothian, VA) was set up in the sitting position in the center of the chamber. Specifically for this purpose, a larger size glass bead dust cloud was generated in the chamber with and without the mannequin. The larger particle size dust was selected so that the effect of the presence of the mannequin would be more pronounced. The vertical elutriator was not utilized during this procedure. The evenness of the dust concentration across the chamber as well as the size distribution of the airborne glass beads were measured in the same way as described previously.

2.3. Concentration of fly ash

As stated previously, a vertical elutriator was used for the removal of large particles of fly ash. Therefore it is not possible to determine the rate of generation before the test material is measured inside the chamber. To describe the concentration in the chamber at different rates of generation, a correlation was obtained between the four different RPM settings of the dust generator and the concentration obtained inside the chamber. For each RPM setting, 5 consecutive dust generations were made and each run lasted 60 min. The concentration of the test material during each run was determined with the TEOM. An average profile of the dust concentration was obtained for each RPM



Fig. 2. Patterns for Determination of Distribution of Concentration.

setting.

Studies reported in the literature showed that there is a difference between the results of the TEOM and gravimetric analysis [28,29]. Therefore, determination of total, inhalable and respirable dust fraction concentrations, were made in comparison to the TEOM.

2.4. Statistical analysis

2.4.1. Evenness of concentration

Twelve open face 37 mm cassettes with polyvinyl chloride (PVC) filters were operated for determination of concentration levels across the chamber. As described earlier, dust concentration values were gathered in different group pattern for comparison. Wilcoxon rank-sum test was used as the statistical test for this comparison. This non parametric test was selected because of low sample size and normality of distribution could not be assumed. Multiple comparisons were made for each pattern and a Bonferroni correction was performed depending on the amount of comparisons made. The same procedures were followed for the glass beads.

2.4.2. Particle size distribution

Five particle size distributions were obtained for each RPM setting. The five particle size distributions were then combined to obtain an average particle size distribution for each RPM setting. The same procedures were followed for the glass beads. Wilcoxon rank-sum test was also used as the statistical test for this comparison because of the low sample size and the particle size distributions have a log-normal distribution. A Bonferroni correction was made for six comparisons.

3. Results

3.1. Concentration profiles of fly ash particles

The evaluation and characterization of the inhalation challenge system for particulates was made at four different RPM settings: 0.2, 0.4, 0.8 and 1.6. Five consecutive particle generations were made at each RPM setting. An example of the consistency of dust generation is shown in Fig. 3.

Average concentration profiles obtained with the TEOM instrument at different RPM settings are shown below in Fig. 4.

The dust introduced into the chamber is passed through a vertical elutriator and the larger particles are removed. This means that the true rate of particle generation is unknown. However, the rate of generation can be estimated from the concentrations measured. An example of the estimated dust concentration and the average profile of dust concentration at RPM 1.6 are shown in Fig. 10.

The observed lag between the actual and estimated values may be attributed to instrument performance. The direct reading instrument measuring particle concentration (TEOM) displays a moving average



Fig. 3. Five Consecutive Particle Generation at RPM Setting 1.6.



Fig. 4. Average Concentration Profile of Fly Ash at Different RPM Settings.

that is determined over a period of 10 min. In a previous report [21] about the performance of the system with gases and vapors, direct reading instruments showed excellent agreement with the estimated values of the model.

The maximum concentrations values measured for particles and those predicted by the model agree. It is believed the actual buildup and decay profiles for gases and particulates are similarly close to the model, but the limitations of the TEOM are probably the cause of this disagreement.

Total, inhalable and respirable dust concentrations were also determined at different RPM settings of the dust generator and correlated with the concentrations obtained with the TEOM. Total and inhalable dust concentrations were approximately 1.2 times higher than the concentrations measured by the TEOM. The coefficient of correlation for both regression lines were 0.992 and 0.997. These results are in agreement with results published elsewhere [28,29]. Good agreement was obtained between the inhalable button sampler and the 37 mm open face cassettes. The inhalable fraction concentrations were found to be similar to the total dust concentrations. Respirable dust concentrations were about 80% of the concentrations of the TEOM. Similarly, respirable dust concentrations were found to be 67% of the total dust concentrations.

Total dust, inhalable and respirable fraction concentrations were determined at each RPM setting as described before. The obtained concentrations were plotted against average concentrations determined by the TEOM. The results are presented in Figs. 5–7.

The calculated coefficient of determination or R^2 for each of the three regression lines were 0.992, 0.997 and 0.969. The intercepts in Figs. 5 and 6 are not statistically different from zero (p > 0.05). The intercept in Fig. 7 is statistically different from zero (p < 0.05). A comparison between the total dust concentrations and the inhalable and respirable fraction concentrations can be seen in Fig. 8.



Fig. 5. Total Dust Concentrations vs. TEOM Average Readings at Different RPM Settings.



Fig. 6. Inhalable Fraction Concentrations vs. TEOM Average Readings at Different RPM Settings.



Fig. 7. Respirable Fraction Concentrations vs. TEOM Average Readings at Different RPM Settings.



Fig. 8. Regression of Average Total Dust Concentrations vs. Inhalable and Respirable Fraction Average Dust Concentrations.

The calculated coefficient of determination or R^2 for both regression lines was 0.999. The intercept for the inhalable regression is not statistically different from zero (p > 0.05), but the intercept for the respirable fraction is significantly different from zero (p < 0.05). A summary of average dust concentrations obtained at different RPM settings is shown in Table 1.

3.2. Evenness of concentration across the exposure chamber

The concentration across the exposure chamber was determined by placing twelve open face cassettes for collection of test material. The positions of the cassettes inside of the exposure chamber are shown in Fig. 2. The evenness of distribution in the chamber was obtained by normalizing the dust concentrations at different rates of generation. Dust concentrations are shown in Table 1. The coefficient of variation

Table 1			
Average Dust Concentrations	Obtained by	Gravimetric	Analysis.

RPM	Total Dust Concentration (μg/m³)	Inhalable Fraction Concentration (µg/m ³)	Respirable Fraction Concentration (µg/m ³)
0.2	135	158	134
S.D.	20.5	28.3	33.9
C.V.	15.2%	17.9%	25.3%
0.4	200	210	181
S.D.	34.9	18.7	39.8
C.V.	17.5%	8.90%	21.9%
0.8	333	337	276
S.D.	18.0	10.2	24.0
C.V.	5.40%	3.03%	8.70%
1.6	891	898	644
S.D.	27.0	7.55	54.9
C.V.	3.04%	0.84%	8.50%

RPM - Revolutions per minute.

S.D. – Standard deviation.

C.V. - Coefficient of variation.



Fig. 9. Particle Size Distributions at Different Rates of Generation.

of the dust concentration inside the chamber was 7.6%. For the characterization and analysis of the evenness of concentration across the exposure chamber, the concentrations obtained by the open face cassettes were grouped in three different patterns. As stated before, this potential effect was investigated with the mannequin and coarse glass beads. No significant differences were observed (p > 0.42). It is possible; a person sitting in the chamber will change the air flow pattern. However, this problem can be readily solved by using personal air sampling equipment positioned in the breathing zone of the test subject.



Fig. 10. Observed and Estimate Dust Concentrations at Rate of Generation RPM 1.6.

Table 2MMD and GSD Obtained at Each RPM Setting.

RPM	MMD (µm)	GSD
0.2	1.27	2.35
0.4	1.39	2.22
0.8	1.46	2.08
1.6	1.15	2.20

3.3. Particle size distributions

Five consecutive particle size distributions were obtained at each RPM setting, and then an average particle size distribution was calculated for each RPM setting. The results are shown in Fig. 9. Mass median diameters (MMD) and geometric standard deviations (GSD) are presented in Table 2. Particle distributions were not significantly different (p > 0.60) for all the RPM settings of the dust generator. The mass median diameters of the fly ash ranged from 1.15 µm to 1.46 µm and the geometric standard deviations ranged from 2.08 to 2.35. These results were expected because the same batch of fly ash was used for all dust generations. The mass median diameter of the glass beads ranged from 7.61 µm to 8.43 µm and the geometric standard deviations ranged from 1.39 to 1.50. The use of a mannequin showed no effect on the particle size distributions generated inside the chamber (p > 0.15).

4. Conclusion

The whole-body human exposure chamber at the USF Sunshine ERC Breath Laboratory offer several advantages over many inhalation challenge systems previously reported in the literature. These advantages include fast response in terms of buildup and decay for both gases and particulates, predictability and reproducibility of reaching target concentrations, low variability of concentration within the chamber and its ability to be operated for the generation of particles within the respirable and thoracic size ranges. The description of this work and the detail of the methodology used in this experiment constitutes a guideline for future investigators conducting this type of work resulting in significant savings of time and resources.

Availability of data and materials

Please contact authors for data request.

Author's contributions

YYH designed and built the exposure chamber. LFP and YYH generated and analyzed the data. LFP and YYH worked together on the development of this paper.

Competing interests

The authors declare that they have not competing interests.

Transparency document

The Transparency document associated with this article can be found in the online version.

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