

The Integral Role of Neurotrophins, Growth Factors, and Aminopropyl Carbazole in the Attenuation of Cognitive Decline

Abstract

This review analyzes 8 studies on the attenuating effects neurotrophins, growth factors, and aminopropyl carbazole (P7C3) have on neurodegeneration and the hypothesized mechanisms for this relationship. Increases in brain-derived neurotrophic factor (BDNF), nitric oxide (NO), insulin-like growth factor-I (IGF-I), transforming growth factor beta 1 (TGF- β 1), and fibronectin type III domain-containing protein-5 (FNDC5) have positive correlations with increases in hippocampus volume and improved scores on memory tests. Conversely, upregulation of the protein leucine-rich repeats and immunoglobulin-like domains 1 (Lrig1) was determined to decrease the neuroregenerative potential conferred by BDNF. The results indicate that BDNF, NO, IGF-I, TGF- β 1, and FNDC5 may positively influence neurogenesis and the maintenance of the neuronal niche in the hippocampus. The therapeutic agent P7C3 was shown to have similar potential by directly regulating BDNF. Further studies are needed to assess the effects that manipulation of each molecule has on mitigating cognitive decline caused by chronic disease.

Keywords

BDNF, VEGF, IGF-1, PDGF-C, NO, Lrig1, TGF- β 1, FNDC5, P7C3, hippocampus, memory, cognition

Introduction

Regression in memory and cognitive function is a growing concern in the aging society of the United States and of the world and has been linked to many chronic diseases and conditions such as diabetes, addiction, chronic anxiety, cardiovascular disease, dementia, and

Alzheimer's. Within the U.S. alone, 9.4% of the population suffered from diabetes in 2015 and 21.5 million adults were dealing with substance abuse in 2014 [1], [2]. Furthermore, 18.1% of the population experiences some form of anxiety, 795,000 people suffer from a stroke each year, and more than 5 million people currently live with Alzheimer's disease [3]-[5].

The hippocampus is recognized as being the short and long-term memory storage center of the brain, and a decrease in the density of neurons, or neurodegeneration, has been associated with a decrease in memory function [6]. A hostile cellular environment and a decrease in the rate of new neuron formation, also known as neurogenesis, are two factors that cause a decrease in hippocampus density. Past studies utilizing human and mouse models have identified that certain molecules and molecular pathways such as brain-derived neurotrophin factor (BDNF), nitric oxide (NO), insulin-like growth factor-I (IGF-1), leucine-rich repeats and immunoglobulin-like domains 1 (Lrig1), transforming growth factor beta 1 (TGF- β 1), and fibronectin type III domain-containing protein-5 (FNDC5) have neuroprotective effects in aging populations, but the results have been largely conflicting and there is no consensus within the scientific community [6]-[15]. Studies utilizing mice have assessed the relationship between the growth factors BDNF, IGF-1, vascular endothelial growth factor (VEGF), and platelet-derived growth factor-C (PDGF-C) and concluded they may stimulate neuron and blood vessel formation as well as improvements in the environment, or niche, in which neurons reside [6]. Past research in an elderly population found that IGF-1 was positively associated with increases in hippocampus volume [6].

Given that research on the topic of memory and the associated molecular pathways is still in its infancy, this review aims to summarize the factors that are currently proposed as playing an integral role in cognition. Analysis of the aforementioned factors and other molecules involved, such as nitric oxide, is warranted for the identification of therapeutic targets that may be

potentially developed into pharmacological agents in the future [7]. To assess which of these biological compounds have the greatest likelihood of being therapeutic candidates, 8 articles identifying molecules and molecular pathways with a direct effect on memory and the structure of the hippocampus were selected for a thorough review. Lastly, studies utilizing P7C3, a pharmacological agent with the potential to attenuate neurodegeneration, were assessed [12], [15].

Methods

Northeastern University's Scholar OneSearch engine was used to identify primary literature that was peer-reviewed and presented original research. Scholar OneSearch utilizes a large subscription base and a number of databases such as Web of Science, Science Direct, PubMed Central, and the Public Library of Science to search for articles relevant to the initial query. Articles were chosen if their contents focused on the analysis of BDNF, IGF-1, VEGF, PDGF-C, NO, Lrig1, TGF- β 1, FNDC5, P7C3 or one of the other molecular substituents and their role in memory function and deterioration. Eight articles published in *Elsevier*, *Cell Reports*, *European Journal of Neuroscience*, *Brain Structure Function*, *Science*, and *Journal of Applied Physiology* met the search criteria and were included for analysis within this review. These articles were published within their respective journals from the years 2015 to 2018.

Results

Brain-derived Neurotrophic Factor

Brain-derived neurotrophin factor (BDNF), a molecule that is produced faster during exercise, influences memory function. Maejima and others drew this conclusion by exercising senescence-accelerated (SAM) mice who had a predisposition to memory decline caused by neurodegeneration. The SAM mice were separated into a sedentary control group and an exercise

group. The exercise group ran on a treadmill for 4 weeks, 5 days a week, for 1 hour a day, and testing was performed on both the control and exercise groups following the completion of the 4-week period. The researchers concluded that mice that exercised exhibited a 10-15% improvement in recognition memory and a 0.2 pg/mg increase in BDNF concentration within the hippocampus. The improvements in memory and the production of BDNF were statistically significant, as evident by the 0.25 relative expression factor of BDNF [8].

The results of the study indicated a positive correlation between BDNF and memory. This correlation directly supported the hypothesized relationship Maejima and others identified between BDNF-promoted neuron formation and memory improvements. The researchers, however, failed to differentiate between the presence of BDNF and pro-BDNF, the inactive precursor to BDNF. Therefore, exercise may have stimulated the release of pro-BDNF, which would create the false impression of correlation or possible causation when compared against the memory test results. Without separately measuring the concentration of BDNF and pro-BDNF, it is difficult to draw the conclusion that exercise stimulated the release of the active neural growth factor from this study [8].

The importance of discerning between active and inactive forms of BDNF before drawing conclusions about a positive correlation between memory and BDNF is evident when comparing the findings of Maejima and others to Ghodrati-Jaldbakhan and others [8], [9]. Ghodrati-Jaldbakhan and others similarly studied the effects of exercise on memory function and BDNF concentration but used mice that modeled the effects of addictive behavior [9]. Addictive behavior was created in mice by administering morphine before the protocol and then ceasing its administration before beginning the exercise program to cause symptoms of withdrawal. Non-addicted mice served as the control group for this study. The mice experiencing morphine

withdrawal were separated into low and high-intensity exercise groups. The exercised group performed 30 minutes of treadmill exercise for 5 days a week for a duration of 4 weeks, and the high-intensity exercise group ran at a final speed that was 12m/min faster than the speed of the low-intensity exercise group [9]. Low-intensity exercise had no effect on BDNF concentration in the hippocampus of the mice, whereas high-intensity exercise resulted in a significant 10% decrease in BDNF concentration. While high-intensity exercise was found to decrease the concentration of BDNF in the hippocampus, it was conversely found to improve spatial memory by 25%. The negative correlation between spatial memory and BDNF concentration in the study by Ghodrati-Jaldbakhan and others directly refutes the claims in the article published by Maejima and others [8], [9].

Ghodrati-Jaldbakhan and others hypothesized that the negative correlation between BDNF and memory function was because exercise stimulated other biochemical pathways unrelated to BDNF. The researchers indicated that biochemical pathways involving dopamine and serotonin may have a direct effect in memory function, but these factors were not measured. Another shortcoming of the research was that Ghodrati-Jaldbakhan and others did not differentiate between the presence of pro-BDNF and active BDNF in the hippocampus, which brings the BDNF measurements under scrutiny [9]. Without having measured both pro-BDNF and BDNF it is possible that exercise may have produced BDNF that remained in the hippocampus while the increased blood perfusion removed the inactive form. Removal of pro-BDNF from the circulatory system would have significantly lowered the concentration of BDNF and potentially impacted the researchers' conclusions. Further experimentation that differentiates between the two BDNF states is necessary to confirm or deny the conclusions drawn by Maejima and others and Ghodrati-Jaldbakhan and others [8], [9].

Nitric Oxide

Nitric oxide (NO), which is produced and transported by the endothelial cells within the body's vascular system, has been shown to directly regulate BDNF within the hippocampus in a rat model. Banoujaafar and others identified NO as a regulator of BDNF within the hippocampus by utilizing an experimental rat model with occluded carotid arteries and assessing both sedentary and exercised conditions. Stopping blood flow to the brain resulted in a decrease of activated NO (p-eNOS) by more than 0.2 units and a decrease in BDNF by 0.15 units within the hippocampal region, and the researchers deduced that NO and BDNF display a linear relationship in rats. To determine causation, brain slices from the rats were incubated with glyceryl trinitrate, which acts as an NO donor, and BDNF and pro-BDNF expression increased by 0.3 units and 0.7 units, respectively [7].

The study by Banoujaafar and others identified NO as a regulator of BDNF and, as a result, neurogenesis within the hippocampal region of the brain. They illustrate that NO is not only important for proper vasodilation and lowering the risk of cardiovascular disease in people with diabetes and hypertension, but it also has implications for memory deterioration. While Banoujaafar and others utilized a clever methodology to deduce correlation and causation between NO and BDNF, they failed to examine the concentration of nNOS, an inhibitor of BDNF, in the brain. As a result, it is difficult to deduce whether a decrease in NO or an increase in nNOS resulted in the reduction in BDNF. Furthermore, 75% of the rats died from the blood flow restriction procedure, and those that survived had developed alternative vascularization to maintain a necessary blood supply to the brain. It is likely that the perfusion of NO to the brain was not as well controlled as indicated by the experimental design and may have affected the BDNF measurements [7].

Insulin-like Growth Factor-I

Maass and others constructed a study of 40 healthy elderly adults that assessed the effects of exercise on the concentration of growth factors known to influence neurogenesis, hippocampal volume, and changes in memory. They found that, independent of the exercise condition, the percent-change in insulin-like growth factor-I (IGF-I) had a positive and linear correlation with the percent-change in volume of the hippocampus and performance in verbal recall memory tests. From a regression analysis, the change in IGF-I and hippocampal volume had a path coefficient of 0.42, whereas the change in hippocampal blood flow (rCBF) and volume had a path coefficient of 0.51. This result indicates that the volume change was more likely associated with the increased blood flow to the specific portion of the brain. IGF-I was determined to have a statistically significant correlation coefficient of 0.42 with late verbal recall, but no causal relationship was determined [6].

The findings of the study by Maass and others were largely inconclusive and may have been due to the study's low power, the effects of natural aging during the 3-month study, and the timing of growth factor measurements. IGF-I levels were found to be elevated for only a duration of 10 minutes following exercise and indicated that the insignificant findings relating to growth factor concentration may be due to the measurement intervals [6]. If exercise has a direct effect on the concentration of growth factors, it would be valuable to conduct future research to determine if IGF-I concentration peaks during exercise and the rate at which it is flushed from the vascular system. In conclusion, the findings from Maass and others indicate a weak correlation between IGF-I and neurogenesis that warrants further studies to deduce if there is a causal relationship.

Leucine-rich Repeats and Immunoglobulin-like Domains 1

In an article published by Trincherro and others, the upregulation of the protein leucine-rich repeats and immunoglobulin-like domains 1 (Lrig1) was demonstrated to have a negative effect on dendritic growth within the hippocampus of mice. The study utilized female mice that were 8 months old divided into either a sedentary or exercise condition. The exercised mice ran on a running wheel for 10-20km a night for a duration of 14 or 21 days, whereas the sedentary mice remained in a cage without a running wheel. Lrig1 was upregulated in 21 sedentary and 23 exercised mice by genetic expression of RV-GFP-oeLrig1. The control group consisted of 12 sedentary and 14 running mice. In the exercised groups, overexpression of Lrig1 resulted in a significant decrease in dendrite length of nearly 300 μm and a decrease of 4 dendrite branching points. On the other hand, downregulation of Lrig1 by expression of the RV-GFP-*sh*Lrig1 resulted in an increase in both dendrite length and branching points in the exercised mice. A 200 μm increase in dendrite length and of 2 branching points was noted between the exercised control and *sh*Lrig1 groups [10].

Dendritic growth and branching are important hallmarks of the integration of new neuron granule cells into the hippocampus. An increased rate of granule cell integration is characteristic of neural plasticity that is present in younger mice populations and tends to decrease with advancing age. Trincherro and others validated the importance of the Lrig1 protein in regulating neurogenesis by its ability to influence the rate at which neurons establish their roots in the hippocampus. An increase in Lrig1 expression resulted in a significant decrease in neurogenesis potential and mitigated the effects of exercise on hippocampal development, whereas a downregulation of Lrig1 allowed for the full potential of an exercise intervention to be realized. Thus, Trincherro and others identify Lrig1 as a negative regulator of the previously identified molecular pathways associated with exercise. Trincherro and others further emphasize this

relationship by presenting findings that Lrig1 proteins decrease neurogenesis by blocking tyrosine kinase (Trk) receptors from binding with BDNF [10].

Trincherro and others identify Lrig1 as an important regulator of neuron maturation in adult mice and as a potential target for a therapy that would downregulate its protein expression capabilities. The researchers, however, failed to assess the effect of Lrig1 regulation on memory capability in mice within their study through learning assessments such as water maze navigation and novel object recognition tests. Furthermore, the causal relationship between Lrig1 and BDNF-Trk binding was speculative and lacked any experimental support [10]. The study could have been improved if the researchers had measured the concentration of molecules downstream of Trk and BDNF in both the Lrig1 upregulation and downregulation mice models. For instance, measuring the concentration of mitogen-activated protein kinase (MAPK) and Ras pathway would have provided conclusive evidence to support or refute the proposed causal relationship [11]. The shortcomings of this research warrant further experimentation and verification of these results in organisms with a more comparable brain development and regulation mechanism to that of humans.

Transforming Growth Factor Beta 1

Transforming growth factor beta 1 (TGF- β 1) is known to be an anti-inflammatory protein that is produced in the hippocampus and assists with neuronal preservation. Choi and others conducted an experiment where 9 male mice, 6-8 weeks of age, received temozolomide (TMZ) to cause neuronal death in the hippocampus. 12 other male mice of the same age received TMZ along with a treatment of LV-TGF- β 1 to overexpress TGF- β 1. The LV-TGF- β 1 treated mice had a 40% increase in expression of TGF- β 1 compared to the control TMZ group. Most significantly,

when measured for the presence of Caspase 3 (Casp3), the LV-TGF- β 1 treated mice had 50 fewer Casp3 expressing cells [12].

The findings by Choi and others are noteworthy because Casp3 is known to be expressed in cells undergoing planned cell death, or apoptosis. The overexpression of TGF- β 1 in transgenic mice was able to substantially reduce the number of dying neurons expressing Casp3 in the hippocampus of male mice, proving the neuroprotective potential of TGF- β 1. Reducing the number of apoptotic neurons in the hippocampus would result in greater preservation of hippocampal volume and the potential for neuronal regeneration to occur at a faster rate than neuronal death. All of these factors are important for maintaining memory function and deterring the onset of Alzheimer's disease in a mouse model. The research at hand is limited in that it only investigated the causal relationship between Casp3 and TGF- β 1 in male mice. The preliminary experimentation of overexpressing TGF- β 1 in female mice yielded insignificant results. This study was restricted by only studying one form of cytokine, TGF- β 1, and its role in the inflammatory pathway that occurs in the hippocampus. Given the promising nature of Choi and others findings, it would be beneficial to study the effects of regulating other proteins in the transforming growth factor family [12]. For instance, in a study by Fukushima and others, transforming growth factor beta 2 (TGF- β 2) was identified as playing an integral role in synaptogenesis, or the formation of new neuronal synapses [13].

Fibronectin Type III Domain-containing Protein-5

Fibronectin type III domain-containing protein-5 (FNDC5) is a molecule that works upstream to regulate the production of BDNF. The level of FNDC5 in the hippocampus was measured in 8 exercised mice and 10 sedentary mice by Choi and others. The exercise group spent 3 hours within a running wheel equipped cage. All 18 mice were administered the

pharmacological agent P7C3 and WNT3 protein to promote neuronal growth and proliferation. The researchers concluded that exercise resulted in a significant 31.62 increase in FNDC5 levels, and a subsequent increase in BDNF levels of 58.42, confirming their proposed causal relationship between FNDC5 and BDNF. The researchers then established that BDNF, and therefore FNDC5, improve cognitive ability in mice by having mice complete cognitive tests. Female mice were either exercised using the previously described protocol or administered LV-BDNF to increase BDNF protein levels within the hippocampus, before completing a delayed nonmatch to position (DNMP) task. DNMP tasks were used to differentiate the pattern memory capabilities between the two mouse groups. The mice administered BDNF were determined to perform as well as the mice that exercised on the DNMP task, and the researchers drew the conclusion that FNDC5 and BDNF were important mediators of memory function in mice [12].

The positive relationship between FNDC5 and BDNF identified by Choi and others was corroborated by a study published by Wrann and others [12], [14]. Wrann and others transduced FNDC5 into a cell-culture of cortical neurons that had been cultured for 7 days (DIV 7) and assessed the concentrations of FNDC5 and BDNF two days later using qPCR. From the qPCR results the researchers observed a 4-fold increase in BDNF expression following the administration of FNDC5 genetic information two days prior [14].

Choi and others and Wrann and others presented conclusive evidence indicating that FNDC5 is directly responsible for production of BDNF in both mouse and cell-culture models. Choi and others further identified that FNDC5 is an important mediator of mouse performance in certain cognitive tasks. Choi and others assessed mouse cognition using behavior, pattern, and spatial oriented tasks and determined that performance was relatively equal across the different tests. Future studies could compare BDNF and exercise model performance on novel object and

object location memory tests to see if alternative tasks would significantly affect the outcome. The researchers also noted that memory improvement only occurred when BDNF and FNDC5 were supplied in conjunction with a pharmacological agent to increase the plasticity of the mouse hippocampus. Future research may deduce what other molecules must be regulated in combination with FNDC5 to result in not just increases in BDNF, but memory retention as well [12], [14].

A question arises when assessing the production and interaction of BDNF and FNDC5: Where are the molecules produced, and are their effects a result of binding to membrane receptors or direct interaction between the molecules? This is a question that Wrann and others were unable to answer from their study. They overlooked the effects that other intermediary molecules may play in the efficacy of the identified BDNF and FNDC5 pathway. However, they hypothesized that FNDC5 is capable of passing through the circulatory system and across the blood-brain barrier [14]. If future studies can verify that FNDC5 crosses the blood-brain barrier, then it may serve as a useful pharmacological agent that can be administered at any point in the circulatory system.

Aminopropyl Carbazole

Many of the previously mentioned molecules such as Lrig1, TGF- β 1, and FNDC5 have been identified because researchers have yet to discover a drug that is highly effective against Alzheimer's disease. However, one drug whose preliminary results have shown to have neuroprotective effects in mice is aminopropyl carbazole (P7C3). Choi and others administered P7C3 in combination with the protein Wnt3 and observed an increase in the number of doublecortin cells (DCX) within the hippocampus of male and female mice. Specifically, treatment of mice with P7C3 resulted in a significant increase in DCX cells of 50%. Measuring

an increase in the number of DCX cells allowed for Choi and others to conclude that neurogenesis was promoted within the hippocampus of mice by administering P7C3. The effects of P7C3 on cognitive test performance by the mice, however, was less significant than the mice that exercised or were also administered BDNF. Therefore, P7C3 promotes neurogenesis but not cognitive improvement when provided as the sole form of treatment [12].

Another study by Jiang and others, however, yielded results that contradict the findings by Choi and others. Jiang and others created a male mouse model that exhibited cognitive impairment by treatment with an amnesic compound called scopolamine. The scopolamine-treated mice were then subjected to a passive avoidance test to assess their memory capabilities. Scopolamine mice that were administered P7C3 performed 3 times better than other scopolamine mice and only 25 seconds worse than the control mice that were not treated with any compounds. From the results of the study by Jiang and others, it is apparent that treatment with P7C3 improves cognition. Furthermore, by analyzing the BDNF protein content of the mouse hippocampus, the researchers determined that P7C3 treated scopolamine mice had higher BDNF protein concentrations than their control counterparts. The scopolamine mice who didn't receive any other form of treatment only had 50% of the BDNF concentration present in the control mice. The researchers' findings indicate that administration of P7C3 results in an upregulation of BDNF production, counter to the results of the study by Choi and others [12], [15].

The conflicting results published on the use of the pharmacological agent P7C3 as a treatment for neurodegeneration proves the need to identify other potential therapies. It should be understood that the study by Choi and others did not directly assess the influence of P7C3 on BDNF. Had all other variables been controlled, there may have been an observable pattern between P7C3 and production of BDNF. Choi and others also used a dosage of 250 mg/kg of

P7C3 a day, whereas Jiang and others noticed significant improvement in cognition and BDNF production by administering only 20 mg/kg of P7C3. The dosage of P7C3 used by Choi and others was at therapeutic levels according to the model utilized by Jiang and others but may have yielded different results because of the use of mice with Alzheimer's disease. The last point indicates the importance of ensuring that the response to P7C3 is similar across many disease models [12], [15].

Discussion

From the articles analyzed for this review, BDNF, NO, IGF-I, TGF- β 1, and FNDC5 have been identified as having a positive correlation with memory improvement and increased hippocampal volume. The findings from these studies indicate that these molecules may regulate neurogenesis within the hippocampus through the BDNF pathway [6]-[9], [12]-[14]. While BDNF expression was determined to have a positive effect on memory tests in healthy mouse models, a similar positive relationship was not evident in the addiction model utilized by Ghodrati-Jaldbakhan and others [9]. While past studies have determined that BDNF is integral to the formation of new neurons, this effect remains to be demonstrated in mouse disease models. Even if future research can determine that overexpression of BDNF is effective in improving memory function within diseased mice, preliminary findings indicate that administration of BDNF may not be an effective treatment due to the size of the molecule. As such, administration of active NO would be more efficient if the causal relationship between NO and BDNF identified by Banoujaafar and others [7] can be verified by future studies.

Maass and others identified a correlation between an increase in IGF-I and hippocampal volume which makes IGF-I a promising and unique candidate for future research, particularly because it is likely to work in a pathway unrelated to BDNF. Concentration of IGF-I is strongly

dependent upon the metabolism of glucose and, therefore, directly affected by chronic diseases such as diabetes [6]. The identification of proper dosing of IGF-I for promotion of an ideal neural environment may allow for better management of diabetes related cognitive decline.

The molecules mentioned to this point were identified because of their positive correlation with BDNF regulation. The findings from the study by Trinchero and others, however, described the molecule Lrig1 as having a negative correlation with BDNF [10]. The ability to downregulate the production of Lrig1 would increase the concentration of BDNF in the hippocampus and stimulate neurogenesis. Future studies should aim to identify where Lrig1 is produced and if a pharmacological agent can be produced to regulate the production of Lrig1.

At this point in time there are relatively few effective therapies available to consumers that specifically control the progression of neurodegenerative diseases such as Alzheimer's disease, dementia, and addiction. Within this review two articles were analyzed that presented the results of using the pharmacological agent P7C3 to treat amnesia and Alzheimer's disease in mice. The results were largely conflicting, with Choi and others concluding that the effects of P7C3 on memory were minimal, whereas Jiang and others determined P7C3 was capable of restoring normal cognitive function [12], [15]. This lack of consensus between the two studies highlights a greater issue which appears to be a significant hurdle to overcome. It appears that there is large variability in the outcomes after using P7C3 that depends on the specific condition that is being treated. While the analysis of P7C3 does not encompass the entire landscape of neuroregenerative drugs, it demonstrates that there is still a significant amount of research that must be completed before a drug such as P7C3 is ready for consumers.

Further research is needed to analyze the effects that overexpression of BDNF, NO, IGF-I, TGF- β 1, and FNDC5 has on the creation of new blood vessels and neurons and the

maintenance of the ideal neural environment. By analyzing current research that presents hypothesized mechanisms for BDNF's positive effects on attenuating cognitive decline, this review highlights therapeutic targets worthy of further research to assess their potential as pharmacological agents. Chronic diseases such as cardiovascular disease, diabetes, and dementia affect the concentration of these molecules which, in turn, has the effect of deregulating the environment in the hippocampus necessary for optimal neuron health and memory function. If the concentration and expression of these molecules can be controlled, there may be the potential to attenuate further neurodegeneration.

References

- [1] American Diabetes Association, "Statistics About Diabetes," *American Diabetes Association*, March 22, 2018. [Online]. Available: <http://www.diabetes.org/diabetes-basics/statistics/>. [Accessed: Nov. 4, 2018].
- [2] S. Thomas, "Statistics on Drug Addiction," *American Addiction Centers*, 2018. [Online]. Available: <https://americanaddictioncenters.org/rehab-guide/addiction-statistics>. [Accessed: Oct. 21, 2018].
- [3] Anxiety and Depression Association of America, "Facts and Statistics," *Anxiety and Depression Association of America*, 2018. [Online]. Available: <https://adaa.org/about-adaa/press-room/facts-statistics>. [Accessed: Nov. 4, 2018].
- [4] Center for Disease Control and Prevention, "Stroke Facts," *Center for Disease Control and Prevention*, 2017. [Online]. Available: <https://www.cdc.gov/stroke/facts.htm>. [Accessed: Oct. 21, 2018].
- [5] Alzheimers.net, "Alzheimer's Statistics," *Alzheimers.net*. [Online] Available: <https://www.alzheimers.net/resources/alzheimers-statistics/>. [Accessed: Oct. 21, 2018].
- [6] A. Maass *et al*, "Relationships of peripheral IGF-1, VEGF and BDNF levels to exercise-related changes in memory, hippocampal perfusion and volumes in older adults," *NeuroImage*, vol. 131, pp. 142-154, 2016. Available: <https://www.sciencedirect-com.ezproxy.neu.edu/science/article/pii/S1053811915010150>. DOI: 10.1016/j.neuroimage.2015.10.084.
- [7] H. Banoujaafar *et al*, "Brain BDNF levels are dependent on cerebrovascular endothelium-derived nitric oxide," *Eur. J. Neurosci.*, vol. 44, (5), pp. 2226-2235, 2016. Available: <https://doi.org/10.1111/ejn.13301>. DOI: 10.1111/ejn.13301.
- [8] H. Maejima *et al*, "Exercise enhances cognitive function and neurotrophin expression in the hippocampus accompanied by changes in epigenetic programming in senescence-accelerated mice," *Neuroscience Letters*, vol. 665, pp. 67-73, 2018. Available: <https://www.sciencedirect-com.ezproxy.neu.edu/science/article/pii/S0304394017309217>. DOI: 10.1016/j.neulet.2017.11.023.
- [9] S. Ghodrati-Jaldbakhan *et al*, "Low- and high-intensity treadmill exercise attenuates chronic morphine-induced anxiogenesis and memory impairment but not reductions in hippocampal BDNF in female rats," *Brain Research*, vol. 1663, pp. 20-28, 2017. Available: <https://www.sciencedirect-com.ezproxy.neu.edu/science/article/pii/S0006899317300963>. DOI: 10.1016/j.brainres.2017.02.024.
- [10] M. F. Trinchero *et al*, "High Plasticity of New Granule Cells in the Aging Hippocampus," *Cell Reports*, vol. 21, (5), pp. 1129-1139, 2017. Available: <https://www.sciencedirect-com.ezproxy.neu.edu/science/article/pii/S2405471317131111>.

com.ezproxy.neu.edu/science/article/pii/S2211124717313542. DOI:
10.1016/j.celrep.2017.09.064.

- [11] J. Romeika, M. Wurzelmann and D. Sun, "Chapter 14 - TrkB Receptor Agonist 7,8-Dihydroxyflavone and Its Therapeutic Potential for Traumatic Brain Injury," *New Therapeutics for Traumatic Brain Injury*, pp. 225-234, 2017. Available: <http://www.sciencedirect.com/science/article/pii/B9780128026861000146>. DOI: [//doi.org/10.1016/B978-0-12-802686-1.00014-6](https://doi.org/10.1016/B978-0-12-802686-1.00014-6).
- [12] S. H. Choi *et al*, "Combined adult neurogenesis and BDNF mimic exercise effects on cognition in an Alzheimer's mouse model," *Science*, vol. 361, (6406), 2018. Available: <http://science.sciencemag.org/content/361/6406/eaan8821.abstract>. DOI: 10.1126/science.aan8821.
- [13] T. Fukushima, R. Liu and J. H. Byrne, "Transforming growth factor- β 2 modulates synaptic efficacy and plasticity and induces phosphorylation of CREB in hippocampal neurons," *Hippocampus*, vol. 17, (1), pp. 5-9, 2007. Available: <https://doi.org/10.1002/hipo.20243>. DOI: 10.1002/hipo.20243.
- [14] C. Wrann *et al*, "Exercise Induces Hippocampal BDNF through a PGC-1 α /FNDC5 Pathway," *Cell Metabolism*, vol. 18, (5), pp. 649-659, 2013. Available: <http://www.sciencedirect.com/science/article/pii/S155041311300377X>. DOI: [//doi.org/10.1016/j.cmet.2013.09.008](https://doi.org/10.1016/j.cmet.2013.09.008).
- [15] B. Jiang *et al*, "P7C3 Attenuates the Scopolamine-Induced Memory Impairments in C57BL/6J Mice," *Neurochem. Res.*, vol. 41, (5), pp. 1010-1019, 2016. Available: <https://doi.org/10.1007/s11064-015-1783-y>. DOI: 10.1007/s11064-015-1783-y.