Video Article

# Visualization of Ambient Mass Spectrometry with the Use of Schlieren Photography

Gregory T. Winter[1], Joshua A. Wilhide[1], William R. LaCourse[1]

[1]Department of Chemistry and Biochemistry, University of Maryland, Baltimore County

Correspondence to: William R. LaCourse at lacourse@umbc.edu

URL: https://www.jove.com/video/54195
DOI: doi:10.3791/54195

Keywords: Chemistry, Issue 112, mass spectrometry, ambient ionization, spray half angle, schlieren photography, visualization technique, gas stream imaging

Date Published: 6/20/2016

Citation: Winter, G.T., Wilhide, J.A., LaCourse, W.R. Visualization of Ambient Mass Spectrometry with the Use of Schlieren Photography. *J. Vis. Exp.* (112), e54195, doi:10.3791/54195 (2016).

## Abstract

This manuscript outlines how to visualize mass spectrometry ambient ionization sources using schlieren photography. In order to properly optimize the mass spectrometer, it is necessary to characterize and understand the physical principles of the source. Most commercial ambient ionization sources utilize jets of nitrogen, helium, or atmospheric air to facilitate the ionization of the analyte. As a consequence, schlieren photography can be used to visualize the gas streams by exploiting the differences in refractive index between the streams and ambient air for visualization in real time. The basic setup requires a camera, mirror, flashlight, and razor blade. When properly configured, a real time image of the source is observed by watching its reflection. This allows for insight into the mechanism of action in the source, and pathways to its optimization can be elucidated. Light is shed on an otherwise invisible situation.

## Video Link

The video component of this article can be found at https://www.jove.com/video/54195/

## Introduction

Mass Spectrometry, an analytical tool available for molecular mass identification, has become one of the most powerful analytical techniques to date. Over the last decade a whole host of new ambient ionization sources have become available for mass spectrometry detection. For the data collected in this manuscript, the Direct Sample Analysis (DSA) source was utilized. Although these sources are extremely versatile, a more detailed knowledge of the physical ionization process is needed for its optimization and extension of purpose. The aim of this experiment is to gain a better understanding of the ionization process within the ambient sources through visualization of the nitrogen stream on the device using a technique called schlieren photography.

Scientific study often initiates through observation, which is difficult if the object of study is transparent to the naked eye. Schlieren photography is a technique that allows the invisible to become visible through relying on changes in the refractive index within transparent media[1]. The inhomogeneity of the refractive indices causes a distortion of the light allowing for visualization. The schlieren technique has been routinely used in a variety of specialty fields including ballistics modeling, aerospace engineering, general gas detection and flow monitoring, and at times to visualize protein bands in gel electrophoresis[2-5].

Most ambient ionization sources use a stream of gas in order to facilitate the ionization. A wide range of conditions can exist for source options, however the parameters of this experiment must involve the utilization of a gas with a refractive index that differs from the surrounding lab air. This specific study utilizes hot nitrogen. It should be noted that only a small difference in refractive index is observed between pure nitrogen from the gas stream and air at RT[6], mainly because air is composed mostly of nitrogen. This issue is overcome in this instance due to the high temperatures of the pure nitrogen in the gas stream which produces a significant enough change in refractive index for the gas to be observed.

Other mass spectrometry sources such as a Desorption Atmospheric Chemical Ionization (DAPCI)[7], Flowing Atmospheric Pressure Afterglow (FAPA)[8-10], and Direct Analysis in Real Time (DART)[11] ionization sources have used schlieren photography. The intention of this protocol is to discuss how to study ambient ionization using a basic schlieren photography configuration. This technique, however, is applicable to any number of different analytical techniques that involve gaseous streams.

## Protocol

## 1. Schlieren Photography

1. **Establishment of the Test Region**
   **Note:** The test region exists directly in front of the mirror.

 

1. Clamp a spherical concave mirror (150 mm diameter, focal length 1,500 mm) in a ring stand clamp large enough to support the mirror. Attach the ring stand clamp with the mirror to a ring stand perpendicular to the floor. The current study used a 3 foot ring stand, but any height can be used as long as it is tall enough to be able to center the mirror in the viewing window of the source.
2. Place the ring stand and mirror to the side of the mass spectrometer source. Make the face of the mirror parallel to, and at the same height, as the source.
3. Position the mirror so its center is aligned with the center source region of the mass spectrometer. Some overlap of the instrument will occur.

2. **Cutoff, Camera and Light Source**
    1. **Cutoff**
        1. Attach a metal plate to the top of the tripod. The plate will serve as a platform to hold both the razor blade and the light source. The razor blade acts as what is known as the "cutoff" in schlieren photography.
        2. Attach the razor blade to the metal plate using a magnet so that the sharp edge is vertical.
        3. Place the tripod in line with the mirror at twice the focal length of the mirror, 3,000 mm. Align the razor blade orthogonal to the path of light reflected from the mirror.
        4. Manually adjust the height of the tripod so that the sharp edge of the razor blade is approximately aligned with the center of the mirror.
        NOTE: Fine adjustment will happen later.

    2. **Camera**
        1. Mount a digital camera with 300 mm telephoto lens on a separate tripod.
        2. Position the camera so the lens (when at full zoom) is 4 cm directly behind and at the same height as the razor blade. Do not remove the lens cap at this time.

    3. **Optional Monitor**
        1. Connect the video output of the camera to a computer monitor or TV to easily view the schlieren phenomenon in real time.
        NOTE: This is a recommended process. This procedure may vary depending on the type of camera used.

    4. **Pinhole Light Source**
        1. Drill a small hole (approximately 0.6 mm in diameter) into the center of a cover (in this case, a vial cap the same diameter of the flashlight was used) which can be attached/taped to the light source. Ensure that the cover has sufficient diameter to completely cover the flashlight lens.
        2. Attach the cover over a 200 lumen LED flashlight using foil tape.
        NOTE: The flashlight will get warm and a high temperature tape is recommended.

    5. **Light Source Positioning**
        1. First use a laser pointer to align the light source with the mirror, razor blade, and camera, to ensure proper positioning of the light source.
        2. Place the laser pointer on the metal plate next to the razor blade.
        3. Manually move the laser pointer so the beam is hitting the center of the mirror. Adjust as necessary to ensure the reflected beam intersects orthogonally to the razor blade so that approximately half of the beam is blocked.
        4. Manually adjust the mirror's position to aim the beam of the laser pointer directly at the razor blade if the beam alignment was not achieved in 1.2.5.3.
        **CAUTION!** Do not look directly into the laser pointer or the reflected beam.
        5. Ensure that the laser beam is centered on the lens while keeping the lens cap on the camera.
        6. Replace the laser pointer with the covered flashlight while everything is aligned. Ensure that the flashlight is in the same orientation as the laser pointer.
        7. Turn on the flashlight and, using a piece of white paper, observe the reflected light at the cutoff. Ensure that the beam is a small focused spot at the cutoff.
        8. Make any vertical adjustments necessary to block approximately half of the reflected light beam with the cutoff.
        9. Remove the lens cap on the camera and focus on the mirror.
        NOTE: It is recommended that the camera/lens be used in manual focusing mode.

## 2. Example Test Object: Mass Spectrometry Ionization Source

1. Manually align the mass spectrometry ion source within the test region, with a distance of 10 mm between the end of the nozzle and the inlet of the mass spectrometer.
2. Manually open the needle valve to the ambient source allowing nitrogen to flow through the source.
3. Open the software used to control the mass spectrometer. For this study, the software used was "SQ driver". Click on file -open- then select the appropriate tune file.
4. Apply all voltages and temperatures to the ambient source once the manual tune is opened. Each mass spectrometer will have its own software for this step. For the current study, once the manual tune is open, click the button "Source Voltage is off" and the button "All gas and heaters are off" to perform this task.
5. Observe the appearance of the flow exiting the nozzle with the schlieren apparatus on the view screen of the digital camera as the temperature increases. Observe the gas stream (see description in "Results" section) coming out of the end of nozzle. The gas stream can be viewed on the back of the camera, or it can be viewed directly on a LCD monitor.
6. Collect the image by either recording a video from the camera, or taking a picture of the gas stream, once desired images are visualized live on the camera.

7. Transfer the picture(s) collected to a computer with the camera memory card or USB connection and view the image with software of your choosing.

## 3. Determination of Spray Half Angle from a Collected Image

1. Open the collected image using an image viewing software and print out the collected image(s).
2. Draw a line on the printed image(s) defining the center axis of the gas stream parallel to the direction of flow using a ruler.
3. Draw a line along the edge of the visualized gas stream on the printed image(s) using a ruler. This can be visualized better from a recorded video due to a shimmer that is present in video format; use this to help identify the edge in the printed images. Mark the outer edges of the gas streams to obtain a range for the spray half angle.
4. Measure the angle produced between the center axis and the line drawn in 3.2 using a protractor.

## Representative Results

A schematic of the schlieren setup including the mass spectrometry ionization source can be found in **Figure 1**. When all schlieren components are properly aligned, gases within the test region can be seen as contrasting dark and light regions. **Figure 2** illustrates how this contrast can be used to observe how the shape of the nitrogen jet flow from the mass spectrometry source changes as nozzle size decreases.

A full, uncropped schlieren image of the source and gas flow can be found in **Figure 3**. This image illustrates the orientation of the test objects relative to the mirror. The image in **Figure 3** also shows what should be expected when the proper amount, approximately 50%, of light is cut off by the razor blade. If the cutoff is either too high (**Figure 4**), or too low (**Figure 5**), poor images will result.

Once the setup is complete, one can adjust various mass spectrometer parameters while watching their effect on the video screen of the camera. This image, alongside the actual signal of the mass spectrometer, allows for optimized conditions to be reached quickly due to the new understanding of the gas stream.

These images can then be used to calculate the spray half angle of the nitrogen stream. The spray half angle tells the user the overall size of the nitrogen gas stream. This angle is effected by diameter of the nozzle, as well as the pressure and temperature of the gas. **Figure 6** is a representation of the half angle measurements with constant nozzle size and variations in gas pressure. As expected, the half angle increases accordingly with an increase in pressure, signifying an overall size increase of the gas stream. **Figure 7** is a representation of the half angle with constant pressure while changing the nozzle diameter. As expected, the half angle increased with increased nozzle diameter. This signifies an overall scaling increase in size of the nitrogen jet coming out of the source as the nozzle diameter is increased.

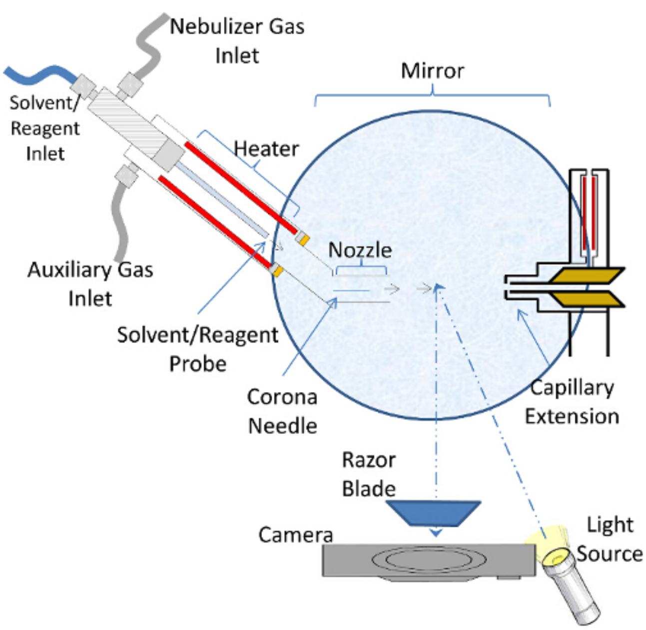


**Figure 1. Schlieren Schematic (re-print with permission from reference 7).** Schematic representation of the schlieren photography apparatus with the mass spectrometry ionization source. Please click here to view a larger version of this figure.

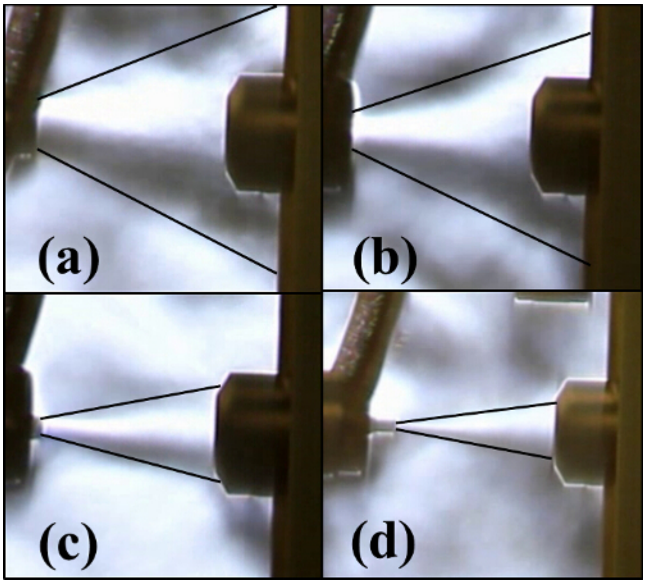


**Figure 2. Visualization of Nitrogen Streams (re-print with permission from reference 7).** Schlieren photographs of gas flow from the ionization source with different nozzle internal diameters of **(A)** 4.8 mm, **(B)** 3.2 mm, **(C)** 1.5 mm, **(D)** 0.5 mm. Please click here to view a larger version of this figure.

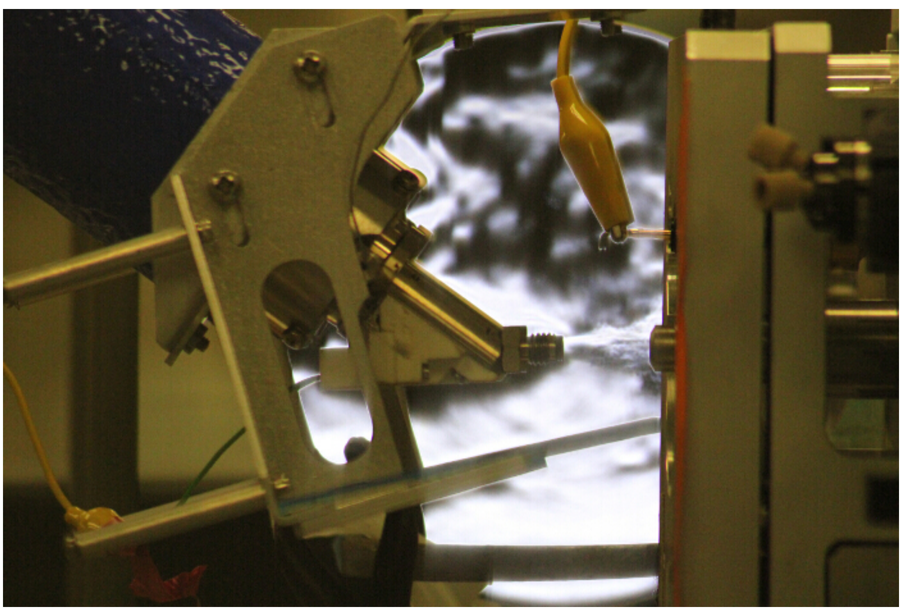

**Figure 3. Visualization of Ambient Source.** Wide angle schlieren photograph of the ionization source with proper positioning of the cutoff. Please click here to view a larger version of this figure.

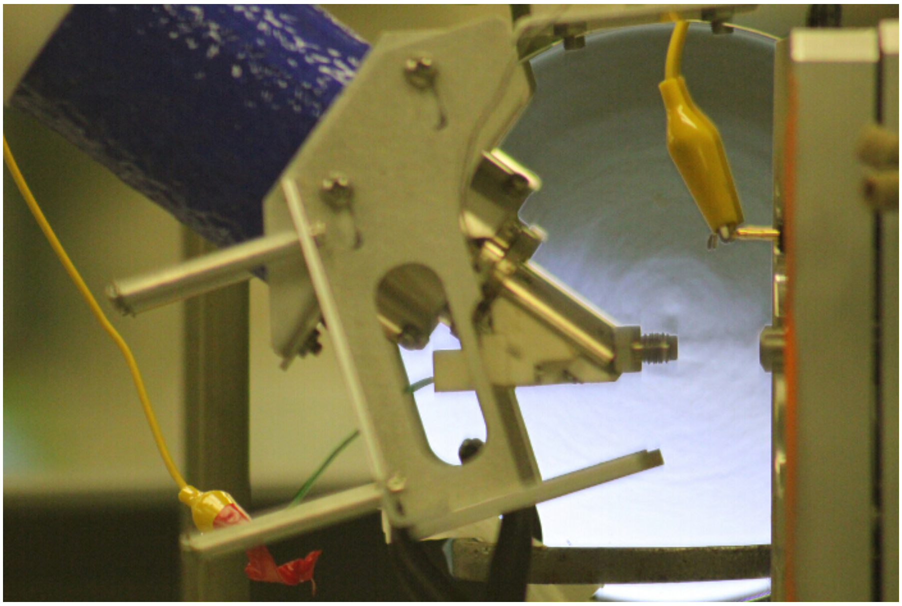

**Figure 4. Poor Visualization with Low Cutoff.** Schlieren photograph with the cutoff positioned too low. Please click here to view a larger version of this figure.

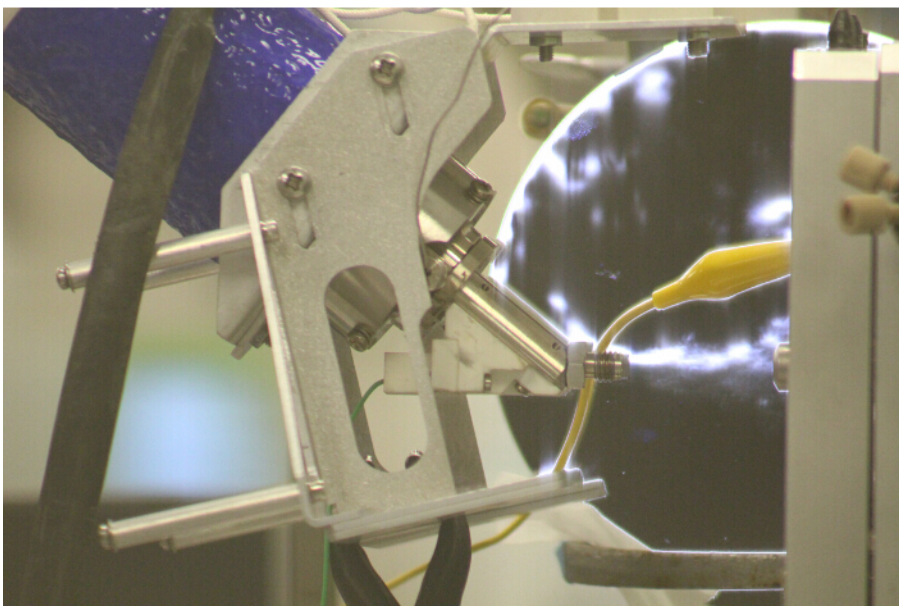

**Figure 5. Poor Visualization with High Cutoff.** Schlieren photograph with the cutoff positioned too high. Please click here to view a larger version of this figure.

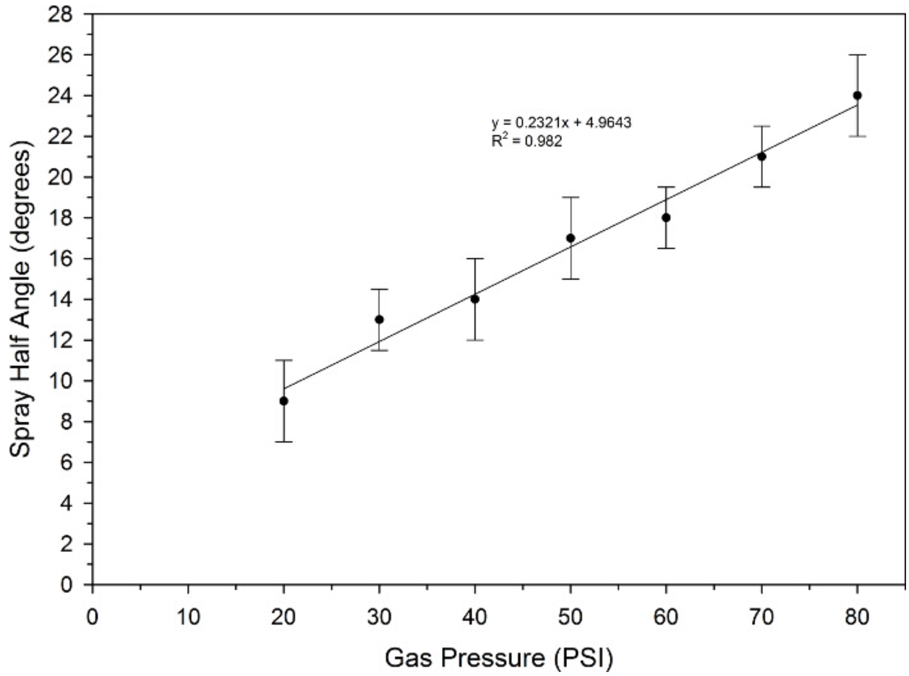


**Figure 6. Half Angle vs. Gas Pressure.** A graph depicting the change in the spray half angle with constant nozzle size with varying gas pressure. Please click here to view a larger version of this figure.

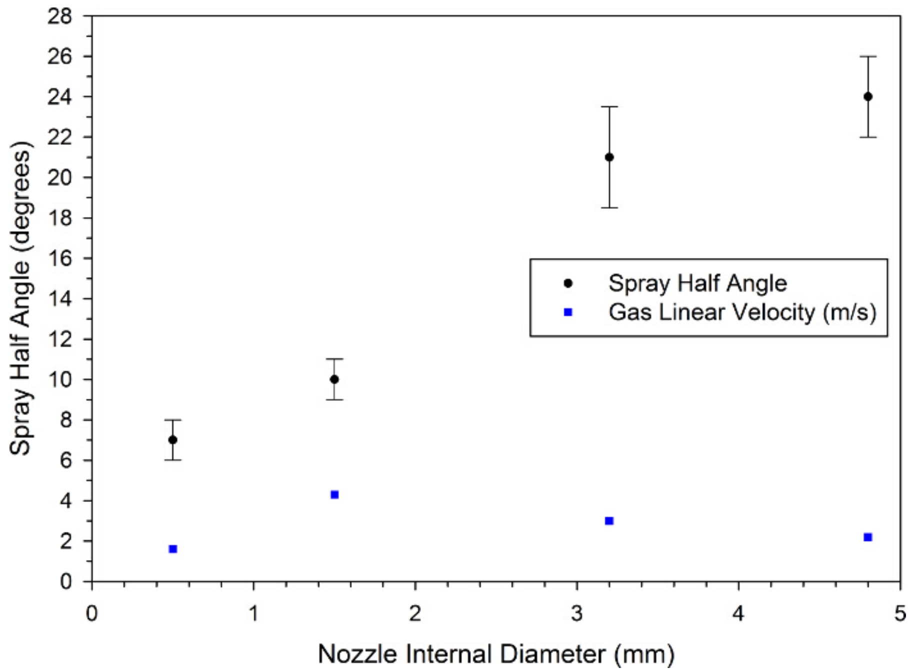


**Figure 7. Half Angle vs Nozzle Size.** A graph depicting the change in the spray half angle with constant pressure with varying nozzle size. Please click here to view a larger version of this figure.

## Discussion

There are several considerations which must be addressed prior to attempting this protocol. In addition to the space around the mass spectrometer for the source and mirror, enough open space must be available to accommodate the distance of twice the focal point of the mirror. Furthermore, the size of the mirror is ultimately decided by the size of the source that is under study. If the mirror is too small, the source will not be fully visualized. It is important to note that some, if not all, of the source covers must be removed to implement the schlieren photography imaging technique.

The most crucial steps of the actual setup are the alignment of each part of the schlieren apparatus. The mirror must be perpendicular to the floor and razor blade must be placed exactly at twice the focal length of the mirror. At this distance, the reflected light will be focused to a small spot. The amount of light blocked by the razor blade is also important. If poor images are produced, the first aspect to adjust would be with the placement of the razor blade. When the razor blade doesn't block enough of the light reaching the camera, no contrast is formed and thus the gas will not be seen. If too much of the light is blocked the images appear dark, making it difficult to distinguish the more subtle details in the nitrogen flow from the object under study.

A limitation of the technique is that there must be a large difference in terms of the refractive index of the background and the area of study. This will depend on temperature and humidity of the laboratory in question. RT nitrogen is ordinarily difficult to see as the background air is composed of approximately 78% nitrogen. This is overcome in the described setup because the temperature of the nitrogen varies from the source which results in changes in the refractive index.

Overall, the significant contribution of this protocol is the ability to understand the physical processes involved with the ionization within the source. This in turn will allow the user to better tune the instrument instead of blindly varying parameters, as well as provide reasoning for the optimized conditions. The advantage of this technique is the ability use all the information from both the physical and chemical processes to gain better sensitivity and selectivity with an ambient ionization source[6]. The user can utilize the schlieren images to determine physical properties of the source while the mass spectrometry data can be used to understand the chemical properties of the source.

Future applications would be to apply this technique to either various other ambient ionization sources available on the market, or a non-commercial apparatus. This can also be applied to any other instruments/machines which utilize gas streams.

## Disclosures

There are no competing financial interests with this article.

## Acknowledgements

The authors would like to acknowledge Caitlin Kowalewski for aiding in the editing and formatting of this publication.

## References

1. Settles, G.S., *Schlieren and Shadowgraph Techniques: Visualization Phenomena in Transparent Media (1sr ed.).* Springer-Verlag, Germany (2001).
2. Strawa, A. W., Chapman, G. T., Arnold, J. O., & Canning, T. N. Ballistic range and aerothermodynamic testing. *J. Aircraft.* **28** (7), 443-449 (1991).
3. Settles, G. S. Imaging gas leaks by using schlieren optics. *Pipeline & Gas Journal.* **226** (9), 28-30 (1999).
4. Takagi, T., & Kubota, H. The application of schlieren optics for detection of protein bands and other phenomena in polyacrylamide gel electrophoresis. *Electrophoresis.* **11** (5), 361-366 (1990).
5. Clark, I. G., Cruz, J. R., Huges, M. F., Ware, J. S., Madlangbayan, A., & Braun, R. D. Aerodynamic and Aeroelastic Characteristics of a Tension Cone Inflatable Aerodynamic Decelerator. *Proceedings from the 20th AIAA Aerodynamic Decelerator Systems Technology Conference and Seminar.* Seattle, May 7, (2009).
6. Froome, K. D. The Refractive Indices of Water Vapour, Air, Oxygen, Nitrogen and Argon at 72 kMc/s. *Proc. Phys. Soc. B.* **68**, 833-835 (1955).
7. Winter, G. T., Wilhide, J. A., & LaCourse, W. R. Characterization of a Direct Sample Analysis (DSA) Ambient Ionization Source. *J. Am. Soc. Mass Spectrom.* **26** (9), 1502-1507 (2015).
8. Pfeuffer, K. P., Schaper, J. N., *et al.* Halo-Shaped Flowing Atmospheric Pressure Afterglow: A Heavenly Design for Simplified Sample Introduction and Improved Ionization in Ambient Mass Spectrometry. *Anal. Chem.* , 7512-7518 (2013).
9. Pfeuffer, K. P., Shelley, J. T., Ray, S. J., & Hieftje, G. M. Visualization of Mass Transport and Heat Transfer in the FAPA Ambient Ionization Source. *J. Anal. At. Spectrom.* **28** (379-387) (2013).
10. Pfeuffer, K. P., Ray, S. J., & Hieftje, G. M. Measurement and Visualization of Mass Transport for the Flowing Atmospheric Pressure Afterglow (FAPA) Ambient Mass-Spectrometry Source. *J. Am. Soc. Mass Spectrom.* **25** (5), 800-808 (2014).
11. Keelor, J. D., Dwivedi, P., & Fernández, F. M. An Effective Approach for Coupling Direct Analysis in Real Time with Atmospheric Pressure Drift Tube Ion Mobility Spectrometry. *J. Am. Soc. Mass Spectrom.* **25** (9), 1538-1548 (2014).

 