The Long Term Effects of SARS-COV-2

Abstract:

In 2020 COVID-19 shut down the whole world. Since then, cases have steadily been dropping to a point where now, in 2023, the world is completely back to normal. However, this return to normality does not mean that COVID-19 is still not affecting our society today, as questions about its long-term effects still remain unanswered. Fortunately, due to COVID-19's global reach, there are many publicly available data sets that can be analyzed to find possible hints at the long-term effects of COVID-19. Therefore, we decided to analyze former COVID-19 patients' tissue samples using gene ontology techniques to identify specific proteins that COVID-19 had affected, which could be a cause for the lasting effects of COVID-19.

Introduction:

On December 12, 2019, a group of people in China's city of Wuhan began to experience flu-like symptoms, including fever, fatigue, dry cough, and gastrointestinal issues. Even though no one knew at the time, these symptoms were the first signs of the Sars-Cov-2(COVID-19) outbreak. As the outbreak progressed and the virus began to spread globally, scientists began to understand the symptoms of COVID-19 more and more. However, unexplainable symptoms emerged from the virus as well, including a loss of smell and/or taste that nearly twenty-five percent of all patients diagnosed with COVID-19 noted. Furthermore, other serious symptoms like seizures were reported by a small group of patients (Burke, 2020). These symptoms led scientists to explore COVID-19's effect on the human brain. So far, many studies have shown that COVID-19 has been linked to abnormalities in the brain. However, it is still unclear what causes these abnormalities and whether or not contracting COVID-19 will have long-lasting consequences. This led us to look deeper into the one critical organ already proven to be

affected by COVID-19, the lung, and ask if any genes are linked to long-term effects like post-acute sequelae of COVID-19(Long COVID).

Literature Review:

The larger scientific community has already established that the human lung is a key organ that COVID-19 affects. Additionally, common symptoms such as fever, pneumonia, and a cough are often caused by the disease. Finally, the diseases have been relatively fatal, with roughly a two to three percent fatality rate. However, 31.7% of all people who contract the disease will visit the intensive care unit (Wen-Lin Su et al., 2021). In terms of the mechanism of COVID-19 on the lungs, COVID-19 is packaged in a spike protein which is used to attach itself to the ACE2 receptor of its host cell. In humans, these ACE2 proteins are located abundantly in the repertory tract. After the cell is bound to the ACE2 protein, it is able to start replicating itself. This series of events allows for the initiation of a "cytokine storm," which is a hallmark of acute respiratory distress syndrome, a major reason for death due to COVID-19 (Forchette et al., 2021). Finally, while the long-term effects of COVID-19 are still largely unknown, a phenomenon known as long COVID or post-acute sequelae of COVID-19 has been reported by an estimated ten percent of all COVID-19 cases meaning that around 65 million people have the syndrome. While the cause for this phenomenon is unknown, there are several leading hypotheses, including persistent reservoirs of COVID-19 in tissues and immune system deregulation. Additionally, long COVID has many symptoms ranging from serious cognitive impairment(brain fog), which is compared to nearly ten years of cognitive aging, to heart, lung, liver, kidney, pancreas, or spleen damage. Finally, long COVID currently does not have any treatment, and more research is needed to understand and treat the syndrome fully.

Methods and Results:

In order to obtain reliable data without doing our own lab testing, we chose a premade data set of 24 patients who have died from COVID-19(N et al., 2020). We then exported the data set into Matlab and created a volcano plot that visualized all the data in a Log2 ratio. The data we were showing in the table shows the upregulation and downregulation of a specific gene. We then narrowed down the results by setting a minimum p-value of .01, a minimum fold change value of 4, and by only using data specific to the lung(Figure 1). By doing this, we created a list of genes that COVID-19 up-regulates. We then plugged this list into a GO enrichment analysis which shows how the specific gene up-regulations affected the body. We sorted this generated list in two different ways. The first was putting the results with the highest False Discovery Rates(FDR) at the top and the lowest at the bottom(Table 1). We did this in order to filter out false positives. Next, we sorted the list, from highest to lowest, by Fold Enrichment in order to find what didn't happen due to just random chance(Table 2). Using these two sorts, we compiled a list of pathways that were likely upregulated by the infection, and using the gene ontology website, we were able to see what genes caused these upregulations. The pathway with the largest Fold Enrichment Value, Spontaneous Neurotransmitter Secretion, is associated with the protein Synaptotagmin which is connected to senses like smell and sight. Additionally, when Synaptotagmin is upregulated, neurotransmitter activity and release are increased due to the Synaptagmin being a calcium sensor on synapses.

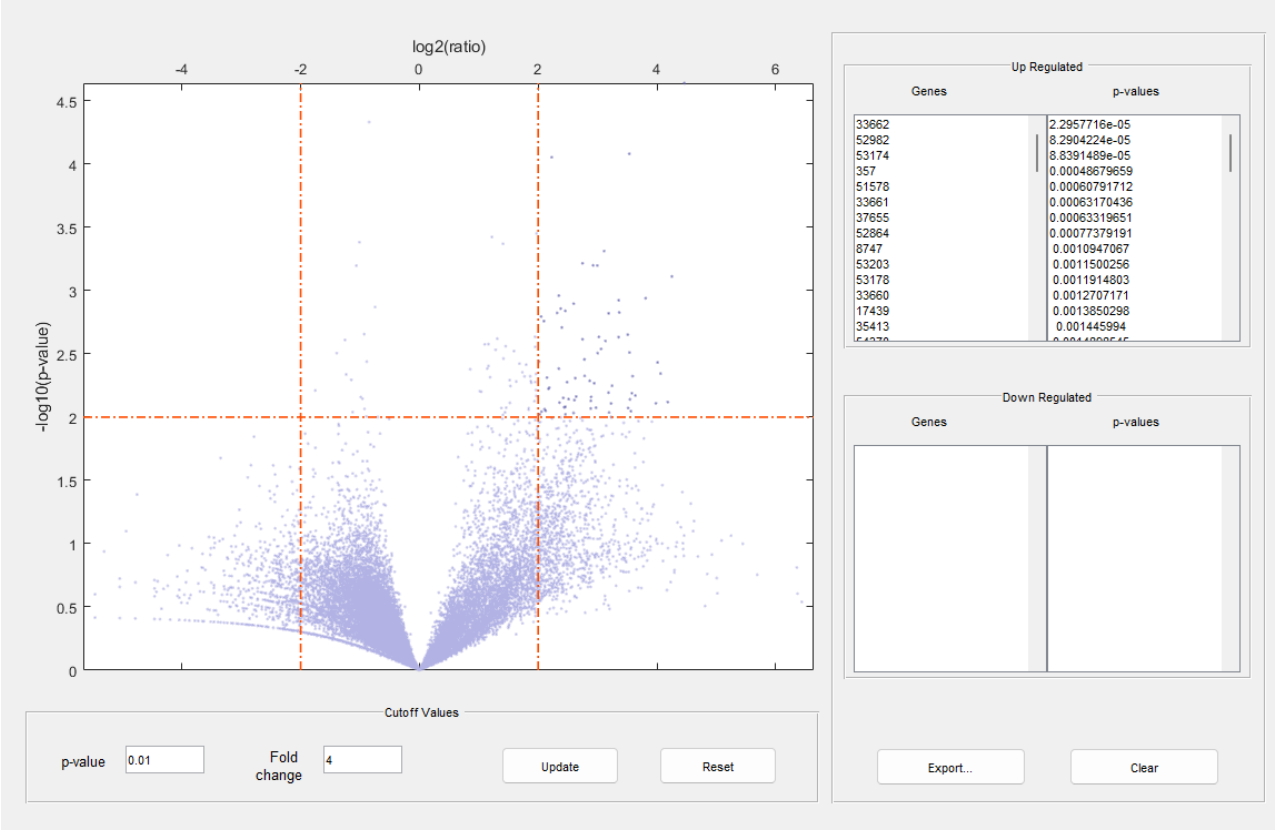

Figure 1. Volcano plot of DEGs of human lungs infected with SARS-COV2 shows a large number of upregulated genes in COVID infection. Genes defined as differentially expressed met the conditions of: adjusted p-value < .01, fold change > 4. Samples taken from 24 infected patients, relative to 3 control patients. Experimental data obtained from (N et al., 2020)

Analysis Type:	PANTHER Overrepresentation Test (Released 20221013)
Annotation Version and Release Date:	GO Ontology database DOI: 10.5281/zenodo.6799722 Released 2022-07-01
Analyzed List:	upload_1 (Homo sapiens)

Reference List:	Homo sapiens (all genes in database)
Test Type:	FISHER
Correction:	FDR
GO biological process complete	Fold Enrichment
spontaneous neurotransmitter secretion (GO:0061669)	**17.15**
calcium ion-regulated exocytosis of neurotransmitter (GO:0048791)	10.72
embryonic digestive tract morphogenesis (GO:0048557)	8.83
ligand-gated ion channel signaling pathway (GO:1990806)	8.08
ionotropic glutamate receptor signaling pathway (GO:0035235)	8.08
phagocytosis, recognition (GO:0006910)	7.01
glutamate receptor signaling pathway (GO:0007215)	6.52
B cell receptor signaling pathway (GO:0050853)	6.51
regulation of dopamine secretion (GO:0014059)	6.49
complement activation, classical pathway (GO:0006958)	6.2
phagocytosis, engulfment (GO:0006911)	6.14
cellular response to DNA damage stimulus (GO:0006974)	0.28
detection of chemical stimulus (GO:0009593)	0.23
RNA processing (GO:0006396)	0.21
detection of chemical stimulus involved in sensory perception (GO:0050907)	0.18
sensory perception of smell (GO:0007608)	0.13
translation (GO:0006412)	0.08
generation of precursor metabolites and energy (GO:0006091)	0.07
ncRNA processing (GO:0034470)	0.07
detection of chemical stimulus involved in sensory perception of smell (GO:0050911)	0.07
ribonucleoprotein complex biogenesis (GO:0022613)	0.07
energy derivation by oxidation of organic compounds (GO:0015980)	< 0.01

Table 1: Data table obtained through GO enrichment analysis(Version 2022-07-01). Results from highest to lowest with respect to Fold Enrichment Values.

GO biological process complete	upload_1 (FDR)
spontaneous neurotransmitter secretion (GO:0061669)	**2.91E-02**
energy derivation by oxidation of organic compounds (GO:0015980)	2.98E-02
developmental process (GO:0032502)	3.08E-02

protein modification by small protein conjugation or removal (GO:0070647)	3.33E-02
regulation of nucleic acid-templated transcription (GO:1903506)	3.71E-02
embryonic organ morphogenesis (GO:0048562)	3.75E-02
excitatory postsynaptic potential (GO:0060079)	3.93E-02
regulated exocytosis (GO:0045055)	3.93E-02
locomotion (GO:0040011)	3.94E-02
mRNA metabolic process (GO:0016071)	4.03E-02
response to chemokine (GO:1990868)	4.15E-02
modulation of chemical synaptic transmission (GO:0050804)	4.15E-02
regulation of DNA-templated transcription (GO:0006355)	4.17E-02
regulation of trans-synaptic signaling (GO:0099177)	4.17E-02
cellular response to chemokine (GO:1990869)	4.18E-02
export from cell (GO:0140352)	4.18E-02
import into cell (GO:0098657)	4.19E-02
regulation of transmembrane transport (GO:0034762)	4.48E-02
lymphocyte chemotaxis (GO:0048247)	4.59E-02
regulation of synaptic transmission, glutamatergic (GO:0051966)	4.70E-02
cellular component biogenesis (GO:0044085)	4.80E-02
catabolic process (GO:0009056)	4.81E-02
regulation of immune response (GO:0050776)	4.87E-02
positive regulation of calcium ion transmembrane transport (GO:1904427)	4.89E-02
secretion (GO:0046903)	4.90E-02
embryonic digestive tract morphogenesis (GO:0048557)	4.91E-02
cellular response to stress (GO:0033554)	4.93E-02
nucleobase-containing small molecule metabolic process (GO:0055086)	4.95E-02
synaptic vesicle cycle (GO:0099504)	4.95E-02

Table 2: Data table obtained through GO enrichment analysis(Version 2022-07-01). Results from lowest to highest with respect to FDR.

Discussion:

My findings agree with the existing literature because it has already been shown that there is an activation of the immune system in the lungs when a person is infected with COVID-19 (Yapici-Eser et al., 2021). However, the knowledge of activation in the immune system due to COVID-19 is very limited; thus, more research is necessary (Guillon et al., 2020).

Therefore, the discovery of Synaptotagmin in these pathways could be monumental and could be the cause of loss of smell functions in COVID-19 patients. Loss of smell in COVID-19 patients has been heavily studied as an early diagnosis of COVID-19 and a symptom that could be used to reduce the possibility of spreading the virus to others during the virus' contagious stage (Santos et al., 2021). Additionally, in the data, one of the pathways Synaptotagmin is detected in is the spontaneous neurotransmitter secretion pathway. This pathway is responsible for the release of neurotransmitters from neurons and, when upregulated, can alter neural networks and synaptic plasticity, which both play a role in long-term changes in neural activity. The possibility of long-term effects due to the spontaneous neurotransmitter secretion pathway in combination with Synaptotagmin, which is linked to loss of smell and sometimes even seizures due to increased neurotransmitter release, creates a worrying sign for the long-term effects of COVID-19.

Conclusion:

In investigating tissue samples of former COVID-19 patients using tools like gene ontology, we discovered possible target proteins, like Synaptotagmin, that COVID-19 upregulates during infection that could lead to long-term effects such as seizures. This discovery could be monumental as these target proteins could be targeted by future drugs in order to treat the effects of COVID-19. In the future, we hope that our findings will be used when developing new drugs to treat long COVID-19. However, there is still alot of room for more research and experimentation as, due to a lack of available lab space, we were unable to complete hands-on experiments, which would have allowed us to take this research further.

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