Experimental Design for the Observation of Saliva Droplets Using a High-Speed Camera

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Abstract: Disease transmission is often caused by viruses or bacteria contained in salivary droplets. Salivary droplets are produced from breathing, coughing, and sneezing activities. The extent of droplet dispersal can determine the safe distance between individuals when interacting, many studies have taken a simulation and modeling approach, arguing that the risk of exposure to pathogens. In this study, we attempted to set up an instrument to experimentally observe the flow of saliva droplets emitted during sneezing using a High-Speed Camera (HSC) model Phantom T-1340 without using a light sheet. Additionally, this study did not use lasers for the comfort and safety of individuals during recording of sneezing phenomena. The observation results explain that the sneezing phenomenon occurs within a time frame of 300 ms. The number of droplets observed was 246 with an observation probability of only 66%. The saliva fluid emitted is considered as large particles (bulk droplets) that undergo refraction due to two main factors, namely external and internal factors. External factors are influenced by environmental air flow rate, humidity, and temperature. Internal factors refer to the contents present in saliva such as water, protein, enzymes, and mucus or mucin.

Keywords: Droplets; HSC; PIV; Saliva; Sneezing

Introduction

Contagious diseases such as Tuberculosis (TB), Pneumonia, Middle East Respiratory Syndrome (MERS), Severe Acute Respiratory Syndrome (SARS), and Coronavirus Disease 19 (COVID-19) (Casanova et al., 2010; Induri et al., 2021; Shih et al., 2007) are caused by bacterial and viral infections such as Mycobacterium Tuberculosis (Pai et al., 2016), Streptococcus Pneumoniae, and Syndrome Coronavirus 2 (Borro et al., 2021; Morcatty et al., 2021). The transmission of these viruses or bacteria generally occurs through pathogens contained in droplets that undergo a phase change from liquid to gas that mixes with the air (oxygen) present in the air. Droplets are fluids or water droplets that come out of a person through the respiratory tract. These droplets are produced by patients when they talk, cough, and sneeze (Ho, 2021; Maehata et al., 2021; Viola et al., 2021). The size of the droplets emitted varies from 5µm to 100 µm (Bi, 2018; Yin, 2009). The speed of the droplets also varies, where the speed of the droplets is influenced by several factors including the pressure generated from the nasopharynx and gastritis. Additionally, factors that affect the speed of droplets include their size and mass (Bahl et al., 2021; Basu, 2021; Dbouk & Drikakis, 2020).

Several previous studies have reported on the mechanism of droplet formation when coughing or sneezing (Nishimura et al., 2013). It has been concluded that saliva droplets originating from sneezing are the primary indicators of pathogen transmission from one individual to another. The World Health Organization (WHO) defines that within a 1 meter distance, there is a risk of pathogen transmission through respiratory, coughing, or sneezing (Vadlamudi S K Thirumalaikumaran Chakraborty Dipshikha Saha

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Abhishek Basu Saptarshi, 1 C.E.; World Health Organization (WHO), 2020). However, research conducted by Bahl et al (2020) suggests that virus transmission in hospitals can occur at distances of up to 4 meters which differs from the WHO guidelines. Therefore, research on droplet dynamics serves as a key indicator for determining policies in controlling the spread of contagious pathogens, especially in the field of healthcare. Previous studies have used modeling techniques to analyze the properties of droplets, which play a crucial role in determining the rate of droplet spread.

Research related to the direction of droplet dispersion, flow velocity, and estimation of saliva droplet size has often utilized Computational Fluid Dynamics (CFD) simulations since the COVID-19 pandemic in 2020 (Faleiros et al., 2022; Mohamadi & Fazeli, 2022; Zoka et al., 2021). The choice of using CFD methods aims to minimize the risk of pathogen transmission or infection to observers or researchers. Furthermore, the selection of CFD as an alternative method is due to the difficulties in directly involving human subjects for various reasons, such as the potential transmission of pathogens contained within droplets or aerosols (Leuken et al., 2016). Aerosols produced by coughs form complex clouds with numerous interactions influenced by the environment (Bourouiba et al., 2014a). Several factors affecting droplet flow rate include airflow distribution, temperature, and air humidity (Al-Safran, 2021). Hence, to assess the risk of infection spread or pathogen transmission, it is essential to study the flow of droplets released by an individual, particularly during sneezing. The regulation and control of disease transmission originating from respiratory droplets rely on calculating a safe distance based on the area of droplet dispersion during respiration, which is grounded in aerobiological studies (Leuken et al., 2016; WHO, 2020).

Modeling is not sufficient to find out the droplet flow rate. Therefore, experiments on the droplet flow dynamics are also needed to support the modeling research on this subject. In most of the literature we have reviewed, high-speed cameras, light sheets, and lasers are used to track the motion of droplet flow released by individuals during sneezing. However, in our study, we used a High-Speed Camera (HSC), specifically the Phantom Model T-1340 equipped with a Nikkor 18-55 mm Macro lens to observe droplet flow. They used light sourced from LED lamps of the CSVITEC Multiled G8 brand for illumination. The LED lamps had a divergence angle of 150 degrees, with the LED intensity set at 85%. The subject was positioned in front of a black background to enhance the clarity of the main object during both the recording and video processing phases. The HSC resolution used was 2048 x 1952 pixels, with a frame rate of 1000 frames per second. If we model the X-Z plane as perpendicular to each other, the positions of the subject, camera, and LED are shown in Figure 1.

![Figure 1. Subject Position, HSC, and Source Light](image)

In the process of capturing images under air conditioning, considerations such as air temperature and humidity were not factored in. The camera was set up 60 cm away from the subject, angled approximately 80 degrees from the light source, aiming at the subject's mouth. This setup was designed to balance light properties in fluids. The fluid is seeded with tracer particles which, for sufficiently small particles, are assumed to faithfully follow the flow dynamics (the degree to which the particles faithfully follow the flow is represented by the Stokes number). The motion of the seeding particles is used to calculate speed and direction (the velocity field) of the flow being studied. We conducted our study with volunteers in good health. The use of human samples in this research was approved by the Research Ethics Committee of Health (KEPPKN Registration Number: 1171012P) at the Faculty of Medicine, Syiah Kuala University, Aceh, Indonesia. Our research is unique in that we used an experimental setup to observe the flow of saliva droplets during sneezing and employed specific instrumentation for the study.

**Method**

**Set-up Instrument**

This research used a High-Speed Camera (HSC) of the Phantom Model T-1340 equipped with a Nikkor 18-55 mm Macro lens to observe droplet flow. They used light sourced from LED lamps of the CSVITEC Multiled G8 brand for illumination. The LED lamps had a divergence angle of 150 degrees, with the LED intensity set at 85%. The subject was positioned in front of a black background to enhance the clarity of the main object during both the recording and video processing phases. The HSC resolution used was 2048 x 1952 pixels, with a frame rate of 1000 frames per second. If we model the X-Z plane as perpendicular to each other, the positions of the subject, camera, and LED are shown in Figure 1.
scattering and perspective distortion, as discussed in previous studies (Bahl et al., 2020). However, a disadvantage of this camera positioning is the tilt that occurs between the plane of focus for observation and the plane of the resulting image. To mitigate this, an f-stop setting of f4 was used to keep all droplet particles within the plane of focus. To induce sneezing, dry tissues were inserted into the subject’s nasal cavity. Tissues were chosen over pepper powder to minimize irritation to the subject, a decision that was ethically approved.

**Image Processing and Analysis**

HSC recordings with extension (.cine) are analyzed using matlab software R2021b. Two main stages are performed, namely pre processing and post processing. The pre processing stage includes converting (.cine) to (.mp4) format then extracting video frames to image with (.tiff) format. The next step is to convolve RGB images to grayscale and continue with thresholding into binary images. Calibration of measurements is done by setting-scaling at a resolution of 2048 x 1536 where at that resolution 1 cm of actual measurement is represented by 50 dot pixels. To obtain the exact coordinate position of the pixel coordinates of observation, calibration is performed on 25 x 25 pixels at a distance of 5 mm and applied to the entire image sequence. To stabilize the movement of the recorded object, normalization is performed using a 2D cross-correlation algorithm. The correlation results are applied to the entire image sequence. The post-processing stage is performed to calculate the number and distribution of droplet sizes that are dispersed, while to analyze the droplet velocity using the Particle Image Velocimetry (PIV) method. The flow of image processing steps is shown in Figure 2.

**Result and Discussion**

The experiment successfully tracked the path of saliva droplets during a sneeze using a High-Speed Camera (HSC) set at 1000 frames per second. The total duration of the observation was 1140 milliseconds, with the sneeze itself happening within a quick 300 milliseconds. This is in line with earlier studies that estimate a sneeze to last between 200 and 400 milliseconds (Bahl et al., 2020; Bourouiba et al., 2014b; Han et al., 2021). The movement of droplets during the sneeze is show in Figure 3.
Figure 3 presents the progression of droplet movement at three specific instances: 400 ms, 450 ms, and 500 ms. The images demonstrate the refraction phenomenon in saliva as it is projected from the subject’s mouth at intervals of 50 ms. Figure 3.a captures the initial scattering of droplets from the mouth, seen as individual droplets. Figures 3.b and 3.c depict the expulsion and fragmentation of saliva fluid that coats the mouth’s edge due to pressure from the nasopharynx (Wu et al., 2020). During the sneezing event, it was observed that the average number of droplets emitted was 246. This is represented in Figure 4, which also shows a total Probability Density Function (PDF) of 66%.

This reduction could be attributed to the decreasing pressure in the nasopharynx as the sneeze event unfolds, preventing saliva from being ejected or broken apart. It might also suggest that the detected droplets are moving into the later phases of the sneeze event. Additionally, due to the limited aperture of the focus lens, it’s possible that not all droplets are captured in the field of view during recording, thereby reducing the chances of droplet detection in each frame.

Saliva droplets are formed and freely dispersed in the air due to hydrodynamic instability and the viscoelastic properties of saliva. In simpler terms, the released saliva, viewed as a large particle or bulk droplet, undergoes refraction due to two primary factors: external and internal. External factors include the speed of the surrounding airflow, humidity, and temperature. Despite these factors being overlooked in this study, our vector flow visualization of droplets (as seen in Figure 4) suggests that they cannot be disregarded. The internal factor pertains to the released saliva itself, specifically its components like water, protein, enzymes, and mucus or mucin. These elements influence the viscosity of the released saliva. The thickness of the saliva influences how strongly the particles stick to it, which affects how well the particle bonds can resist force or change in shape.
The movement of saliva droplets from a sneeze at 150 ms is shown in Figure 5(a). The red lines indicate the direction and speed of the droplets. The droplets act like spheres that rise freely in the air. Some of the droplets split into smaller ones at a point called bifurcation, where the flow pattern changes (Veldhuis & Biesheuvel, 2007). This phenomenon is influenced by the density ratio of saliva droplets (Horowitz & Williamson, 2010). So that the impact of this density causes complex changes in droplet particles, including irregular changes in shape, mass, and flow direction. When saliva droplet particles are at a certain density (critical density), then the particles start to move up freely. Particles that have a denser density remain on a relatively stable path and move downward at an angle (Albert et al., 2015). Environmental parameters are external factors such as temperature, humidity, temperature, and air flow velocity. We assume that these environmental parameters significantly influence the internal processes within the saliva droplet.

The information from Figure 5.b shows that the droplet velocity vector ranges from 0.5 to 3.9. The highest velocity is at 150 ms. This is assumed to happen when the nasopharynx has the maximum pressure, and then the droplet velocity decreases over time (Han et al., 2021; Kwon et al., 2012).

Conclusion

This research used the particle image velocimetry (PIV) method to observe the movement of saliva droplets that originate from sneezing. The PIV technique allowed for accurate analysis of the movement of saliva droplets. The study found that the sneezing phenomenon occurs within a time interval of 300 ms. The number of droplets observed was 246 with a probability of observation of only 66%. The saliva fluid that is released is considered as a large particle (bulk droplet) that undergoes refraction due to two main factors, namely external and internal factors. External factors are influenced by the speed of the ambient air flow, humidity, and temperature. Internal factors refer to the content contained in saliva such as water, protein, enzymes, and mucus or mucus. The saliva droplets that are released undergo bifurcation. The phenomenon is influenced by the density ratio of saliva droplets, so that the impact of the density causes complex changes in droplet particles that include changes in shape that are not fixed, mass, and flow direction.

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Author Contributions
For research articles with several authors, a short paragraph specifying their individual contributions must be provided. The following statements should be used “Conceptualization, Hamdani Umar, Samsul Rizal, Rachmad Almi Putra; methodology Maimun Syukri; software, M Salamul Fajar Sabri”. All authors have read and agreed to the published version of the manuscript.

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Conflicts of Interest
Declare conflicts of interest or state “The authors declare no conflict of interest.”

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