CHARACTERISATION OF MICROVASULATURE CHANGES AFTER CLOSED SOFT TISSUE TRAUMA BY CONTRAST ENHANCED MICRO-CT IMAGING

[1]Zohreh Barani Lonbani, [1]Tony Parker, [1,2]Michael Schuetz, and [1]Roland Steck

1 Institute of Health and Biomedical Innovation (IHBI), Queensland University of Technology (QUT), Brisbane, QLD
2 Trauma Service, Princess Alexandra Hospital, Woolloongabba, Brisbane, QLD
Email: z.baranilonbani@qut.edu.au

INTRODUCTION

It is known that the vascular morphology and functionality are changed following closed soft tissue trauma (CSTT) [1], and bone fractures [2]. The disruption of blood vessels may lead to hypoxia and necrosis. Currently, most clinical methods for the diagnosis and monitoring of CSTT with or without bone fractures are primarily based on qualitative measures or practical experience, making the diagnosis subjective and inaccurate. There is evidence that CSTT and early vascular changes following the injury delay the soft tissue tissue and bone healing [3]. However, a precise qualitative and quantitative morphological assessment of vasculature changes after trauma is currently missing. In this research, we aim to establish a diagnostic framework to assess the 3D vascular morphological changes after standardized CSTT in a rat model qualitatively and quantitatively using contrast-enhanced micro-CT imaging.

METHODS

An impact device was used for the application of a controlled reproducible CSTT to the left thigh (Biceps Femoris) of anaesthetized male Wistar rats. After euthanizing the animals at 6 hours, 24 hours, 3 days, 7 days, or 14 days after trauma, CSTT was qualitatively evaluated by macroscopic visual observation of the skin and muscles. For visualization of the vasculature, the blood vessels of sacrificed rats were flushed with heparinised saline and then perfused with a radio-opaque contrast agent (Microfil, MV 122, Flowtech, USA) using an infusion pump. After allowing the contrast agent to polymerize overnight, both hind-limbs were dissected, and then the whole injured and contra-lateral control limbs were imaged using a micro-CT scanner (µCT 40, Scanco Medical, Switzerland) to evaluate the vascular morphological changes.
Correlated biopsy samples were also taken from the CSTT region of both injured and control legs. The morphological parameters such as the vessel volume ratio (VV/TV), vessel diameter (V.D), spacing (V.Sp), number (V.N), connectivity (V.Conn) and the degree of anisotropy (DA) were then quantified by evaluating the scans of biopsy samples using the micro-CT imaging system.

RESULTS AND DISCUSSION

A qualitative evaluation of the CSTT has shown that the developed impact protocols were capable of producing a defined and reproducible injury within the region of interest (ROI), resulting in a large hematoma and moderate swelling in both lateral and medial sides of the injured legs. Also, the visualization of the vascular network using 3D images confirmed the ability to perfuse the large vessels and a majority of the microvasculature consistently (Figure 1). Quantification of the vascular morphology obtained from correlated biopsy samples has demonstrated that V.D and V.N and V.Sp were significantly higher in the injured legs 24 hours after impact in comparison with the control legs ($p<0.05$). The evaluation of the other time points is currently progressing.

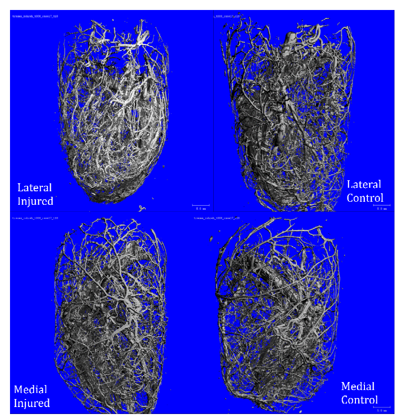

Figure 1: While 3D images of the rat hindlimb's vascular tree obtained 24 hours after injury from micro-CT scans showed no clear differences between injured and non-injured control limbs, the quantification of VV/TV revealed higher values in the injured legs, ($p<0.05$).

CONCLUSIONS

The findings of this research will contribute to a better understanding of the changes to the vascular network architecture following traumatic injuries and during healing process. When interpreted in context of functional changes, such as tissue oxygenation, this will allow for objective diagnosis and monitoring of CSTT and serve as validation for future non-invasive clinical assessment modalities.

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