Combinatorial diagnostics of sports-related concussion

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Mild traumatic brain injury (TBI) or concussion (the terms can be used interchangeably) is defined as a head trauma resulting in brief loss of consciousness and/or alteration of mental state.[1] Today, this is a clinical diagnosis based on self-reported symptoms after blunt head trauma, and the condition presents an everyday challenge in emergency care units around the world. Concussion is common in contact sports, such as boxing, American football, ice hockey and rugby, but also highly prevalent in horse-riding.

There are several reasons for why the diagnosis should not be neglected. First, 7-20% of concussion patients presenting to the emergency room have an intra-cranial abnormality (*e.g.*, contusion or hemorrhage) on a day-of-injury CT scan.[2] Second, a significant number of concussion patients get incapacitating symptoms for >10 days following the injury.[3] Third, repetitive concussions may cause traumatic encephalopathy syndrome (TES), a clinical disorder associated with neuropathologically diagnosed chronic traumatic encephalopathy (CTE), which is a neurodegenerative disease,[4] suggesting that care should be taken to minimize the risk of a new concussion before proper recovery from the first one has been secured.

In the current issue of *Journal of Neurotrauma*, McDonald *et al*. undertook a longitudinal prospective study of professional flat-track jockeys with and without incident concussion to examine how the recently developed blood biomarkers for neuroaxonal injury (neurofilament light [NfL] and tau) and astrocytic activation/injury (glial fibrillary acidic protein [GFAP]) can be used together with computerized cognitive testing to diagnose the condition and characterize its time course.[5]

Among 64 professional flat-track jockeys who donated baseline (non-concussed) blood samples and underwent cognitive testing using a computerized test (CogSport), 15 medically diagnosed concussions occurred during the training and competition season. The concussed jockeys were followed with repeated blood sampling and cognitive testing during a year at time points of high relevance to concussion dynamics (2- and 7-days, and 1- and 12-months post-concussion).

A number of important observations were made. First, the blood biomarker concentrations showed different dynamics, corroborating earlier results from moderate to severe TBI.[6] GFAP concentration peaked within two days post-injury, underscoring that it is a fast marker (samples collected during the first day post-concussion would likely have been useful to capture peak values, but this was logistically challenging). GFAP is a protein specific to central nervous system astrocytes. Astrocytic end-feet extending into the neurovascular unit could give rapid release of GFAP into the bloodstream following trauma-related blood-brain barrier injury and astrocytic activation. NfL, on the other hand, was considerably slower with peak values observed 1 to 4 weeks post-concussion. This structural protein is a building block of large-caliber myelinated axons. Its expression is high in long subcortical axons that are vulnerable to rotational forces to the brain. It takes time for NfL to be released from injured axons into the brain extracellular fluid, the CSF and the blood, due to the relatively slow breakdown of cytoskeletal structures. Furthermore, continued release of NfL from the distal part of disconnected axons (so-called Wallerian degeneration) may contribute to increased NfL concentrations in CSF and blood weeks to month after the injury. Tau is another intra-axonal structural protein. In contrast to NfL, it is more highly expressed in thin unmyelinated axons and shows GFAP-like fast dynamics in moderate to severe TBI.[6] Its utility in concussion is, however, uncertain, which is underscored by the fact that no clear concussion-related tau changes were seen among the concussed jockeys. Another candidate biomarker for concussion, ubiquitin carboxy-terminal hydrolase L1, did not produce useful results with most sample concentrations measured below the lower limit of quantification of the assay.

At 2 days post-injury, most of the concussed jockeys showed cognitive impairment compared with their own baseline norm, as determined using the CogSport test, making it possible to link biomarker changes suggestive of concussive brain injury to functional impairment. When combined, 2-day blood biomarker and CogSport results differentiated concussed from non-concussed jockeys with an area under the receiver operating characteristic curve of 0.94, which is excellent.

How can all this be utilized practically? If an individual is suspected to have sustained a concussion, acute cognitive and blood biomarker tests could identify such individuals. This can be corroborated 7-10 days after the injury by an NfL test (GFAP has lost its diagnostic power by then). Cognitive testing will objectify that a concussion with functional brain deficits has occurred, whilst the biomarkers will detect the structural brain injury. The study by McDonald *et al*. shows that cognitive symptoms disappear faster than biomarker changes indicating brain injury. Therefore, emphasis should probably be placed on biomarker normalization and not only clinical improvement to ensure that the brain has recovered before the athlete returns to the sport.

What about individuals with sustained blood biomarker changes post-injury or even pre-season. Stably increased blood NfL concentration is a typical feature of a wide range of neurodegenerative diseases, CTE included.[7] It is possible that individuals who do not normalize in their blood biomarker levels have ongoing neurodegenerative disease. Two of the jockeys showed such profiles. However, before drawing such daunting conclusions more research is needed to exclude potential confounders (these biomarkers are still relatively new, and we need to interpret their results with caution). TBI and other CNS injuries and diseases may expose the immune system to intraneuronal antigens, which could lead to the development of auto-antibodies.[8] Auto-antibodies against a biomarker may prolong its half-life resulting in increased levels in the biofluid. To determine whether sustained biomarker increases indeed indicate neurodegeneration, extended longitudinal studies are needed.

The elegant study by McDonald *et al*. could serve as a blueprint for such studies. A more acute sample could be added, and the study protocol could be expanded to other sports to increase the participant number, as well as the generalizability of the results. Extended follow-up over years of individuals with abnormal biomarker results could also be added (if possible, with a brain donation option). Such studies would require hard work and collaboration but are needed to finally determine if this type of combinatorial cognitive and biomarker test is clinically useful, if objective concussion diagnostics with biomarker-based return-to-play criteria can prevent chronic symptoms, and if sustained biomarker abnormality indeed indicates ongoing neurodegeneration and potentially CTE.

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Competing interests

HZ has served at scientific advisory boards and/or as a consultant for Abbvie, Alector, ALZPath, Annexon, Apellis, Artery Therapeutics, AZTherapies, CogRx, Denali, Eisai, Nervgen, Novo Nordisk, Pinteon Therapeutics, Red Abbey Labs, reMYND, Passage Bio, Roche, Samumed, Siemens Healthineers, Triplet Therapeutics, and Wave, has given lectures in symposia sponsored by Cellectricon, Fujirebio, Alzecure, Biogen, and Roche, and is a co-founder of Brain Biomarker Solutions in Gothenburg AB (BBS), which is a part of the GU Ventures Incubator Program (outside submitted work).

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