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## Histopathology assay of the lung after intratracheal injection of SARS-CoV-2 spike protein recombinant in mice: A preliminary study

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COVID-19 can cause ARDS, characterized by Diffuse alveolar Damage (DAD) generated by cell death, surfactants, and proteins to the alveolar. When the hyaline membrane accumulates or thickens, oxygen exchange in the alveoli is disrupted. Recent research employing animal model mice has been designed and carried out on various methods that could be used to investigate the mechanisms of inflammation and infection in COVID-19 disease. In this study, we injected SARS-CoV-2 spike recombinant protein into the trachea to make an animal model for studying DAD and infiltration with a histological assay. Methods: To initiate mouse models, an incision is made in above the trachea, the muscles and glands above the trachea are moved. A 26-gauge needle delivered 15 µg of SARS-CoV-2 recombinant spike protein (SC-2 RSP) in 50 µl saline, followed by a 100 µl air injection. The control group was given 50 µl saline intratracheally. Afterward, mice were euthanized with intraperitoneal injections of ketamine and xylazine at 1, 2, and 7-days postinjection (d.p.i.) to harvest the lungs. Hematoxylin & Eosin were used to stain lung tissue for histological examinations. Result: Lung histopathology of BALB/c mice injected with SARS-CoV-2 recombinant spike protein at 1-, 2-, and 7-days post instillation showed mild immune cell infiltration. Cell infiltration was also found at alveolar, perivascular, and peribronchiolar locations compared with untreated mice. According to our study, SARS-CoV-2 recombinant spike protein has been shown to trigger mild lung inflammation in BALB/c mice.