Neurological Findings in Aged Rats Using Translational Imaging Technologies: PET, SPECT and MRI

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1 INTRODUCTION
We have shown that aging causes neurological and memory deficits. Mild neurological changes in neurological function, including minor impairment in memory and cognition can result naturally from aging. In this study, the neurological changes caused by aging were studied with translational imaging techniques in healthy naturally aged rats. Small animal imaging is an important tool for preclinical drug research as it can provide valuable information of disease progression and treatment longitudinally. In this study, we imaged young and aged male rats (2 and 10 months, respectively) with MRI, SPECT and PET to measure changes in blood-brain-barrier (BBB) integrity, cerebral blood flow (CBF) and neuroinflammation to study the effects of aging.

2 MATERIALS AND METHODS
The blood-brain-barrier (BBB) integrity was evaluated using MRI with T1-enhancing contrast agent Gadolinium (Gd). Intravenously administered Gd does not diffuse across intact BBB under normal conditions. Thus, Gd extravasation assessed by T1-weighted MRI is an effective marker of BBB disruption. The extent of Gd entry into the brain was determined as the percent difference in pre- and post-Gd images taken before and 8 min after Gd administration. Single photon emission computed tomography (SPECT) imaging after intravenous injection of 99mTc-exametazime (99mTc-HMPAO) is a conventional method to assess CBF in vivo. It has shown to convincingly strongly with regional brain perfusion and is used in clinical nuclear imaging to detect stroke and other cerebrovascular diseases. To study CBF without restraint or anesthesia, the rats were cannulated in jugular vein and the tracer was injected throughuffed cannula. Alterations in CBF between various brain regions were quantified as regional radioactivity concentration / radioactivity concentration in the whole brain *100%.

To evaluate the level of neuroinflammation in aged rats, dynamic PET imaging of 11C-FETPA was performed. Due to lack of reference tissue, individual arterial input function (AIF) was measured. In this case blood input function was measured from artery-venous shunt during the dynamic PET scan and population based corrections for plasma fraction and metabolism were generated for blood input function from the imaged young and aged rats. The dynamic PET imaging data was fitted to two tissue compartment model (PMOD).

3 RESULTS
When BBB integrity was measured with contrast enhanced MRI, a trend of signal ratio of pre- and post-images in aged rats was observed both in cortex and striatum. However, high variation in the signal ratio was observed in aged group when compared to young animals (Figure 1). Accumulation of Gd was seen in the pineal gland (saifal sinus area between cerebrum and cerebellum). This phenomenon was seen in all aged rats, which also showed enlarged pineal gland mass compared to young rats (Figure 2). Pineal gland doesn’t contain BBB, which explains the higher Gd permeability in aged rats at that region.

In the brain perfusion study with SPECT, the weight and the body composition of the studied groups varied greatly. Average body weights of young and aged rats were 295 ± 14 g and 595 ± 58 g, respectively. As body weight is one of the factors to calculate total brain volume, to avert the bias, the total brain volume was calculated. This value is shown in Figure 3. Aged animals had higher relative blood flow in circulate cortex, medial prefrontal cortex and orbitofrontal cortex, whereas they had lower relative blood flow in inferior colliculus, pons, ventral tegmental area and in cerebellum blood flow in comparison to young rats. Average body weights in PET study of young and aged rats were 288 ± 11 g and 661 ± 94 g, respectively. Again the average SUV images showed higher accumulation in the aged rats, which was due to bodyweight bias. The dynamic PET images were fitted to two tissue compartment model and total distribution volumes (VT) were compared between the groups. No significant difference were observed in Vt values (Figure 4).

However, clear difference was seen in the plasma activity profile used to correct AIF. Aged rats had larger fraction of radioactivity bound to the blood cells, whereas young rats had more radioactivity in the plasma, especially towards the end of the 60 min PET scan. The metabolism of 18F-FEPPA was fast with both groups (Figure 5). This finding highlights the importance of metabolite corrected AIF in small animal PET imaging, especially in the case where the studied groups have different phenotype.

4 CONCLUSION
Translational imaging techniques were used to evaluate alterations in BBB integrity, brain perfusion and neuroinflammation in aged rat model. Subtle changes were seen in BBB integrity and brain perfusion, whereas no signs of neuroinflammation were present. Noteworthy are the changes in tracer kinetics in PET imaging between aged and young animals, which highlights the requirement of full AIF with metabolite correction in small animal PET imaging.

As a summary, translational imaging techniques provide a powerful research tool for CNS disease models allowing comprehensive evaluation of disease progression and treatment interventions for in vivo studies. Metabolite corrected AIF can be obtained from rats without significant blood loss, enabling the use of multiple compartment models and longitudinal imaging without reference tissue.