

## Introduction

The COVID-19 pandemic along with wildfires have sparked an urgent need for improved guidelines regarding face mask use. The United States Centers for Disease Prevention and Control recommends reuse of face masks to conserve supplies for essential workers. Although previous studies have demonstrated the protective nature of wearing masks to limit the dispersal of exhaled respiratory droplets from the wearer to other individuals, there remains the potential to facemask-wearers of inhaling particles generated from the mask itself. The goal of this project is to examine whether inhalable face mask debris is present for new or mechanically aged masks using scanning electron microscopy (SEM) and Raman Spectroscopy. To determine their biological effects, particles collected from these masks were instilled intranasally in mice. Pulmonary function testing was performed, as well as bronchoalveolar lavage to determine if facemask debris particles alter the physiologic function of the lungs and/or produce inflammatory changes in the lungs of mice.

## Experimental Design

### Particle Collection

1. Prefilter deionized water through a 0.4 $\mu$ m polycarbonate membrane filter.
2. Sonicate N95, surgical, and fabric masks individually with 300mL of prefiltered deionized water.
3. Filter the sonicated solution through a 0.8 $\mu$ m polycarbonate membrane filter.

### Particle Analysis

#### Scanning Electron Microscopy

1. Image acquisition.
2. 100 random particle counts to categorize particles by size and morphology.

#### Raman Spectroscopy

1. Acquire the reference spectrum for each mask layer.
2. Determine if the particle spectrum matches with the reference spectrum.

### Mouse Exposure

1. 25 $\mu$ L intranasal administration of particle suspension (1 $\mu$ g/ $\mu$ L) in C57 mice for each type of mask on 3 consecutive days.
2. Pulmonary function test with methacholine challenge.
3. Bronchoalveolar lavage to determine total cell count and cell differential.

## Results

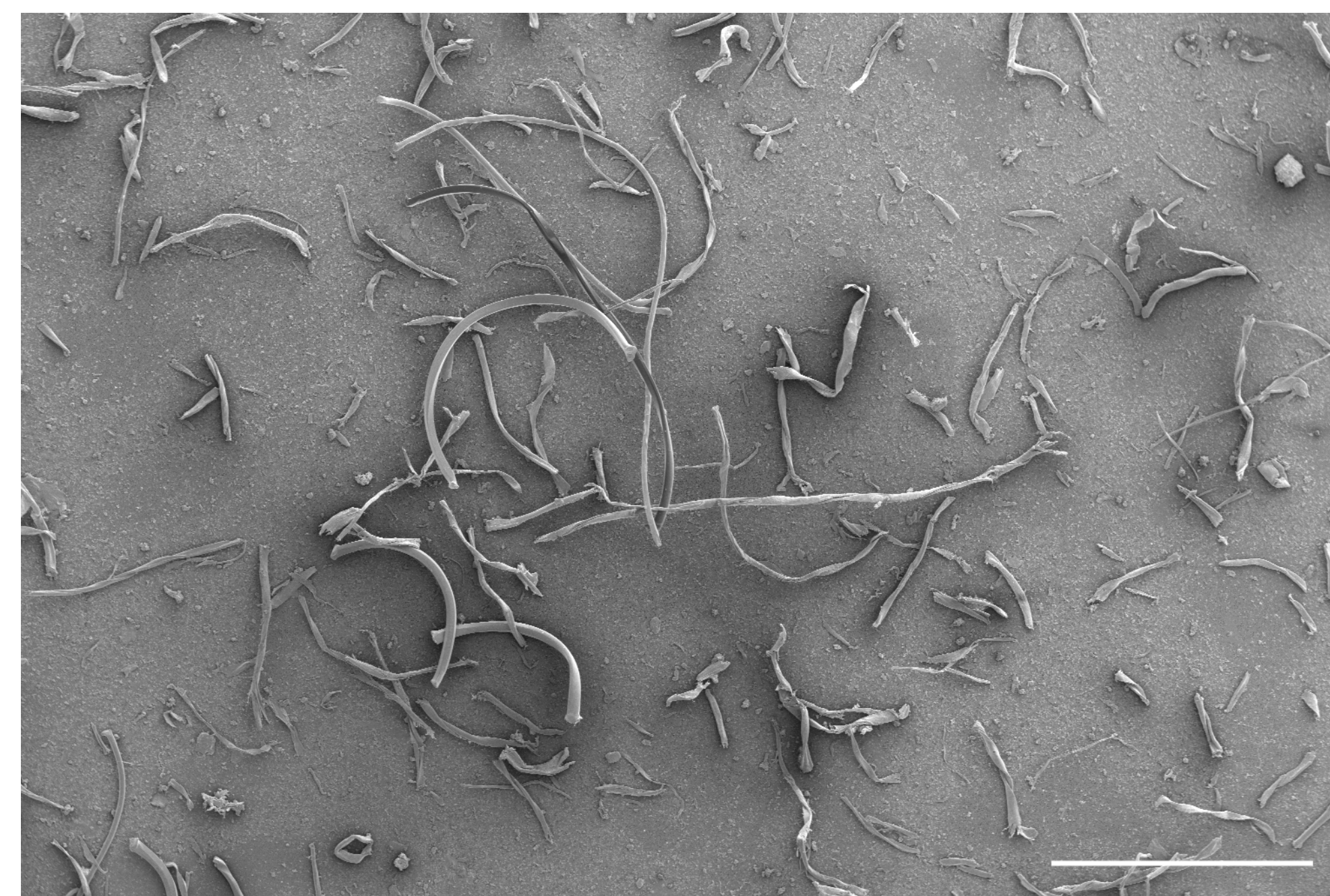


Figure 1. SEM image of particles from a new fabric mask. The scale bar represents 500 $\mu$ m.

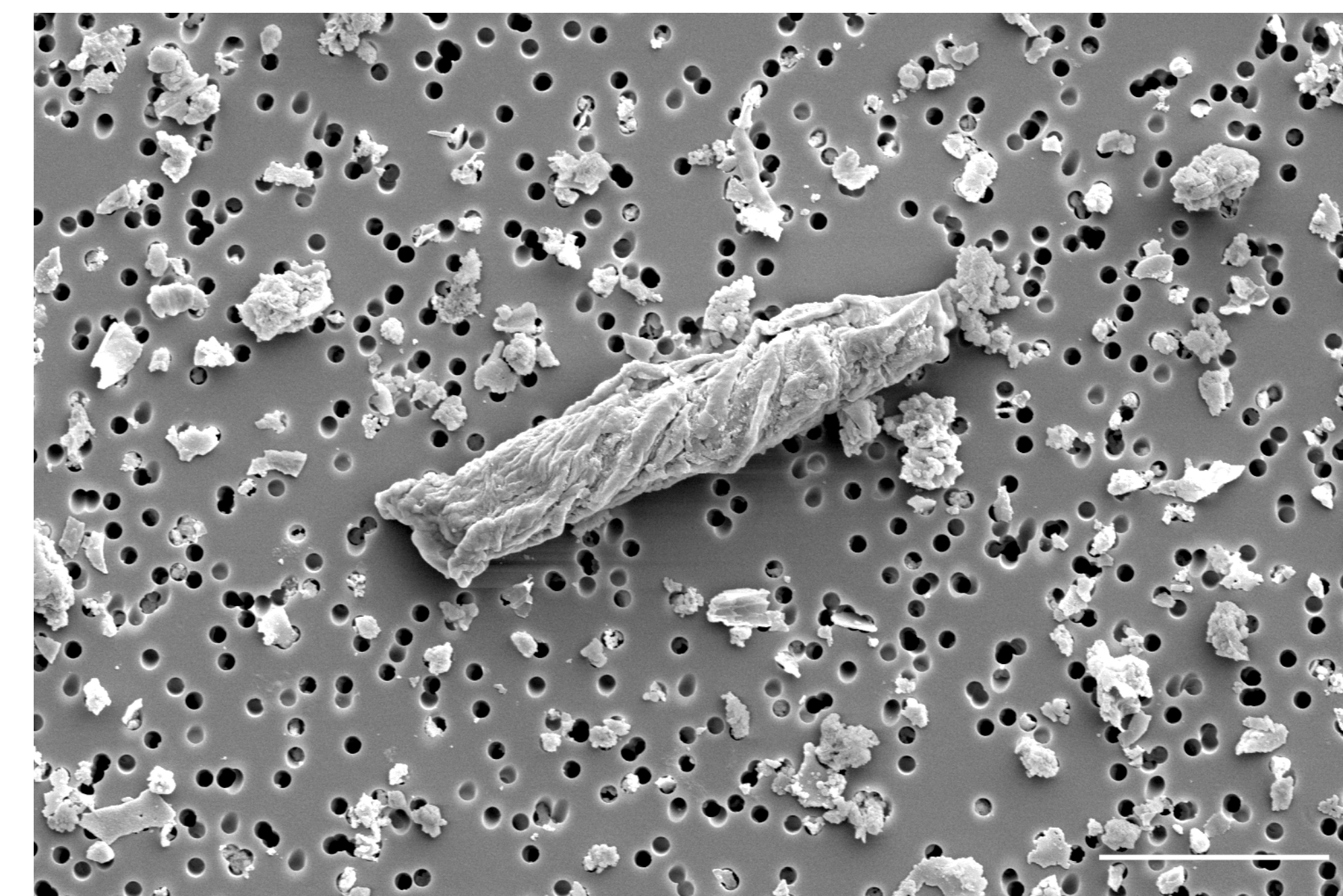


Figure 2. SEM image of particles from an aged N95 mask. The scale bar represents 10 $\mu$ m.

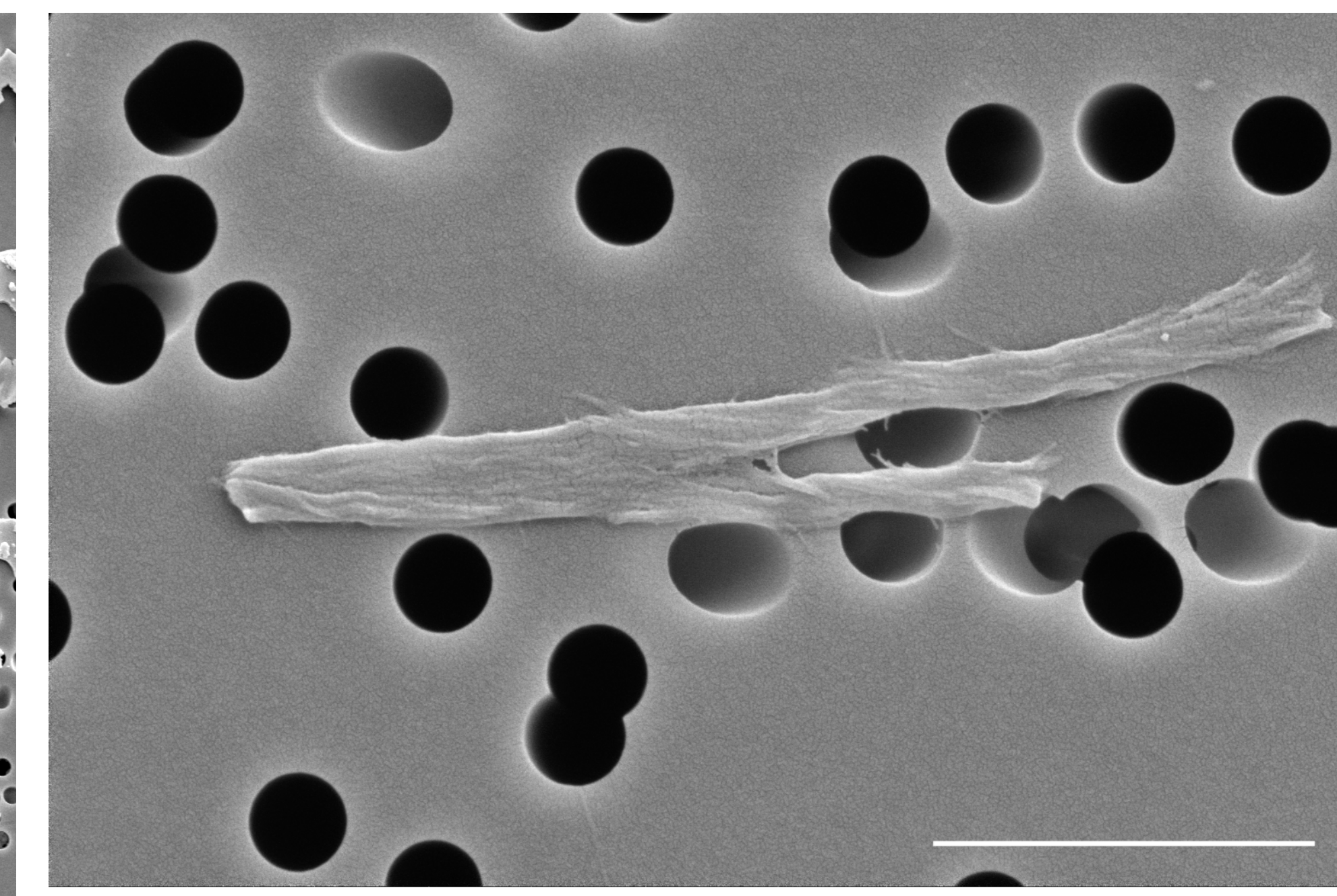


Figure 3. SEM image of a fiber from a new N95 mask. The scale bar represents 3 $\mu$ m.

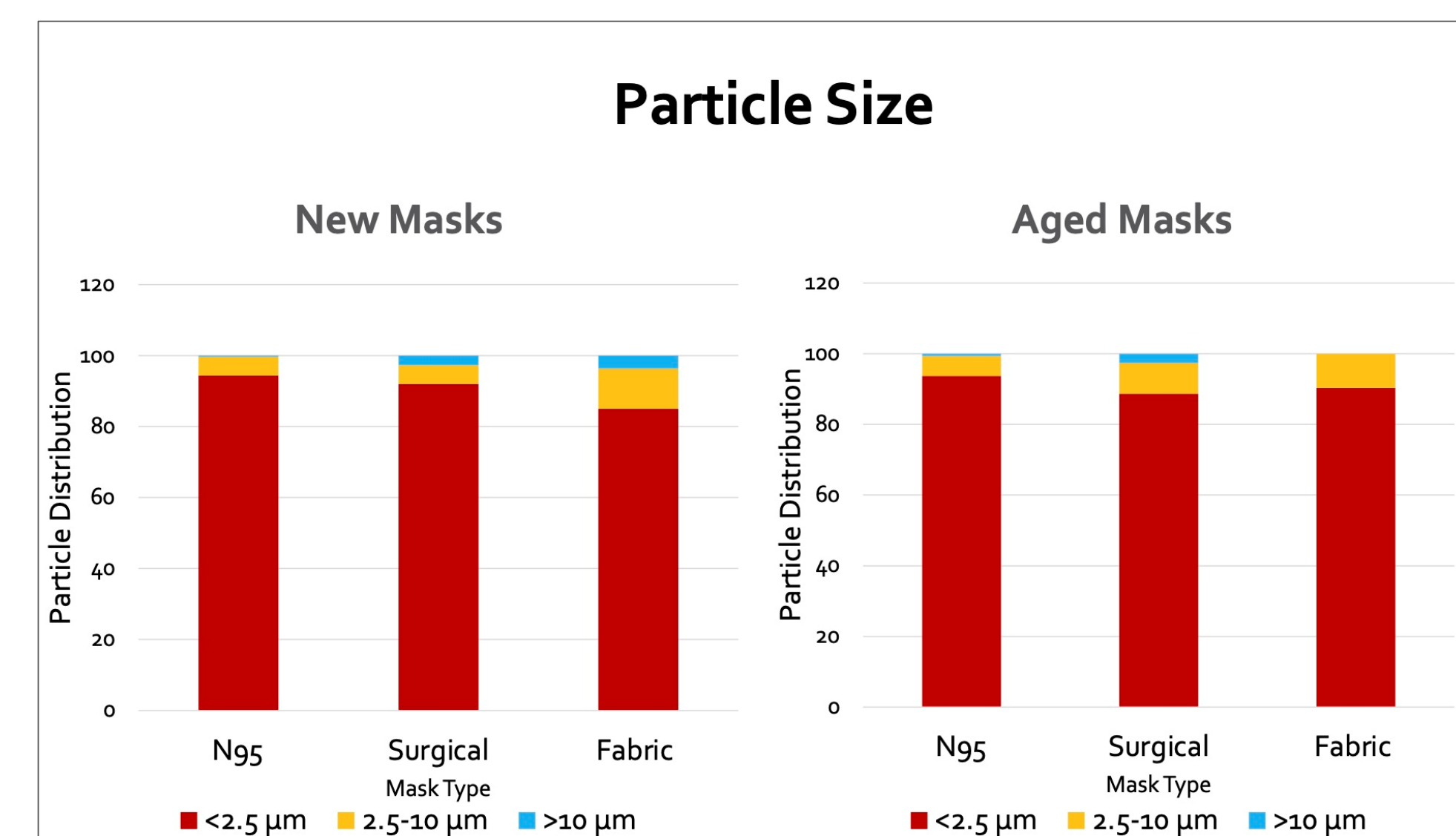


Figure 4. Particle size distribution of new and aged masks. Each bar represents the findings from three masks for each mask category.

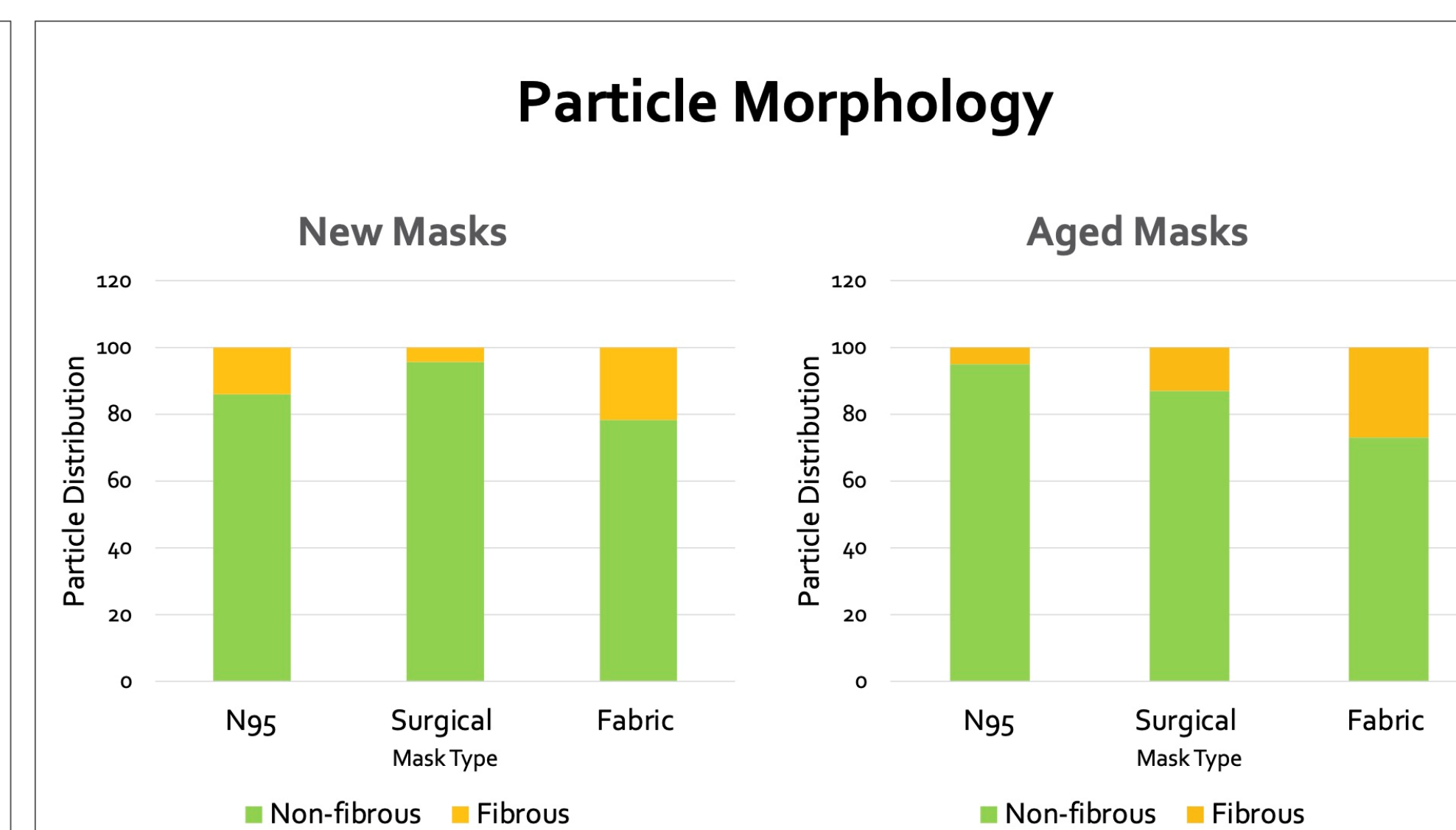


Figure 5. Particle morphology distribution of new and aged masks. Each bar represents the findings from three masks for each mask category.

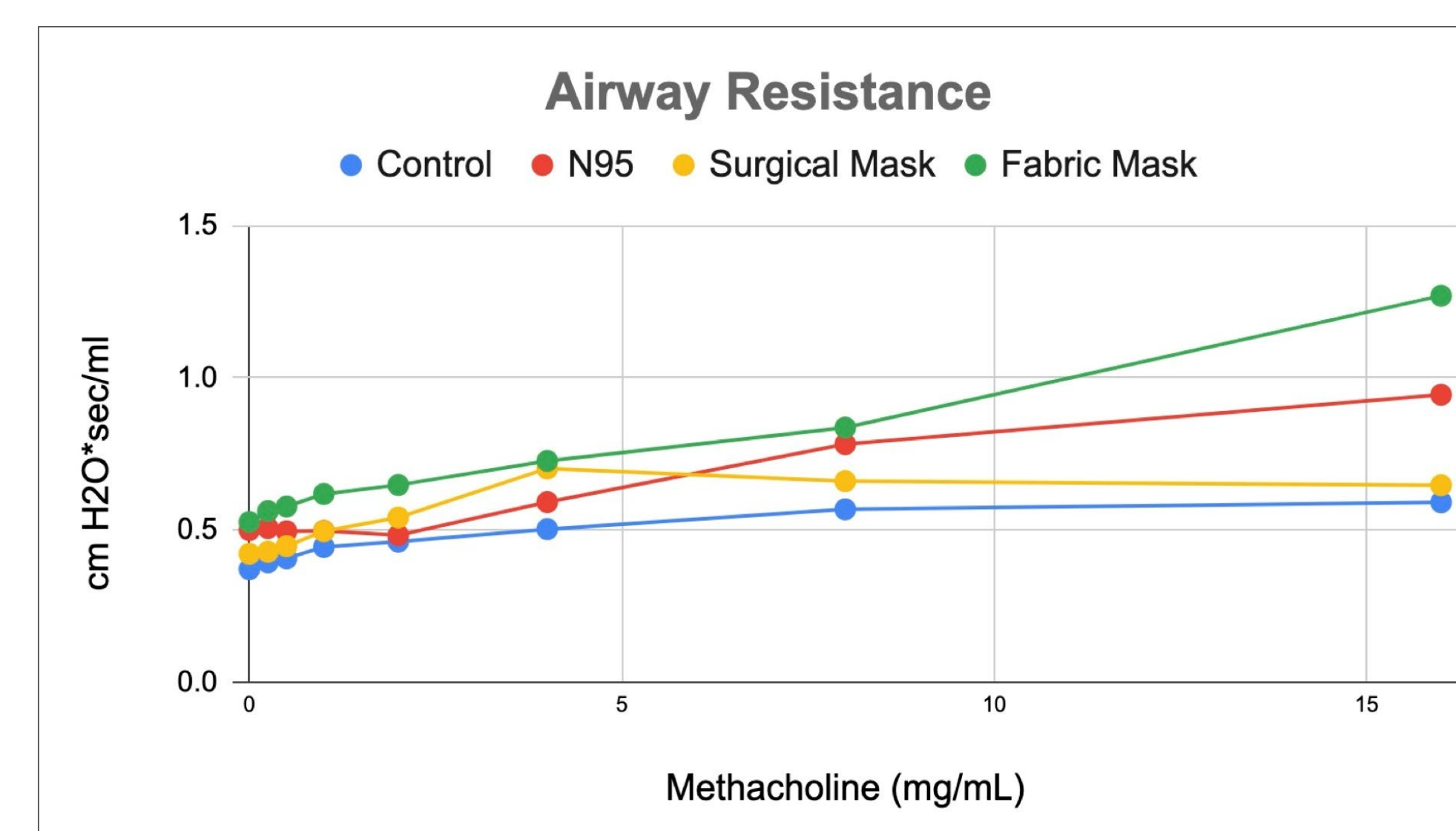


Figure 6. Methacholine induced airway resistance. (n = 5 mice / treatment group).

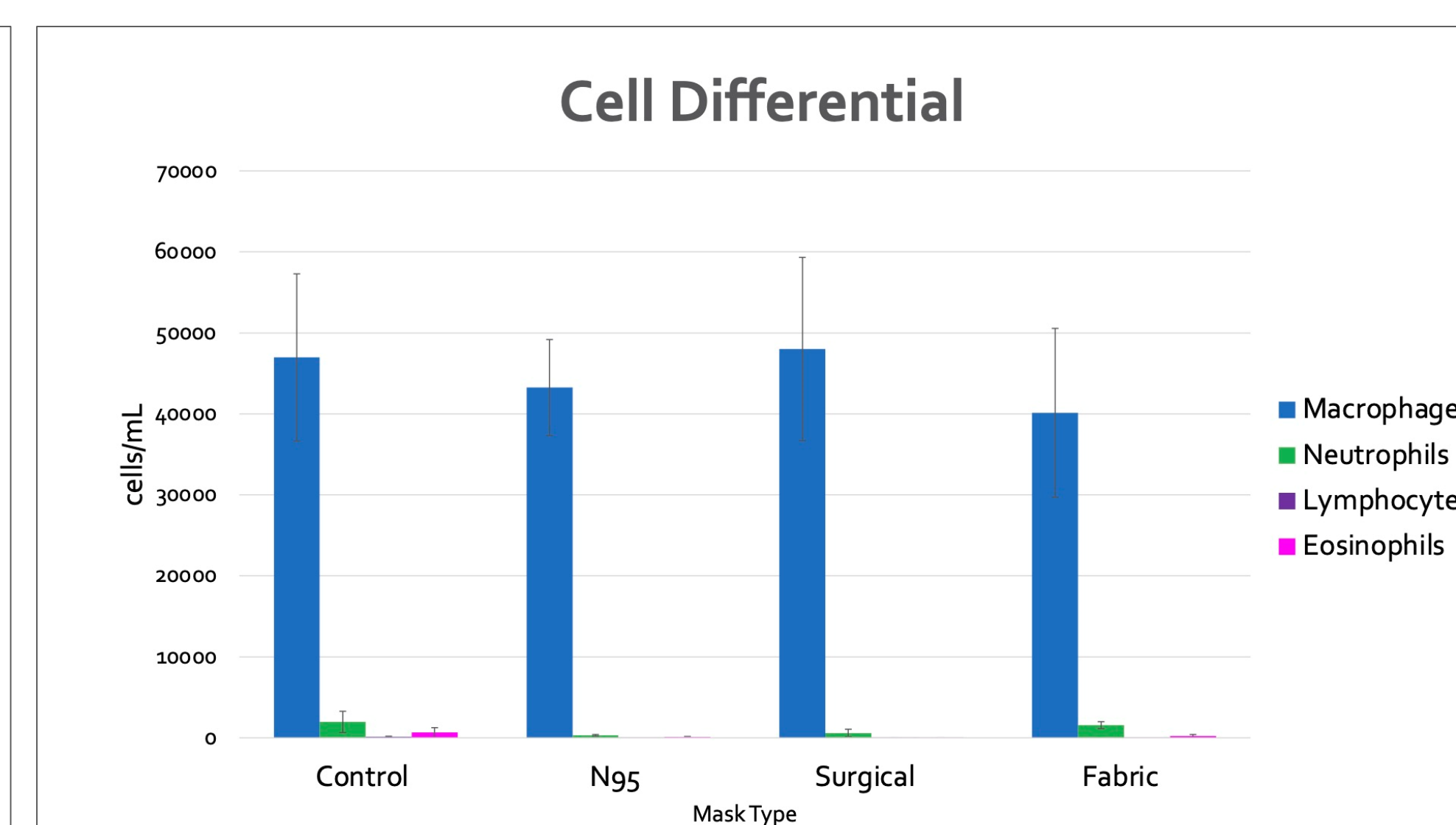


Figure 7. Cell differentials by bronchoalveolar lavage. (n = 5 mice / treatment group).

## Conclusions

From sonicated new and aged **N95, SURGICAL, and FABRIC MASKS**, we found:

- Greater than 90% of particles released from all masks were inhalable (<10 $\mu$ m).
- N95 masks generated the greatest proportion of respirable particles (<2.5 $\mu$ m).
- Fabric masks generated the greatest proportion of inhalable fibrous particles. (aerodynamic diameter <2.5 $\mu$ m)
- Repeated intranasal exposure of mice to face mask debris did not result in a significant difference in airway resistance following methacholine challenge compared to sham controls.
- No significant differences were noted in total cells or the cell differential recovered by bronchoalveolar lavage.
- Despite the presence of inhalable mask debris, no clear respiratory toxicity was noted in mice.

## Acknowledgements

Financial support was provided by the UC Davis SVM Students Training in Advanced Research (STAR) Program and NIH grant 5T35OD010956-22 and the CDC/NIOSH grant U54 OH07550 (Western Center for Agricultural Health & Safety) Rapid Response Grant.