HIGH SCHOOL EDITION
Journal of Student Research
Volume 12 Issue 3 (2023)

Identifying the Relationship between Microglia and Suicide: Analyzing Microglial Densities in Suicide and Non-Suicide Brains in Deceased Patients

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ABSTRACT

Suicide is one of the worst outcomes that can occur in clinical psychiatry. This issue affects 700,000 people globally every year. While currently no cure exists to stop suicide completely, recent research has led to new discoveries in the field which has improved the knowledge and awareness regarding the issue. Recent findings have shown an increase in microglia activation (innate immune cells) in the brains of suicide victims. This research paper seeks to quantify this trend by analyzing microglial densities in the white matter and dorsal raphe nucleus regions of the brain using post-morterm human brains from suicide victims and controls. This paper seeks to answer the question if microglial densities in the white matter and dorsal raphe nucleus regions of the brain are increased in suicide victims. This papers purpose is to help create a wider understanding on the role of microglial densities in suicide victims.

Introduction

Suicide is a major public health crisis that stems from psychiatrical factors in the brain. According to the World Health Organization, over 700,000 people die by suicide on an average every year (World Health Organization [WHO] 2021). A majority of people who die by suicide have mental health disorders. However, not all people who have mental health disorders die by commiting suicide. This suggests that external factors might play a role in facilitating suicide. As a result, this topic has been studied vigorously by psychiatrists and psychologists over the years. While no cure has been found to stop suicide, creating awarness and producing research about this topic is acutely needed for reducing the number of deaths. Although health professionals can determine if a person is at risk for suicide based on their psychological health, it still important to understand suicide from biological side. This information offers an enhanced perspective into what is occurring in the brain specifically by identifying differences in the brains characteristics between suicide victims and healthy people (Goncalves de Andrade et al., 2022). Recent advances in psychiatry have shown a correlation between suicide and microglial activation (in microglia cells). Microglial cells are known to play a crucial role in the fight against the invading pathogens, which generate the release of immune response signals.This can ultimately create dysregulation and lead to neuroinflammation in the brain (Goncalves de Andrade et al., 2022). Currently there is not enough research to understand the specific role on how microglia cells act in suicide, such as whether they are only associated in suicide, or whether they related to the development of psychiatrical diseases (Suzuki et al.,2019). This paper will investigate the role of microglia in suicide by observing microglial densities in post-mortem brain samples.

Microglia Cell Biology

In order to understand this research paper, it is first important to understand the biology of the microglial cells. Identified by Spanish neuroanatomist Pio del Rio Hortega, the microglial cells are innate immune cells mostly prevalent in the human body's Central Nervous System (CNS). The CNS is part of the nervous system, consisting of the spinal cord and brain (Suzuki et al.,2019). Microglial cells can be located in other parts of the brain in varying amounts and densities. Microglial cells are derived from the primitive hematopoietic cells (cells in the bloodstream), which play a crucial role in maintaining the brain's homeostasis - regulation of the energy supply. These cells also originate in the embryonic yolk-sac (structure developed in the uterus) and then migrate into the neural tube. These cells reproduce through cell division with the process occurring as the fetus develops into a mature form in the womb (Lannes et al., 2017). Upon complete development microglial cells account for 10% of cells in the brain, regulating brain development and function (Sharma et al., 2021).

The cell body of the microglia contains a nucleus, as well as organelles such as the mitochondria and endoplasmic reticulum. Furthermore, the cytoplasm of the microglia is involved with the production of proteins and enzymes involved with immune response. In their resting state, microglia have a small cell body, typically oval shaped. This physical characteristic helps the microglia continuously monitor their environment in search of invaders (Sharma et al., 2021).

Scope of Current Microglial Research

Cutting edge research has shown that microglial activation, the resulting cytokine production and the increased neuroinflamation in the CNS are observed in the victims of suicide. However limited research is available regarding these topics. Much of this stems from complexities on whether to analyze post-mortem human microglia samples or not. Analyzing human samples can prove to be difficult to research due to the ethical, and biological complexities as procedures require consent to ensure the privacy and rights of scientific samples. Yet studies regarding human microglia do exist and this research has created groundbreaking discoveries in the suicide field. However the need for more research regarding specific characteristics of microglial cells in suicide victims exists to create further replicable data on the relationship between microglia and suicide (Suzuki et al.,2019).

Microglia Activation and Steps

Before delving deeper into specific characteristics of the microglia it is important to understand how microglial activation in the brain and leads to suicide.

In healthy people's brains, the maintenance of the brain environment is based on immune receptors and signals such as CSF1R (transmits intracellular signals), extracellular signal regulated kinases, scavenger, ApoER2 and TAM receptors. **(Refer to Figure 1)** These receptors keep the function and stability of the Microglia in check. Furthermore, microglia cells also have chemokine receptors that regulate their location in the Central Nervous System, which in turn create stability in microglial cells. Microglia cells are highly dynamic and can undergo rapid changes. In microglial activation, the cells undergo functional, and morphological changes in response to external stimuli. Usually such stimuli are foreign invaders and pathogens. This process is exemplified when the microglia, affected by invading pathogens, are activated resulting in an extension of their cell body, and heightened speed allowing them to move towards the invading pathogens. **(Refer to Figure 2)** As a result the immune receptors are activated and produced in higher quantities. Overall the the main goal of this process is to help the microglia protect itself however the effects of microglial activation can increase suicide risk. This correlation is seen in two key ways: increased cytokines production and neuroinflammation in the CNS (Lannes et al., 2017).

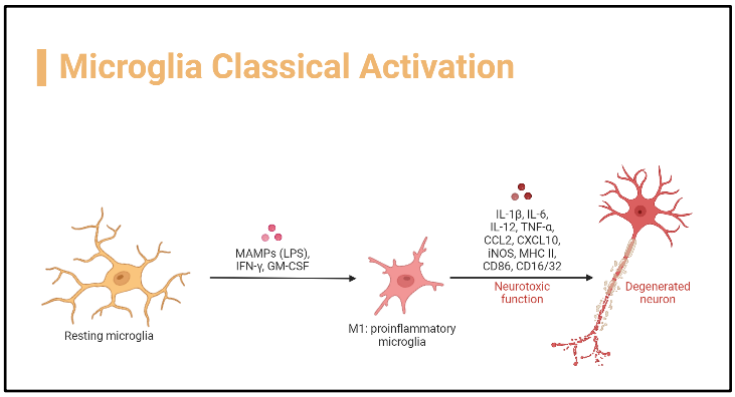

Figure 1. This image shows the steps that occur during microglial activation, and how the microglia cell attacks an invading pathogen. Created and copyrighted by Tanish Joshi-Apte

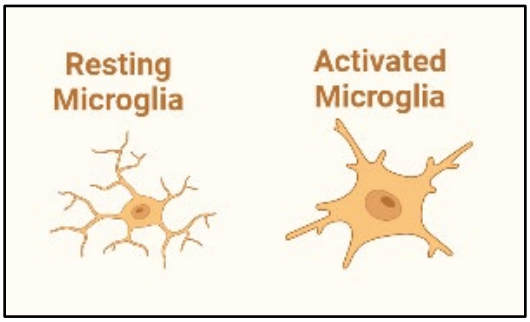

Figure 2. This image shows the difference between a resting microglia cell compared to an activated microglia cell. Note the increased size in an activated microglia cell. Created and copyrighted by Tanish Joshi-Apte.

Cytokines

Cytokines are small proteins important in cell signaling and function that are produced by the microglia. Upon microglial activation, cytokines are produced in order to aid in regulating brain function, immune response, and cell signaling. Usually the microglia produces various cytokines to respond to various stimuli such as IL-1 and IL-6. Upon activation the cytokines can recruit and incorporate other immune cells to the infection site to help treat it. As a result cytokines have pattern recognition receptors (PRRs) that detect pathogens or damaged tissues. However there is increased evidence that dysregulated and increased cytokines production can contribute to the heightened development of psychiatric disorders. Ultimately this combination of heighted cytokine production can destroy neural networks and neurons in the brain creating increased risk for suicide. This risk has been displayed in studies that have shown that suicide victims have increased amounts of IL-1 and IL-6 in their bloodstream and blood cell walls. The elevated IL-6 levels is one of the most prominent findings in suicide victims and has been groundbreaking for further research into the role of cytokines (Brites & Fernandes, 2015).

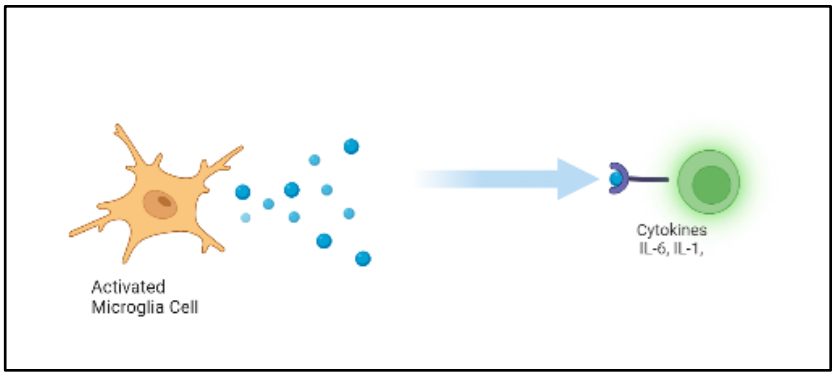

Figure 3. This image displays the process of cytokines IL-6 and IL-1 being released from the activated microglia in order to fight against the attacking pathogen. Created and Copyrighted by Tanish Joshi-Apte.

Neuroinflammation

Neuroinflammation is a process characterized by dysregulated amounts of cytokines, chemokines, and reactive oxygen species in the CNS. Similarly, recent research demonstrates a link between the activation of the microglia and neuroinflammation. If neuroinflammation is detected, the microglia is activated and is equipped with toll-like-receptors (TLR) to defend the Central Nervous System from attacking pathogens. Psychological triggers such as stress, anxiety, or depression can cause the microglia to become activated. Microglian activation is seen with a rapid and heightened change in cell shape, behavior, and structure. Furthermore neuro-inflamation can also create suicidal behavior due to the alteration of the neurotransmitters. Neurotransmitters carry chemical signals from one neuron to the next. In the microglia specifically, neurotransmitters can work moderately or release the amount of cytokines needed to create an immune response (DiSabato et al,. 2017). If there is a decrease in the function of neurotransmitters, this can lead to oxidative stress and synaptic plasticity. This trend has been shown with individuals dying by suicide having increased neuro-inflamation in the CNS specifically in regions such as the prefrontal cortex (Brites & Fernandes, 2015).

Microglial Density

Microglial density refers to the number of microglia cells present in a specific area of a brain. The number of microglial cells can vary across different brain regions and is usually influenced by age and sex of a person. Microglial density can be measured in the brain using tools such as sterology and immunohistochemistry which quantify the cell count. Furthermore, microglial activation can also affect the cell density, causing the microglia to have to defend itself in response to an invading pathogen. While the relationship between between microglial activation and suicide is clear, understanding the role of microglial density and suicide is not.

Methodology

The primary goal of this paper was to gather data regarding microglia cell density in suicide victims to understand the connection between suicide and microglia. This method was done by comparing the microglial cell density of suicide victims compared to non-suicide victims, to determine if there was difference in size, and density of the cells. This paper specifically looked at data microglial density since currently limited analysis and literature regarding microglial density has been available, and the conclusions put forth from this paper could create a new understanding and and conclusions about understanding the role of microglial in suicide, which could create new and improved opportunities for future research in the role of microglia in suicide.

The initial hypothesis in this paper was that that a greater microglial density would be found in the white matter and dorsal raphe nucleus regions of the brain in suicide victims compared to non-suicide victims and/or healthy control groups. This hypothesis was based on the fact that previous research had established a relationship between microglial activation and suicide with increased microglial cell growth being a symptom of microglial activation. In order to test this hypothesis, this paper used the data gathered from relevant scholarly works which focused on testing the microglial density post-mortem human microglia samples. In order to narrow down the scope of the hypothesis, a select criteria was established to select the samples used for this paper. The first criteria was to include data from both male and female brain samples, with an age of above 18 years of all brain samples being analyzed. No specific race and gender was required for the data being analyzed. The inclusion of all races, ages greater than 18 years and all genders increased the scope and relevance of the conclusions since it can be applied to a wider population size. Next, all data used in this paper was dated less than 10 years from May 2023, this is to ensure that the data used in this paper is relevant to the current knowledge of microglial density in suicide victims. Since this paper contains data from human brains, the next criteria was to ensure that all ethical guidelines and approvals were followed and seeked when testing brain samples. This is to ensure that the data used in this paper conforms to certain ethical standards. Due to the limited research available on this topic, microglial samples were only analyzed from white matter in the brain and the dorsal raphe nucleus, this is because most microglial cells are found and are prominent in these two regions. Microglial samples from other regions of the brain would have been preferred to create replicable results. Although microglial density can vary across different parts of the brain ultimately the main goal of this paper was to understand whether there is a difference in size of microglia cells, between suicide victims and healthy patients and not to measure the density of healthy microglial cells.

Microglia in White Matter Region

Prefrontal White Matter

The white matter in the brain refers to the tissues in the brain composed of neurons, the name is derived from the whitish color of the tissues. The white matter in the brain helps transmit electrical signals between the brain and the spinal cords. This process is done by the use of axions which create pathways to transmit signals. As a result, the role of white matter is important in cognitive function. The structure of the tissues in white matter is composed of various axions which are surrounded by a fatty substance called myelin. Myelin acts as an insulating layer helping to facilitate the rapid electrical signals taking place. Disruptions in the white matter can trigger an increased risk for suicide, however the current research has been inconclusive when drawing out specific trends.

In order to evaluate microglial density this paper analyzed the data from the prefrontal white matter region in victims of suicide and healthy controls. This study analyzed brain tissue samples within 24 hours of death selected from the New York State Psychiatric Institute. The study group focused on 11 suicide victims and 25 individuals whose cause of death was not suicide. It is important to note that both groups included individuals with various psychiatric diseases and not one specific one. After collection of the brain tissues, the left hemisphere of the brain was sliced and stored at four degree celsius, with two sections being processed for testing. First to determine the microglial characteristics and to identify white matter in the brain immunostaining was performed. Immunostaining is the process that uses different markers to visualize specific components of the cells and brain. The study specifically used two markers called CD68-immunoreactive, and an ionized calcium binding marker.

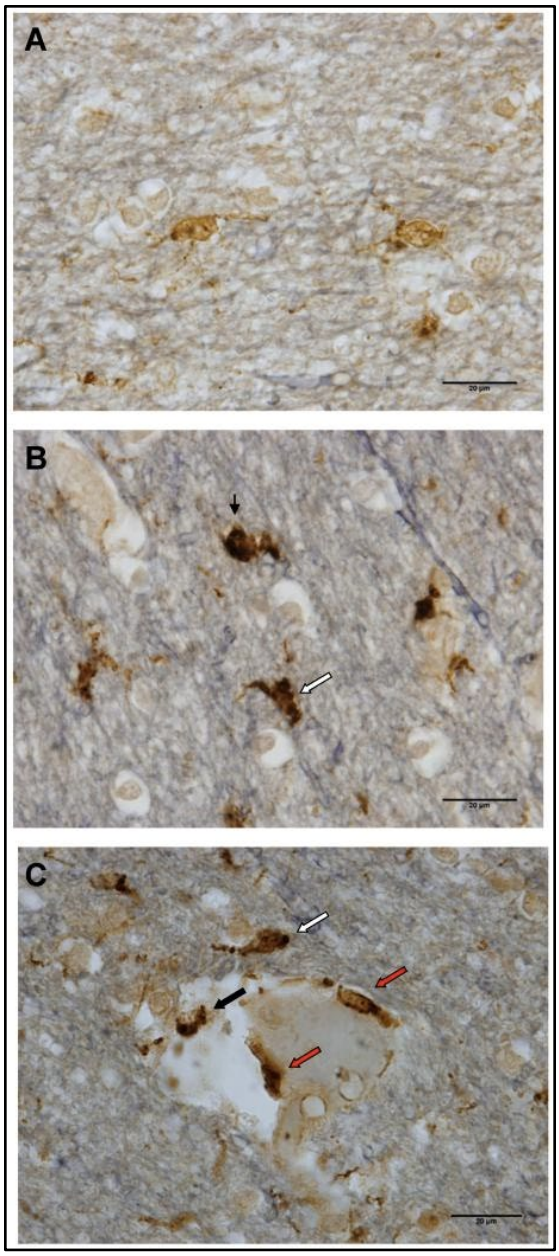

Figure 4. This figure demonstrates the different types of microglial patterns in the four patient groups post immunostaining. From *Microglia of Prefrontal White Matter in Suicide* (Schnieder et al., 2015)

The study grouped together activated microglia and macrophages which are large white blood cells into the category of activated phagocytes. Once microglia undergo activation they are transformed into what is known as activated phagocytes. After collecting the samples, the brain tissues were washed. Then using a physical disector probe, the white matter sections of the brain tissues were studied to estimate their densities. This was done by using two serial sections of the brain tissue (dorsal and ventral) and then counting activated phagocytes, resting microglia, and perivascular cells that appeared in the brain tissues of suicide and nonsuicide victims. **(Refer to Figure 4)** Perivascular cells are found in the blood vessels, and were used as another variable when measuring density change. They

were independent of the resting microglia. Then, correlations between the three control variables, age, sex were determined using linear regression. On average this study used 256 counting frames to analyze 1348 cells per brain tissue.

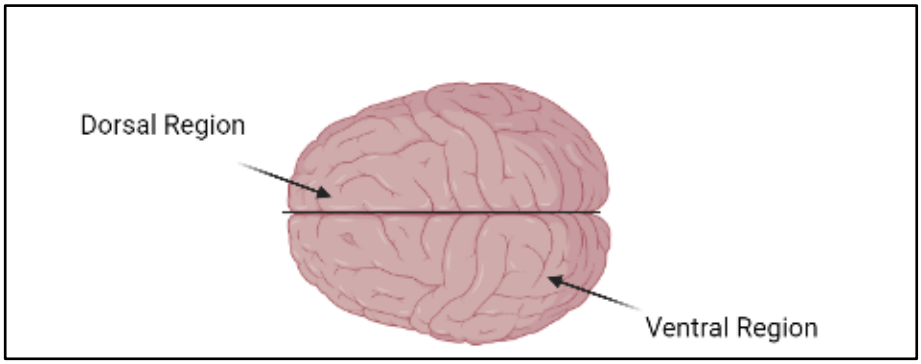

Figure 5. This figure demonstrates the Dorsal Region located above, and the Ventral Region located below in the brain. Created and Copyrighted by Tanish Joshi-Apte.

Table 1. This figure demonstrates the microglial densities by manner of death in dorsal and ventral white matter, with the data being expressed as a mean.

	Suicide Victims		**Non Suicide Victims**	
	Dorsal White Matter*	**Ventral White Matter***	**Dorsal White Matter****	**Ventral White Matter****
Activated phagocytes	5592 ± 2578	6620 ± 2247	6901 ± 2887	6039± 3120
Resting microglia	6292 ± 2267	7001 ± 2415	6897 ± 2489	6897 ± 2489
Perivascular cells	4684 ± 884	4808± 1104	3967 ± 893	4531± 1214
Total cells	16567 ± 1666	18431	16736	17467 ± 3152

*=suicide **=non suicidal

The results demonstrates that there was no significant increase of density in activated phagocytes (includes activated microglia) or resting microglia cells in suicide victims. However in perivascular cells the density was 18% higher in victims who committed suicide than those who did not. This is exemplified in the data from Table 1 which shows the average perivascular density in dorsal white matter being 4684 ± 884 in suicide victims compared to 3967 ± 893 in non suicide victims. However one key phenomenon this data also found was that within dorsal white matter increased microglia cells were found in the blood vessel walls but not in the rest of the white matter. This can allude to a relationship between an increase in the density of microglia cells upon their contact with blood vessel walls. More research is needed to verify and replicate this claim (Schnieder et al., 2015).

Microglia in the Dorsal Anterior Cingulate Cortex

In order to determine if such results can be replicated in other areas of white matter, microglial density in the dorsala anterior cingulate cortex was evaluated to further determine the correlation of microglial density and suicide.

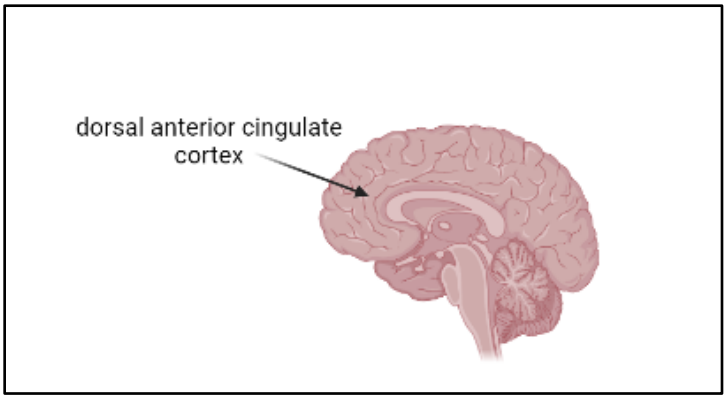

Figure 6. This figure demonstrates the location of the dorsal anterior cingulate cortex in the brain. This region is located in the frontal lobes of the brain playing a role in cognitive processes. Created and copyrighted by Tanish Joshi-Apte.

Dorsal Anterior Cingulate Cortex (dACC): This is a region in the brain located in the frontal area, specifically in the medial wall. The dACC plays a vital role in the cognitive, emotional, and decision making skills of humans. Dysfunction in this region has been associated with psychiatric disorders.

This study used a case control model using immunohistochemistry and phenotypes to differentianate microglial densities between 16 suicide victims of depression, and 13 control subjects through observing macrophages in the dorsal anterior cingulate cortex (dACC) white matter samples. After seeking approval by the Douglas Hospital Research Ethics Board, post mortem brain samples were immunostained using IBA1 microglia marker. One of the key discoveries that this study found that the total density of microglial cells did not differ between depressed suicide victims and the healthy controls. Yet, when observing the relative proportions of different microglial phenotypes, it was shown that victims with suicide has increased amounts of primed microglia. The ratio of primed over ramified microglia significantly increased 2.3 p=0.03. Primed microglia, different from activated microglia, are defined as cells that are in an inflammatory state. Furthermore, an the proportion of blood cells located next to macrophages were double in suicide victims. While these results did not prove an increase in microglial density, the results demonstrate an increase of microglial activation in suicide victims usually associated with neuro-inflammation in the CNS. This data does demonstrate a clear link between microglial activation and suicide. Yet,it is important to understand that this data only focused on dACC white matter and only analyzed suicide victims who had depression. As the sample size was limited and the results may not necessarily be the same if this study was replicated (Torres-Platas et al., 2014).

Discussion of Microglial Density in White Matter Region

While both of the studies looked at white matter in the brain, their conclusions were different. When examining microglia in prefrontal white matter, an increase in microglial density was not observed in suicide victims. Instead in perivascular cells (variable) a clear increase in cell density could be seen in suicide victims. Future studies should analyze perivascular cells in prefrontal white matter to further validate these results. When analyzing microglia in the dACC region a clear increase in primed (specific phenotype) microglial density was observed. This trend was not

present in other microglial phenotypes. The study shows the microglial density from both prefrontal, and dACC regions in the white matter is similar in both experimental and control groups.This could be due to the fact that while both studies tested the frontal white matter, the second study analyzed the dACC region specifically. Similarly, both sample sizes had a limited number of participants. Future studies conducted on microglial cells in white matter should include more sample sizes to create replicable results.

Microglia in the Dorsal Raphe Nucleus

Dorsal Raphe Nucleus (DRN): This is a part of the brain located in the midline of the brain and it helps influence the Central Nervous System's process. It is considered part of the serotonergic system essentially regulating emotions and mood. Abnormalities in the serotonergic system may affect the DRN. Research has demonstrated a change in the DRN function and structure in suicide victims.

In order to create a wider understanding of the role in microglia density in suicide, data from the study "Microglia in the dorsal raphe nucleus plays a potential role in both suicide facilitation and prevention in affective disorders" was analyzed. The data focused on microglial activation and densities. The DRN region was chosen to specifically analyze due to current research indicating a link to suicide. The data was gathered through immunostaining formalin fixed region tissues in the brain with the HLA-Dr Antigen. This antigen is used to determine when understanding the immune response patterns in the microglia. In activated microglia this antigen is usually highly regulated. The microglial densities were analyzed as well to further evaluate the role of the DRN using AgNOR parameters which are quantitative measurements to determine cellular activity. The study sample used post-morterm brains focusing on three groups: non-suicidal patients with psychiatric condition (21), victims of suicide (24) , and healthy controls (22). The non-suicdal and suicidal brain samples included patients with depression, paranoia, and residual symptoms. With the term residual referring to patients with lingering symptoms of a psychiatric condition. When immunostaining these samples the brains were treated with 1.5% H202, and then were incubated with the antibody HLA-Dr.

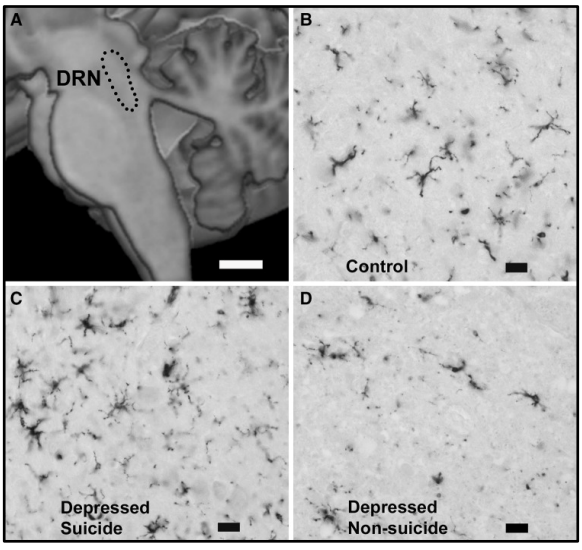

Figure 7. This figure depicts the microglial reaction and activation patterns signified by the black spots in all three testing subjects. Image A also demonstrates the location of the Dorsal Raphe Nucleus region in the brain. From *Microglia in the Dorsal Raphe Nucleus plays a Potential Role in both Suicide Facilitation and Prevention in Affective Disorders.* (Brisch et al., 2017)

Based on **Figure 7**, depressed suicide brains had higher amounts of microglial activation patterns, and darker spots. This pattern supports prior research which established a link between microglial activation and suicide. Furthermore depressed non-suicide brains had the least microglial activation patterns as seen with the least amounts of black spots, followed by the control.

Table 2. This chart demonstrates the median number of microglial cells present in the DRN region of each individual patient group. *refers to brain samples of deceased patients*

Patient Group*	Median Microglial Density (cells/mm^2)
Suicide Patients (parania, depression, and residual included)	198 microglial density
Non Suicide Patients (parania, depression, and residual included)	135 microglial density
Healthy Controls	260 microglial density

This data source used a statistical analysis (STATISTICA version 10) to analyze the samples. In order to measure the microglial density samples were analyzed using spearman correlation coefficients to specifically identify association of microglial density and AgNOR parameters. As seen in **Figure 9**, the median microglia densities were much higher (260 cells/mm^2) in the healthy controls, compared to the suicide and non-suicide patients. With non-suicide patients having the lowest microglial density only having 135. However this data set represents the median of all patient groups and variations in microglial density within the individual groups are possible.

One key trend the results indicated was a lower microglial reaction and significantly decreased microglial density was observed in the non-suicidal depression specific patient group. Furthermore the research identified that this effect was due to any antidepressants or drug usage in the deceased patients. This suggest there are differences in microglial activity in the brains of depressed non-suicide individuals compared to victims of suicide who had depression. Overall, aside from identifying a specific decrease of microglial density in non-suicide patients with depression, the findings did not demonstrate a solid link to increased microglial density in patients with suicide across any underlying psychiatrical condition. As a result these findings are inclusive, with future research being needed to validate the claims (Brisch et al., 2017)

Discussion of Microglial Density in Dorsal Raphe Nucleus

Besides a decrease in microglial reaction and density in non-suicidal patients, no significant increase in microglial density was observed in the suicide victims, thus disproving the hypothesis. However the relationship between the decrease in microglial reaction and density in non-suicidal patients could be seen as positive, since research has demonstrated that the decrease in microglial reaction was a result from anti-depressent medication, with the ultimate goal of that medication being to stop suicide. More research on this specific matter is needed. Furthermore the sample size in this study was very limited with only a total of 67 brain samples being analyzed. A sample size of 150 brains or more for all patient groups would be ideal to create credible results, the complexities of this research inherently creates challenges when selecting brain samples and as a result a very large data set could be infeasible to access.

Limitations

This paper had certain limitations that must be considered. This paper focused on data regarding microglial densities in two parts of the brain: white matter and dorsal raphe nucleus, as a result the data gathered is limited and may not necessarily be replicable in other portions of the brain. Further research studies are recommended to include a wider density data range focusing on all major portions of the brain. This paper analyzed only one data set regarding microglial densities in the dorsal raphe nucleus region compared to two data sets in the white matter region. More data sets when analyzing densities in the dRN region and white matter region would be beneficial to prove the hypothesis. The data this paper used also had a limited number of study participants. The number of post-mortem brains analyzed were limited with all three data sources having less than 70 brains in total for all patient groups. This is due to the nature of the experiment as gathering data from human parts can be challenging due to ethical constraints. Yet increased participants can generate replicable and clear data trends. Increased participants are needed to fully determine if there is an increase in microglial density in suicide victims.

Implications

Based on these three studies: with two focusing on white matter and the third one focusing on microglia in the dorsal raphe nucleus, there is no clear consensus on increased microglial density in the white matter and dorsal raphe nucleus regions of suicide victims. Based on the observations from the data sets, an increase in primed microglial densities in blood vessel walls of suicide victims was established however there was not enough data to validate this claim. Additional studies of primed microglial cells in blood vessel walls, can create new research opportunities. This can potentially facilitate the use of microglia in blood vessel walls in suicide research and prevention. Overall, this paper's hypothesis cannot be proven, with no evidence of increase in microglial density was observed in suicide victims. Additional research into microglia density is warranted, with in-depth research creating more data which can be used to determine new trends, and conclusions. New trials and studies featuring a larger sample size of non-suicide, and suicide victims could generate concrete and replicable data.

However the future of microglia cell research in treating suicide is promising and is not just limited to microglial densities. New research into enabling microglial activation as a biomarker to measure the risk for suicidal behavior or thoughts about commiting suicide are ongoing. Biomarkers are essentially indicators that can be used to measure the progress of a biological process. Microglial activation could be used as a way to determine risk of suicide for a patient, which could enable caregivers to create personalized suicide prevention strategies. Drug development to limit microglial activation is already in progress.These new pharmaceuticals could be used to specifically target microglial activation in patients, with the hopes of preventing suicidal behavior. However indepth research is needed for these ideas to materialize. Ultimately, while the role of microglial densities in suicide victims is inconclusive, the use and function of microglia in determining and treating patients with suicidal behavior is vast.

Acknowledgements

I want to thank Dr. Rajagopal Appavu (Ph.D) for his dedication, and support throughout this research process. I would like to thank my research advisor Jothsna Kethar for helping facilitate this novel opportunity to analyze the relationship between microglial density and suicide.

References

Andoh, M., & Koyama, R. (2021, March 10). *A Comparative Biology of Microglia Across Species*. Frontiers. Retrieved May 25, 2023, from https://www.frontiersin.org/articles/10.3389/fcell.2021.652748/full

Bacskai, B. J., & Garcia, M. (2020, August 24). *Tools and Approaches for Studying Microglia In vivo*. Frontiers. Retrieved May 25, 2023, from https://www.frontiersin.org/articles/10.3389/fimmu.2020.583647/full

Baharikhoob, P., & Kolla, N. J. (2020, July 22). *Microglial Dysregulation and Suicidality: A Stress-Diathesis Perspective*. Frontiers. Retrieved May 25, 2023, from https://www.frontiersin.org/articles/10.3389/fpsyt.2020.00781/full

Brisch, R., Steiner, J., & Mawrin, C. (2017, February 22). *Microglia in the dorsal raphe nucleus plays a potential role in both suicide facilitation and prevention in affective disorders*. NCBI. Retrieved May 25, 2023, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5509773/

Burfield-Bayard, L. (1996, August). *″. ″* - Wiktionary. Retrieved May 25, 2023, from https://pubmed.ncbi.nlm.nih.gov/8880079

The dorsal raphe nucleus and serotonin: implications for neuroplasticity linked to major depression and Alzheimer's disease. (n.d.). PubMed. Retrieved May 25, 2023, from https://pubmed.ncbi.nlm.nih.gov/18772036/

Godbout, J. (2016, May 4). *Neuroinflammation: The Devil is in the Details*. NCBI. Retrieved May 25, 2023, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5025335/

Gonçalves de Andrade, E., González Ibáñez, F., & Tremblay, M. (2022, May 18). *Microglia as a Hub for Suicide Neuropathology: Future Investigation and Prevention Targets*. NCBI. Retrieved May 25, 2023, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC9158339/

Lannes, N. (2017, December). *Microglia at center stage: a comprehensive review about the versatile and unique residential macrophages of the central nervous system.* NCBI. Retrieved May 25, 2023, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5768411/

Microglia in depression: current perspectives. (2020, October 14). PubMed. Retrieved May 25, 2023, from https://pubmed.ncbi.nlm.nih.gov/33068286/

Petković, A., & Chaudhury, D. (2015, November 23). *Neuroinflammation and Depression: Microglia Activation, Extracellular Microvesicles and microRNA Dysregulation*. Frontiers. Retrieved May 25, 2023, from https://www.frontiersin.org/articles/10.3389/fncel.2015.00476/full

Ransohoff, R. M. (2016, January). *Microglia in Health and Disease - PMC*. NCBI. Retrieved May 25, 2023, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4691795/

Schnieder, T. P., Trencevska, I., & Rosoklija, G. (n.d.). *Microglia of Prefrontal White Matter in Suicide*. NCBI. Retrieved May 25, 2023, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4141011/

Sevenich, L. (2018, April 23). *Microglial Dynamics During Human Brain Development*. Frontiers. Retrieved May 25, 2023, from https://www.frontiersin.org/articles/10.3389/fimmu.2018.01014/full

Sorond, F. A., & Gorelick, P. B. (2019, August 8). *Brain White Matter: A Substrate for Resilience and a Substance for Subcortical Small Vessel Disease*. NCBI. Retrieved May 25, 2023, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6721396/

Steiner, J. (2008, January). *Immunological aspects in the neurobiology of suicide: elevated microglial density in schizophrenia and depression is associated with suicide*. PubMed. Retrieved May 25, 2023, from https://pubmed.ncbi.nlm.nih.gov/17174336

Suicide. (2021, June 17). World Health Organization (WHO). Retrieved May 25, 2023, from https://www.who.int/news-room/fact-sheets/detail/suicide

Suzuki, H. (2019, February 13). *Suicide and Microglia: Recent Findings and Future Perspectives Based on Human Studies*. NCBI. Retrieved May 25, 2023, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6381042/

Torres-Platas, S. (2014, November). *Evidence for increased microglial priming and macrophage recruitment in the dorsal anterior cingulate white matter of depressed suicides*. PubMed. Retrieved May 25, 2023, from https://pubmed.ncbi.nlm.nih.gov/24858659/