Quantifying *in vivo* brain motion during injurious head impacts – A pilot study

Tom Whyte, Jie Liu, Vivian Chung, Shelly McErlane, Zelalem A. Abebe, Kurt A. McInnes,
Cheryl L. Wellington, Peter A. Cripton

I. INTRODUCTION

Computational models of the brain are playing an ever-increasing role in understanding the causative factors of traumatic brain injury (TBI), establishing TBI impact tolerances and developing strategies for TBI prevention [1]. The fidelity of computational models to the living brain is critical for these tasks.

Current computational models of the human brain have been almost exclusively validated using human cadaver head impact data [2], despite studies that have shown the mechanical properties of neural tissue degrade quickly after death [3]. It is not known whether the techniques used to prepare and test cadaver brain specimens preserve the *in vivo* mechanical properties. There is an urgent need to precisely measure the deformation of living brain tissue during head impacts relevant to sustaining closed head injury.

The Closed Head Injury Model of Engineered Rotational Acceleration (CHIMERA) was invented at the University of British Columbia (UBC) in 2014 to address the need to provide a clinically relevant animal model of TBI. CHIMERA delivers precise biomechanical inputs that reliably induce repeatable, unrestricted head kinematics following impact, and produces the major pathologies of human TBI, including traumatic axonal injury and haematoma [4]. CHIMERA is currently operational for rodents and ferrets. Ferrets were chosen for this study because of their gyrencephalic brain and larger volume fraction of white matter compared to rodents, enhancing the prospect of translation to human TBI. The aim of this study is to quantify *in vivo* brain motion during CHIMERA-delivered head impacts.

II. METHODS

Three anaesthetized ferrets (1.56–1.6 kg) were used for this study, which was approved by the UBC Animal Care Committee. Buprenorphine (0.04 mg/kg) and Meloxicam (0.2 mg/kg) for analgesia, Glycopyrrolate (0.01 mg/kg) for anticholinergic properties, anti-emetic medication Cerenia (1 mg/kg), antibiotic Ampicilin (20 mg/kg), and Ketamine (6 mg/kg) and Midazolam (1 mg/kg) for sedation were injected subcutaneously. The ferret was anaesthetized with isoflurane (4% for induction and 1–2% for maintenance) and intubated (3.5 cuffed endotracheal tube). The animal was then placed in a stereotaxic surgical frame. A midline incision was made and the cranium was exposed. Three or four holes were burred in the skull, through which fiducials (1.5 mm stainless steel spheres with approximately 8-fold greater density than brain tissue for one ferret and neutral density targets (NDTs) for two ferrets) were inserted into the brain tissue at predetermined stereotaxic coordinates. The NDTs consisted of 1.5 mm stainless steel spheres glued within hollowed 3.2 mm foam beads. Fiducial implantation involved inserting and retracting a solid 16-gauge needle into the brain tissue to form a tract, followed by placing the bead at the top of the tract and using a hollow, flattened-tip 16-gauge needle to push the bead through the induced tract to the desired depth. One fiducial was implanted through each burr hole and into each induced tract. The burr holes were sealed by placing a form-fitting piece of Gelfoam absorbable gelatin (Pfizer, New York, NY) on the brain surface and then applying quick-setting acrylic resin Cortoss (Stryker, Kalamazoo, MI). Additional 2 mm stainless steel spheres were affixed directly to the skull and to a 3D-printed frame that was affixed to the dentition using cyanoacrylate. The incision was sutured and the animal was extubated and maintained under anaesthetic for a minimum of one hour prior to head impacts, allowing the resin to set, the cyanoacrylate to cure and the cerebrospinal fluid to replenish [5].

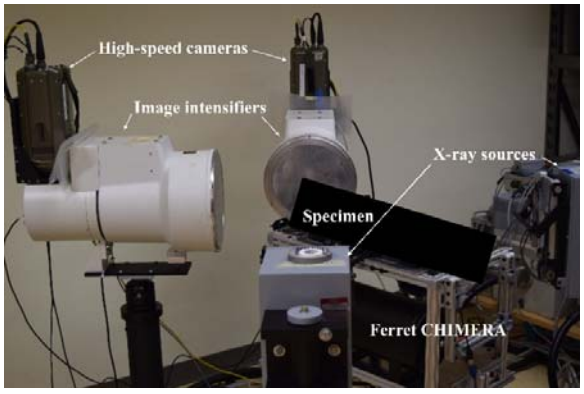

Fig. 1. High-speed fluoroscopy setup and the ferret CHIMERA device.

Each ferret was placed in a supine position on the ferret CHIMERA device (Fig. 1) and subjected to 6–10

T. Whyte (e-mail: tom@mech.ubc.ca; tel: +1 604-652-3004) is a postdoctoral fellow in the Department of Mechanical Engineering at the University of British Columbia, Canada (UBC). J. Liu is a research microsurgeon at the International Collaboration on Repair Discoveries (ICORD). V. Chung is a research engineer at the Centre for Hip Health and Mobility. S. McErlane is a clinical veterinarian at UBC. Z. Abebe and K. McInnes are research engineers at UBC and ICORD. C. L. Wellington is a Professor in the Department of Pathology and Laboratory Medicine at UBC. P. A. Cripton is a Professor in the Department of Mechanical Engineering at UBC and ICORD.

impacts at energies of 17–56 J. This impact orientation produced head translation and rotation primarily in the sagittal plane. For all impacts, an interface was placed between the impact piston and the head of the ferret, which transmitted the load over a greater area of the skull than the piston tip and prevented skull fractures. The head impacts were imaged with dual image intensifiers and X-ray sources in fluoroscopy mode, at 85 kV and 7.5 mA, placed at 90° from one another (Fig. 1). Two Phantom v12.1 high-speed cameras (Vision Research Inc., Wayne, NJ) captured images on the output phosphor of the image intensifiers at 10,000 frames per second. Following the CHIMERA impacts, the ferrets were euthanized and perfused with phosphate buffered saline and 4% Paraformaldehyde. The brains were harvested and dissected to examine the fiducial locations, potential residual tracts and the surrounding brain tissue.

The XROMM software package [6] was used to analyze the collected high-speed videos by performing image intensifier distortion correction, calibration of the 3D space defined by the intersecting fields of view, and tracking of the 2 mm skull beads and 1.5 mm implanted brain fiducials. Fiducial motion data were filtered using a 500-Hz, 4^{th}-order low-pass Butterworth profile.

III. INITIAL FINDINGS

The ferrets showed transient episodes of sinus bradycardia and/or premature ventricular contractions in response to changes in cerebrospinal fluid pressure during the stereotaxic surgery. One required no further intervention and completed the TBI procedure, while two went into cardiac arrest. One was resuscitated using an anticholinergic (Atropine) administered IV and then completed the surgery and TBI procedure. One could not be revived, at which time the surgery was stopped and the TBI procedure was carried out with the *ex vivo* specimen approximately one hour post mortem.

Since it is preferable to disturb as little brain tissue as possible when implanting fiducials, we examined whether it was necessary that the implanted fiducials be neutrally dense (greater overall volume) by comparing the positions of each implanted marker before and after each impact, measured fluoroscopically. It was assumed that if the fiducials remained in place in the tissue, there would be no difference in the pre- and post-impact positions. The average and maximum difference between any pre- and post-impact position of the stainless steel beads over six impacts was 1.07 mm and 4.47 mm. For the NDTs, the corresponding average and maximum was 0.21 mm and 0.62 mm over six impacts.

Relative motion between the NDTs and skull markers was observed during each head impact test. The sagittal plane displacement of one NDT relative to the skull motion in a living specimen is shown in Fig. 2. The range of implanted NDT motion increased with increasing impact severity, see Fig. 2.

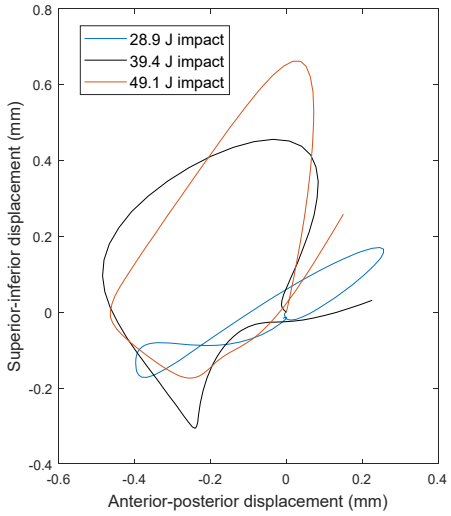

Fig. 2. Sagittal plane NDT displacement relative to the skull during *in vivo* tests for increasing impact severities. Anterior and superior directions are positive.

IV. DISCUSSION

In this pilot study, fiducials were successfully implanted into the living ferret brain and *in vivo* displacements were imaged fluoroscopically during CHIMERA-delivered head impacts. Importantly, these impacts are in the injurious range for closed TBI, with matched severity impacts in non-surgical animals exhibiting imaging abnormalities three hours post-impact [7]. The need for NDTs was confirmed in this series of tests since the stainless steel beads were found to migrate from their implanted positions.

Further testing using NDTs will provide a comprehensive dataset for validating a computational ferret brain model based on *in vivo* brain tissue displacement. Through matched CHIMERA impacts, it will be possible to compare detailed pathological findings with various computational model outputs.

V. REFERENCES

[1] Crandall, J. R., *et al*., *Clin Anat*, 2011.
[2] Hardy, W. N., *et al*., *Stapp Car Crash J*, 2001, 2007.
[3] Oakland, R. J., *et al*., *Proc Inst Mech Eng* [H], 2006.
[4] Namjoshi, D. R., *et al*., *Mol Neurodegener*, 2014.
[5] Begley, D. J., Elsevier Academic Press, 2003.
[6] Knörlein, B. J., *et al*., *J Exp Biol*, 2016.
[7] Hutchinson, E., *et al*., NCA TBI Symposium, 2018.