Appendix 1. Description of Imaging and Brain Stimulation Methods

Method	Description
Functional Brain Imaging	
Functional magnetic resonance imaging (fMRI) is an indirect measure of neuronal activity. It relies on changes in the concentration of oxygen-rich and oxygen-low haemoglobin in the blood caused by different metabolic demands of the neurons during brain activity. Two main methodologies are used in fMRI studies in the context of concussion: task based and resting state.[1,2]	
Task-based fMRI	Investigates how different regions of the brain are activated or deactivated in response to a specific task compared to baseline measures. Brain regions that show an elevated metabolic activity in contrast to a pre-defined baseline during the task are interpreted as associated with the task. Working memory, attention, and executive-control are typically impaired after concussion and therefore are most commonly studied with task-induced fMRI paradigms.[3]
Resting-state fMRI	Relies on the fact that the brain is continually active. Intrinsic brain activity is characterized by synchronized spontaneous activity, with brain regions displaying distinct coordinated patterns or networks, which represent the functional organization of the brain.[4] The differences in synchronization patterns provide key information regarding neurological and developmental diseases.[5,6] A number of distinct resting-state networks have been identified,[7] with the default mode network (DMN) predominantly investigated. The DMN shows higher functional connectivity during a task-free rest state.[8] When the brain is actively engaged in a goal-driven behaviour, the activity in the task-relevant networks is up-regulated while the activity of the DMN is suppressed.[9]
Electroencephalogram (EEG) reflects the electrical activity in the brain. Traditionally, assessments of cognitive processing using EEG relied on event related potentials (ERP) derived from averaging the EEG signal time locked to specific events. These types of studies provide important information about brain function in relation to specific functions such as attention, executive function, visual-perception.[10]	
Structural Brain Imaging	
Diffusion tensor imaging (DTI) provides a measure of white matter integrity by analyzing the three-dimensional shape of water diffusion through axonal tracts and their projections.[11]	
Fractional anisotrophy (FA)	Reflects the integrity of white matter tracts by assessing the degree to which water molecules can diffuse within a voxel.[12] Lower FA values reflect free water diffusion in all directions compared to higher FA values, reflecting diffusion along one axis. Following concussion, mechanical damage to axons disrupts the ratio of intra- and

extracellular water, affecting diffusion along the axon.[13]
Brain Stimulation
Transcranial Magnetic Stimulation (TMS) is based on the principle of electromagnetic induction, whereby a single-pulse over the motor cortex induces an electric field in the cortex, activating intracortical interneurons that, in turn, excite the corticospinal tract to produce a muscle response, indexed through electromyography (EMG). The TMS pulse can be delivered while the participant is resting or as the individual maintains a sustained contraction in the muscle of interest. The TMS pulse provides an index of the excitability in higher threshold interneurons. When delivered during an active contraction, TMS provides a measure of inhibition, seen as a silent period in the muscle response.[14] These indices are important for concussion research as this injury increases the likelihood of axonal disruption and changes excitability.[15] Additionally, pairing two TMS pulses in rapid succession produces an inhibitory response when delivered at an established interval (e.g., 50msec apart).[16] Responses from this paired pulse technique index gamma-aminobutyric acid (GABA)-B receptor activity, which is proposed to be disrupted by concussion.[15]

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