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Evaluating The Course of an Alzheimer's disease Model in the Rat Brain with High Frequency Ultrasound

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The sporadic form of the Alzheimer's disease (AD) is currently the most severe and frequent type of dementia among older people, representing a critical public health issue. Up to date, many animal models of the AD have been developed, giving the opportunity for the discovery of new diagnostic and therapeutic tools. However, most of the established diagnostic tools are either invasive or of high cost.

Undoubtedly, the possibility of using reliable non-invasive, fast and easy to use technology in order to reveal compositional, topographical and geometrical brain changes caused by AD will be of high scientific value. The current development of high frequency ultrasound (acoustic microscopy) allowed us to penetrate the rat head to a depth of 1.5 -2 cm in order to observe certain micro structures, such as the inflammatory lesions of the AD brain.

An AD rat model was generated after streptozotocin (STZ; 3mg/Kg) intracerebroventricular injection (2μl/100gr body weight per lateral ventricle) in young (3-month old) and old (18-month old) male Wistar rats, bilaterally, under a stereotactic device. Intracerebroventricular STZ has been previously shown to disrupt the cerebral insulin signaling, resulting in reduced brain glucose metabolism and severe oxidative stress, as it is known to occur in the human AD brain.

Brains of the STZ-injected rats were scanned with array probes of ascending frequency (16MHz, 20MHz, 32MHz, 50MHz and 75MHz) at 1, 3 and 6 months after the STZ-injection in order to reveal any possible brain lesions due to the STZ injection. Age-matched healthy rats were used as controls. Experimental rats were deeply anesthetized and their brain parenchyma was scanned while moving the probes from the rostral to the caudal end of the brain cavity. Cross sectional ultrasonic images of high frequency starting from 16MHz up to 75MHz were acquired using:

1. A prototype acoustic microscope of "ORMYLIA" Foundation and a commercial (ATYS medical) one with single element transducer probes with operating frequencies 32-250MHz

And

2. The Aixplorer system (SuperSonic Imagine) with phased arrays probes with operating frequencies 16-20MHz.

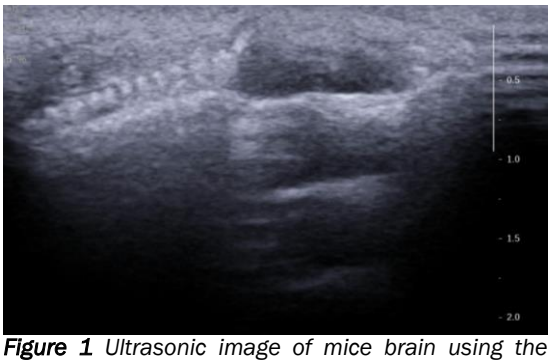

Figure 1 *Ultrasonic image of mice brain using the 16MHz phased array probe.*

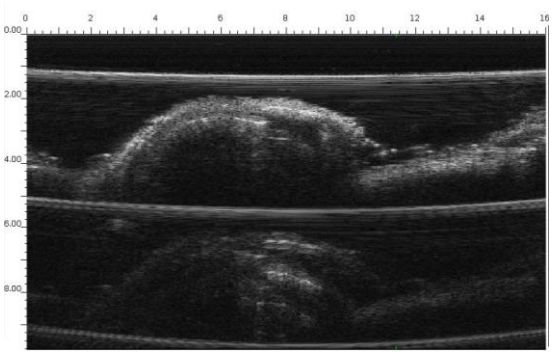

Figure 2 *Ultrasonic microscopy image of mice brain using the 32MHz – 50Mhz single element transducer probe.*

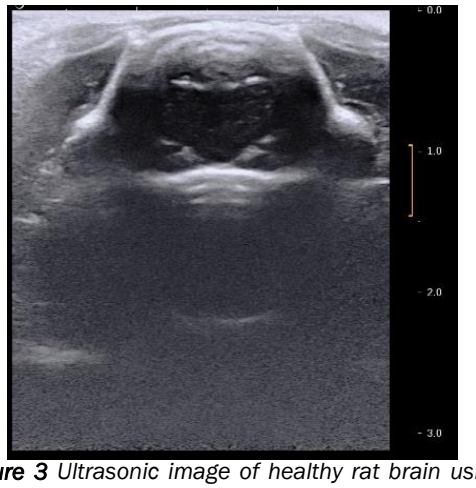

Figure 3 *Ultrasonic image of healthy rat brain using the 20MHz phased array probe.*

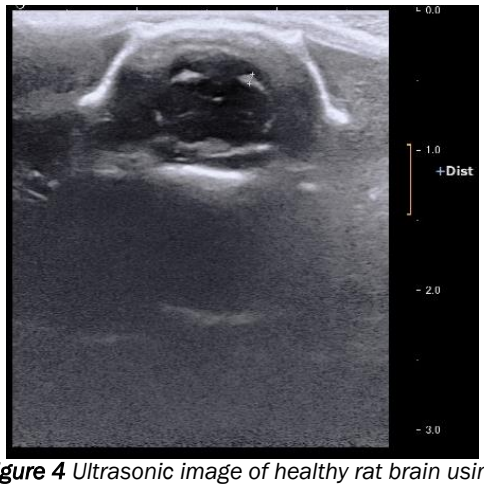

Figure 4 *Ultrasonic image of healthy rat brain using the 20MHz phased array probe.*

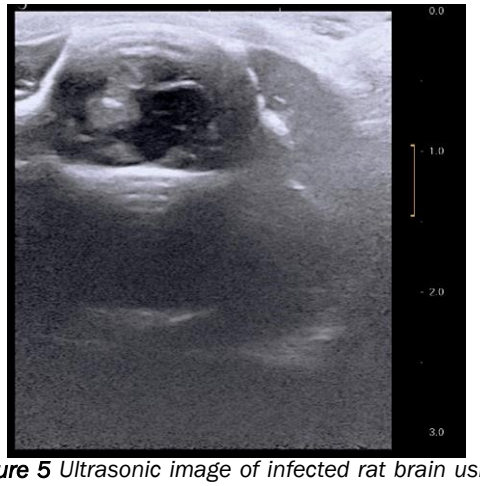

Figure 5 *Ultrasonic image of infected rat brain using the 20MHz phased array probe.*

The acquired images were evaluated using the stereotactic rat brain atlas of Paxinos and Watson. STZ-injected rats were transcardially perfused 3 or 6 months post-injection and affected brains were collected, embedded in paraffin and stained with various histopathological (Nissl, Bielschowvsky silver stain etc.) and immunofluorescence (Thioflavine T, GFAP, Iba1 etc.) techniques, in order to confirm ultrasonographic findings.

Comparative analysis of the images acquired with all types of ultrasonic scanning and optical microscopy indicate that the use of the 20MHz phased array probe produces optimal results, providing a resolution of 77μm, allowing the detection of pathognomonic AD rat brain lesions and their development during AD progression.

References

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