**Focused Ultrasound- Induced Neurogenesis Requires an Increase in Blood-Brain Barrier Permeability**

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Finding methods to accurately treat brain disorders has been an issue across the medical field. The blood-brain barrier (BBB) has hindered progress but new techniques have come along that can make the BBB more permeable. One method is focused ultra-sound (FUS) administered with contrast microbubbles at peak pressure 1.56MPa. By being able to make the BBB more permeable over the hippocampal region of the brain, it can be shown that, under certain conditions, stimulation of neurogenesis can occur. Currently, the exact means of FUS making the BBB more permeable are unknown but if the mechanism can be isolated then drug administration for the treatment of the brain can be altered for the better

**Introduction**

Scientists have long since faced an interesting dilemma when it comes to treating brain disorders. That, of course, is because of the body’s need to keep the brain away from any harmful material at all costs through a highly selective membrane. The blood brain-barrier (BBB) is so effective at its job that it causes problems treating brain ailments because very few particles can pass through the BBB. Because of this problem, scientists have been working to find a way to increase BBB permeability in selective areas of the brain. The focus of this particular study is targeting the hippocampal region of the brain in an attempt to make it more permeable. Former studies that have been done on this subject have shown that magnetic resonance imaging (MRI)-guided focused ultra-sound (FUS) is a technique that has the capability of opening the BBB [1]. Using magnetic resonance imaging-guided focused ultra-sound allows for scientist to be able to isolate a specific region of the brain to study and attempt to make slightly more permeable. The MRI for this study is to see and compare photos after using specific frequencies and specific pressure amplitudes of ultrasound waves in order to see which environment will make the BBB more permeable. By using this method, along with clinically approved microbubble contrast agents injected intravenously, permeability of the BBB in a controlled area was accomplished. The purpose of the microbubble, which is a small bubble less than 1 mm filled with gas, is to enhance the ability of the ultra-sound waves and imaging. This has allowed for the treatment of neurological disorders because drugs that cannot normally cross the BBB are able to pass through. Because of the study that was already done, it was discovered that there is a correlation between FUS and neurogenesis in the hippocampus. Neurogenesis is the process of creating new neurons by using neural stem cells. FUS without microbubbles was shown to increase the density of brain-derived neurotrophic factor (BDNF). It is known that increased levels of BDNF contribute to neurogenesis. However it is not known which specific factors of transcranial FUS leads to neurogenesis. This study was conducted to attempt to pick out which specific factors of FUS leads to neurogenesis.

**Recent Progress**

The way this study was set up was by arranging 16 adult mice (20-66g) into four different groups. Each group was administered FUS with microbubbles or without microbubbles. Groups one and two had microbubbles, while groups three and four did not have microbubbles. Each group also had a specific pressure during this process. Group one and three were at 1.56MPa, group two at 0.39MPa, and group four at 3.00 MPa. In order to conduct this study, a costume built transducer (1.68 MHz) had to be built with a 60mm radius of curvature and 75mm diameter. After the device was built, each mouse was anaesthetized and had the hair removed from the head. While under, a tail vein catheter was attached then they were laid in a supine position. A microbubble contrast agent, Definity, was administered to groups one and two at 0.02ml/kg immediately before the start of the sonication. Then FUS was administered in the hippocampal region at 1 Hz in 10ms bursts for 120s. The average peak pressure was 1.56MPa. Once this was reached, the pressure was then reduced by fifty percent. For groups two, three, and four the average pressure was increased by 25% (0.39MPa), 100% (1.56MPa), and 200% (3.00MPA) respectively. Once the experiment started, mice were injected with 5-Bromo-2’-deoxyuridine (BrdU) intraperitoneally at 50mg/kg once a day for six days. The animals survived for twelve days after BrdU administration then they were anaesthetized and euthanized. The brains were then removed and sliced on the coronal plane in 50 um sections. Every sixth section was stained then mounted on a slide. Images of the dentate gyrus, a section of the hippocampus, were examined. Groups one, two, and three were viewed at 20X magnification while group four was viewed at 63X. Once this was done, BrdU cells were counted along with the cells that were stained, which were BrdU-DCX and BrdU-NeuN. By doing this, it is seen that NeuN stained cell were prominent in the granular layer of the dentate gyrus while DCX cells are mainly in the subgranular layer. Because of the increase in the average number of BrdU cells, it could be seen that FUS- mediated opening of the BBB increases cell proliferation in the dentate gyrus. However, the presence of both BrdU-DCX and BrdU-NeuN shows evidence that FUS doesn’t just increase cell proliferation but induces neurogenesis specifically. It is shown that the animals in group one were the only group to show an increase in BBB opening in the hippocampal region.

**Discussion**

This study examines how focus ultra-sound can lead to an increase in permeability of the blood brain barrier by examining if microbubbles should be used or should not be used and at what peak pressure is needed for optimal permeability. This study was done with the objective of seeing how neurogenesis is affected based on microbubble administration along with an accompanying peak pressure. Neurogenesis was measured by the increased presence of BrdU-DCX and BrdU-NeuN together in a slice of dentate gyrus from a specific location of the hippocampus. After compiling and analyzing the data, it is seen that neurogenesis is increased if microbubbles are administered with a peak pressure of 1.56MPa. It also shows that neurogenesis does not increase at lower pressure amplitudes with microbubbles or at higher pressure without microbubbles. It should also be noted that FUS has been shown to be both safe and reversible [2]. Unfortunately, it is not known which precise FUS mechanism is responsible for the stimulation of neurogenesis. If the mechanism can be pinpointed then the therapeutic effects could be tremendous.

**References**

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