



August 7, 2018 College of Engineering
Department of Chemical and
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Dear Dr. Troyer,

Enclosed please find a manuscript entitled "A New Portable In Vitro Exposure Cassette for Aerosol Sampling" (Authors: Lynn E. Secondo, Nathaniel J. Wygal, and Nastassja A. Lewinski) for your consideration as an article in the Journal of Visualized Experiments. All authors have read and approved the submission of the manuscript before exclusively submitting it to this journal. This paper has not been previously published or accepted for publication.

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In this manuscript, we present a new approach for *in vitro* lung toxicity testing that is enabled by the portable *in vitro* exposure cassette or the PIVEC. To our knowledge, this is the first device of its kind to allow for air-liquid interfaced cell exposures to aerosols within the breathing zone of a person. The PIVEC was designed as an adapter to the 37 mm filter cassette that is commonly used to collect aerosols on a filter. By replacing the filter with a framework to support a cell culture insert, the design is capable of exposing cells at the air-liquid interface (ALI). *In vitro* studies utilizing cells grown and exposed at the ALI have been repeatedly demonstrated to provide a more biologically accurate response.

In addition to describing the design, characterization of the PIVEC was performed by measurement of the deposition efficiency, relating the amount deposited onto the cells to the amount administered to the system, over three sizes of copper nanoparticles. Exposure of cells within the PIVEC to the copper nanoparticles was performed with a focus on cytotoxicity and oxidative stress endpoints. The PIVEC is compared to literature values for commercial systems with the same aerosol flow pattern including the Cultex CG and RFS and the Vitrocell. The portability of the PIVEC allows on-site *in vitro* testing of aerosols by taking the lab to the field to investigate near the source of emission. As an inexpensive device, multiple PIVECs can be used to monitor spatial differences. This portability will provide many opportunities that were not previously available with current ALI exposure systems.

If we can provide any further information that would be helpful in evaluating this manuscript, please contact us at (804) 828-0452 or by email at nalewinski@vcu.edu.

Thank you for your time and consideration.

Sincerely,

Nastassja Lewinski, Ph.D.
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