ORIGINAL RESEARCH ARTICLE

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Abstract

Background Using functional near-infrared spectroscopy (fNIRS) as an objective diagnostic tool, we aimed to (1) compare fNIRS measurements in adult and youth male rugby players against controls over a playing season, and 2) document the fNIRS changes that occur in concussed rugby players during the England Rugby Football Union Graduated Return-To-Play programme (GRTP). Sixty-seven participants (rugby = 41 (26 adults: 27.5 ± 4.4 years; 15 youth: 16.5 ± 0.6 years; control = 26 (11 adult: 30.5 ± 5.2 years; 15 youth: 16.9 ± 0.4 years) completed fNIRS assessments at pre, mid and end-season. Eight players (five youth, three adult) sustained concussions, and completed fNIRS and the Graded Symptom Checklist from the Sport Concussion Assessment Tool version 5 (SCAT5) assessment throughout the GRTP period. Mixed linear models were utilised to assess the effect of group and time on fNIRS measures of oxyhaemoglobin (ΔO_2 Hb) and deoxyhaemoglobin (Δ HHb) during performance tasks. Typical Error (TE) i.e., normal biological fluctuation and measurement error, was calculated to identify 'cut-off' thresholds for identifying effects of concussion.

Results There were significant differences in fNIRS indices over time in adult and youth groups (p < 0.05) but no significant differences between rugby and control groups (p > 0.05). Seven out of eight (87.5%) concussed players showed changes greater than TE during the GRTP period for both ΔO_2 Hb and Δ HHb during performance tasks and these players' ΔO_2 Hb profiles had not returned to within 'normal' levels within the GRTP period. All players' symptom severity and number returned to normal within the GRTP period.

Conclusion Current GRTP protocols alone are problematic and there is a need for a more individualised approach to concussion management, utilising objective biomarker tools such as fNIRS.

Key Points

• Graduated return-to-play protocols are primarily based on subjective, symptom-based reporting despite limitations.

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- Functional Near-Infrared Spectroscopy (fNIRS) can track recovery time course following concussion.
- fNIRS responses remained altered, even after resolution of subjective symptoms.
- Traditional clinical clearance methods might not fully capture the brain's neurophysiological recovery, potentially exposing players to increased risk of subsequent injuries.
- Incorporating objective measures such as fNIRS into return-to-play protocols could enhance the safety and health outcomes of rugby players.

Keywords Functional Near-Infrared Spectroscopy, Concussion, Rugby, Graduated Return To Play

Background

Sport-related concussion (SRC) is a mild traumatic brain injury (mTBI) caused by external forces applied to the brain [1]. Repetitive concussive injuries and sub-concussive impacts can increase the risk of neurodegenerative complications ranging from mild brain impairments to neurological disease such as dementia and chronic traumatic encephalopathy (CTE) [2]. In certain sports (e.g. rugby, American Football, boxing), risk of concussion is inherent [3, 4]. Over the course of one season, Rugby Union players may experience up to 100 direct head impacts [5] and reporting of concussion incidence in professional rugby has risen significantly from 2009 to 2019 [6]. This trend may not only reflect an increase in the physicality of contact or combat sports but could also be attributed to greater concussion awareness and improved head injury assessment reporting. As such research to improve the understanding of risks, methods for diagnosis and, most importantly, improving the long-term brain consequences for those affected is needed [7, 8].

Near-Infrared Spectroscopy (NIRS) provides measurements of relative changes in blood oxygenation [9]. Functional near-infrared spectroscopy (fNIRS) has been shown to be a valid technique for measuring cerebrovascular health [8] and has demonstrated utility in monitoring concussion recovery [9-11]. For example, retired contact sport players with a history of concussion exhibit altered cortical activity and impaired dynamic cerebral autoregulation (dCA) compared to age-matched controls [12–14]. Research has reported elevated oxygenated haemoglobin (O₂Hb) responses in the prefrontal cortex (PFC) during repeated squat-stand manoeuvres, indicative of compensatory mechanisms for impaired perfusion pressure regulation [12]. Additionally, studies have observed reduced total haemoglobin (tHb), a surrogate marker for cerebral blood volume during visual search tasks that induce Neurovascular Coupling (NVC), reflecting impaired oxygen delivery [13]. Retired rugby players exhibit reduced O₂Hb and increased deoxygenated haemoglobin (HHb) during cognitive tasks, suggesting longterm impacts on NVC [14]. Sub-acute and acute impacts across various combat [15, 16] and contact sports [17, 18] have also shown altered cerebral responses. Adolescent soccer players demonstrated increased O₂Hb in the PFC during complex balance and cognitive tasks following a season of repetitive head impact exposure, suggesting compensatory prefrontal activation to maintain performance [18]. Similarly, university athletes showed acutely increased O₂Hb responses following ball heading during single and dual-task assessments, indicative of heightened prefrontal recruitment for motor-cognitive responses [17]. In professional boxers, significant cortical deoxygenation during orthostatic challenges highlighted impaired cerebral oxygenation and dCA due to repetitive trauma [16]. Recently, Martini et al. [10] and Jain et al. [11], have demonstrated the sensitivity of fNIRS to detect persistent alterations in prefrontal cortical activity during gait and dual-task challenges in concussed adolescents compared to controls. Consequentially, these findings support the use of fNIRS as an objective measurement tool for concussion [12]. Additionally, fNIRS derived measurements of NVC and dCA may function as biomarkers of cerebrovascular health, aiding in concussion diagnosis and return-to-play/learn processes.

The purpose(s) of this study were to; (1) compare fNIRS measurements following NVC and dCA inducing tasks in adult and youth male rugby players and non-contact sport controls over a playing season and; (2) document the fNIRS changes that occur in concussed rugby players during the 2022-23 England Rugby Football Union (RFU) Graduated Return to-To-Play (GRTP) programme [13].

We hypothesised that; (1) fNIRS changes in male rugby players over a season would be significantly different from those in a control group; (2) rugby players who sustain a concussion will exhibit more pronounced fNIRS changes than non-concussed players.

Methods

A longitudinal study was used to characterise the effects of a season of rugby union on cerebral oxygenation. fNIRS data were collected at: pre, mid, end-season. An exploratory, prospective cohort study utilising rugby players who sustained a concussion was used to document the fNIRS changes that occur during the England RFU GRTP programme [13].

Participants

Participants were adult male 1st XV and youth players (16–18 years) from a UK amateur rugby club and controls. Control group participants were recruited from

local sports clubs and schools, did not take part in contact or combat sports and had no history of concussion in the last 5 years. Contact or combat sports were defined as sports or activities where participants engaged in physical interactions that involve grappling, striking or forceful body contact [14]. All participants completed a General Health Questionnaire (GHQ). Demographic information is shown in Table 1.

Procedures

Testing took place in the same set of rooms at the same time of day. Participants had not exercised in the previous 12-hours or consumed alcohol/ caffeine within 6-hours. Baseline testing was conducted over two days. Day one involved demographic measures, GHQ, and the Sport Concussion Assessment Tool v5 (SCAT5) (adult rugby only). On day two, fNIRS assessment took place.

Any concussions were identified on field by team medical (physiotherapist) or coaching staff. Following the 2022-23 England RFU GRTP programme guidelines [13], clinical recovery was defined as a resolution of post-concussion symptoms, and resumption of normal activities (school/work/sport).

England RFU GRTP guidelines during the 2022/23 playing season followed the same six stage return to play process as the World Rugby GRTP programme [17], with longer mandatory stand down periods of initial rest i.e., youth players (U19) minimum of 23 days and adult community players 21 days.

The SCAT5 is a standardised tool for evaluating concussions [1, 15]. It was administered to adult rugby players during pre-season assessments. If a concussion occurred, symptom evaluation (number and severity) from the Graded Symptom Checklist (GSC) was repeated during the GRTP process. The SCAT5 has been shown to discriminate between non-concussed and concussed athletes [1, 15], and is most valid within 72 h of concussion [1].

fNIRS Device and Protocol

The fNIRS head cap was placed 1 cm above the eyebrows on the supraorbital ridge to avoid the sinuses (Fig. 1). The 24-channel, continuous wave system (Brite, Artinis Medical Systems, Elst, The Netherlands, www.artinis. com) monitored changes (micro-molar, μ M) in oxyhaemoglobin (Δ O₂Hb), and deoxyhaemoglobin (Δ HHb) in the pre-frontal cortex (PFC). Changes in light attenuation were measured at two wavelengths (762 and 843 nm) and concentration change was calculated using the modified Beer–Lambert law within Oxysoft software (v3.5.15.2.). The differential path length factor (DPF) was calculated for individuals in relation to their age [16].

A modified Neary Protocol, detailed elsewhere [7], was used to assess brain oxygenation during tasks that induced NVC using the "Where's Wally" visual search task paradigm and dynamic cerebral autoregulation (dCA) was assessed using a squat-stand (SS) manoeuvre. Briefly, the "Where's Wally" test was conducted on a high definition (HD) screen (22"size), with participants at a self-selected distance for optimal focus. A block-design was utilised, consisting of five cycles of 20 s with eyes closed followed by 40 s with eyes open during which the participant searched the screen for "Wally". If the participant found "Wally" the slide was immediately advanced to a new slide. This continued for the duration of each 40 s eyes open block. The test took 5 minutes in total. Prior to the SS manoeuvre, the participant sat quietly for 1 minute and then stood up for 1 minute to allow the body to adjust to a standing position. Participants then completed 5 minutes of a SS manoeuvre which consisted of 10 s squatting, followed by 10 s standing (total of 15 squats) [18]. During the squat portion, participants held $a \sim 90^{\circ}$ knee angle, and then returned to standing at a pace dictated by an audio timer on the HD screen. Previous research have shown these tests to be valid measures of NVC [19] and dCA [20] respectively. Task Performance was evaluated by the number of correct object identifications during the "Where's Wally" task.

Table 1 Group demographic and general health questionnaire data (mean ± 9	SD)
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Variable	Adult Rugby	Adult Control	Youth Rugby	Youth Control
Participants (n)	26	11	15	15
Age (years)	27.5 ± 4.4	30.5 ± 5.2	16.5 ± 0.6	16.9 ± 0.4
Height (cm)	181.4±5.8	180.2±6.9	181.1±5.0*	174.2±5.7
Body Mass (kg)	103.0±17.0*	82.1 ± 9.1	83.7±18.6*	69.8 ± 14.2
Previous Concussions	2.3±2.1*	0	1.9±2.8*	0
Physical Activity (mins/week)	514.2 ± 286.9	533.8±349.8	588.0±271.98*	295.3±136.9
Games played	12.5 ± 5.2	N/A	12.1±7.5	N/A
Minutes Played (mins)	866.2±375.5*	N/A	621.8±337.8	N/A

Note: Control subjects' various sports included baseball, 5-aside football (no heading), powerlifting, swimming, and running

*Significant differences between rugby and control groups



Fig. 1 Headcap frontal and lateral views and 3D plot depicting optode arrangement and experimental protocol. T = transmitter and R = receiver. Data were collected at a sample frequency of 50 Hz via multi-wavelength LEDs situated on a soft neoprene head-cap containing 8 receivers and 10 transmitter channel pairs, 30 mm apart. The probe positions of the fNIRS detection device covered the area linking Fp1, F3, F7, and Fp2, F4, F8, corresponding to the left and right prefrontal cortex respectively, according to the international EEG 10–20 system [21]

fNIRS Data Analysis

fNIRS signals were exported into MNE-NIRS, a Python toolbox, for analysis [22]. Each channel was visually inspected for signal quality. Channels with excessive motion artifacts, and/or non-pulsatile signals were excluded [23] utilising the signal quality index (SQI) function (i.e. SQI Value < 3) implemented in Python [24]. A 5th order Butterworth filter was utilised to remove motion artifact [25, 26] and a low pass filter (cut-off frequency of 0.09 Hz) was applied to all channels to keep all frequencies below the lowest systemic frequency [27]. Across all participants and groups, an average of 3% of channels were discarded at the pre-season (range of 0–8%), 4% at mid-season (range of 0–13%), and 4% at end-season (range 0–17%).

To evaluate changes in PFC oxygenation, data across all 24 channels covering the PFC were averaged. This approach was chosen to capture global cortical trends, reflecting the overall activation of the PFC during tasks that induce NVC and dCA.

 O_2 Hb and HHb variables were analysed by calculating the change (Δ) between the average maximal values during the 40-second eyes-open task and the average minimal values during the 20-second eyes-closed within each trial for the "Where's Wally" visual search task. This computation was repeated across five trials for each channel, and the resulting Δ values were averaged across the 24 channels. For the squat-stand manoeuvre, Δ values were similarly calculated as the difference between the average maximal values during the 10-second squat and the average minimal values during the 10-second stand periods, averaged across the 15 trials and 24 channels [28, 29].

Statistical Analysis

Descriptive statistics are presented as mean ± SD (Table 1). Descriptive comparisons were analysed using independent t-tests. Separate mixed-effects linear models were used to assess the effect of group (control, rugby) and time point (pre-season, mid-season, and end-season) on ΔO_2 Hb and Δ HHb during the "Where's Wally" and squat-stand manoeuvre tasks in adult and youth rugby and control cohorts. Models were fitted using Restricted Maximum Likelihood estimation. If statistically significant differences were identified, Bonferroni post hoc tests were applied to correct for multiple comparisons. Given the known protracted development of the prefrontal cortex, which continues to mature into the mid-20s [30, 31] and recent evidence that has indicated concussed adolescents show different responses in NVC compared to adults [32] youth (16.6 ± 0.5 years) and adult (28.4 ± 4.8 years) cohorts were analysed separately to account for age-related differences in neural activation and cognitive performance during working memory tasks. We used repeated measures taken across the playing season from rugby and control participants to calculate Typical Error (TE) represents the variability in observed measurements due to both instrumentation noise (e.g., equipment precision) and biological noise (e.g., circadian influences, hydration status) [33]. It was calculated as the standard deviation of the differences between repeated measurements divided by $\sqrt{2}$.

Equation:

$$\frac{TE = SDdiff}{\sqrt{2}}$$

In this study, repeated measures from only non-concussed rugby and control participants were analysed using the Hopkins reliability spreadsheet to calculate TE [32]. Data are reported as means with 90% confidence intervals (CI). TE values for fNIRS ΔO_2 Hb, Δ HHb, "Where's Wally," and squat-stand were calculated separately for each group (adult and youth, rugby and control participants). These TE thresholds were subsequently used to distinguish meaningful changes in concussed rugby players, beyond the range of normal biological and measurement variability. Independent t-tests were employed to identify differences in TE values between rugby and control groups, establishing group-specific thresholds for further analysis [34]. These values served as thresholds for identifying effects of concussions beyond normal fluctuation / measurement error in concussed rugby players. Statistical analyses were performed using IBM SPSS v.29.0.

Results

Significant differences in baseline characteristics between rugby and control groups were identified by independent sample t-tests with significant differences in height (youth rugby taller than youth controls p = 0.001), and body mass (youth and adult rugby players heavier than controls p = 0.02, p = 0.001 respectively). Significant differences were also seen in concussion history with adult and youth rugby players having experienced a greater number of concussions compared to controls (p = 0.0009, p = 0.0042 respectively. Adult rugby players played significantly more minutes than youth players (p = 0.042: Table 1)

Fig. 2 displays an example participant fNIRS timeline trace for O_2 Hb and HHb responses during the WW, transition and squat-stand tasks.

Playing Season

Changes in fNIRS responses for control and rugby adult and youth groups during neurocognitive visual search ("Where's Wally") and neurophysiological (squat-stand) tasks are shown in Table 2.

Neurocognitive: "Where's Wally"

There was no significant effect of group (adult / youth control vs. rugby), time or interaction effect on ΔO_2 Hb (p > 0.05). There was no significant effect of group (adult / youth control vs. rugby), time (adult) or interaction effect on Δ HHb (p > 0.05) There was a significant effect of time on Δ HHb for youth groups (p = 0.04). In youth groups post-hoc pairwise comparisons indicated that mid-season Δ HHb was significantly higher compared to end-season.

Task Performance: "Where's Wally" Identification

There was no significant difference in "Where's Wally" object identification between pre, mid and end season for all groups (p = > 0.05) (Supplemental 1).

Neurophysiological: Squat-Stand Task

There was no significant effect of group (adult control vs. rugby), time or interaction effect on ΔO_2 Hb or Δ HHb (p > 0.05). There was no significant effect of group (youth control vs. rugby), or interaction effect on



Fig. 2 An individual oxyhaemoglobin (O₂Hb) (solid line) and deoxyhaemoglobin (HHb) (dashed line) signal trace for an adult rugby player at pre-season. The signal profile is similar for all groups and time points within the season

Table 2 Adult and youth, control and rugby group mean \pm se change (Δ) in "where's Wally" (WW) and squat-stand (SS) tasks; Oxyhaemoglobin (ΔO_2 Hb) and Deoxyhaemoglobin (Δ HHb) profiles across a rugby playing season at three time points (pre, mid and end season)

Age Group	Group	Task	Pre-Season		Mid-Season		End-Season	-Season	
			ΔO₂Hb (μM) Mean±SD	ΔHHb (μM) Mean±SD	$\Delta O_2 Hb (\mu M)$ Mean ± SD	ΔHHb (μM) Mean±SD	ΔO₂Hb (μM) Mean±SD	Δ HHb (μ M) Mean ± SD	
Adult	Control	WW	1.06±0.55	0.25 ± 0.09	0.92±0.50	0.25 ± 0.08	0.88±0.45	0.27±0.10	
Adult	Rugby	WW	1.06 ± 0.63	0.28 ± 0.10	1.07 ± 0.44	0.28 ± 0.10	1.43 ± 0.84	0.38 ± 0.16	
Youth	Control	WW	1.17±0.73	0.32 ± 0.17	1.24 ± 0.76	0.31±0.13*	1.21 ± 0.40	0.26 ± 0.10	
Youth	Rugby	WW	0.89±0.61	0.24 ± 0.11	1.14±0.63	$0.33 \pm 0.12^*$	0.88 ± 0.50	0.23 ± 0.08	
Adult	Control	SS	6.75±1.81	1.05 ± 0.38	6.55 ± 1.62	0.98 ± 0.39	6.66 ± 2.46	1.07 ± 0.52	
Adult	Rugby	SS	7.28±1.97	1.17±0.32	7.81 ± 1.88	1.28 ± 0.44	7.29 ± 2.36	1.26 ± 0.43	
Youth	Control	SS	5.79±2.83*	$0.99 \pm 0.50^{*}$	7.79 ± 3.31	$1.28 \pm 0.46 \dagger$	7.00 ± 3.09	1.21 ± 0.44	
Youth	Rugby	SS	6.10±3.07*	$1.08 \pm 0.53^{*}$	7.57 ± 2.79	1.26 ± 0.53	8.21±2.64	1.40 ± 0.41	

*Significant change over time. †significant group x time interaction effect

Table 3 Calculated typical error (TE) values for "where's Wally" (WW) and squat stand (SS). Data are shown as mean with 90% confidence intervals (90%CI)

O ₂ Hb TE	HHb TE
0.29 [0.24, 0.37] +	0.10 [0.08, 0.13]
0.42 [0.30, 0.95]	0.11 [0.09, 0.14] +
1.54 [1.31, 1.90] +	0.27 [0.23, 0.34] +
1.41 [1.19, 1.80] +	0.23 [0.19, 0.29] +
	O₂Hb TE 0.29 [0.24, 0.37] + 0.42 [0.30, 0.95] 1.54 [1.31, 1.90] + 1.41 [1.19, 1.80] +

All TE values can be found in Supplemental 2. + indicates pooled TE values

 ΔO_2 Hb (p > 0.05). Time had a significant effect on ΔO_2 Hb (p = 0.004) and time (p = 0.03) and group by time interaction effect on Δ HHb (p = 0.02). Post-hoc pairwise comparisons indicated pre-season ΔO_2 Hb was significantly lower compared to mid-season (p = 0.013) and end-season (p = 0.01). Δ HHb pre-season was significantly lower than end-season (p = 0.03). The significant interaction effect showed that Δ HHb for the youth control group was higher at mid-season compared to youth rugby (p = 0.02).

Typical Error (TE)

Calculated TE thresholds for ΔO_2 Hb and Δ HHb during "Where's Wally" and squat-stand tasks are shown in Table 3. Independent samples t-tests showed significant differences between control and rugby participants TE scores for Δ HHb (adult) (p = 0.03) and ΔO_2 Hb (youth) (p = 0.001) "Where's Wally" and no significant differences (p > 0.05) for SS (ΔO_2 Hb and Δ HHb) respectively. Therefore, rugby participant only TE values were created for the adult Δ HHb and youth ΔO_2 Hb WW task and nonsignificant TE values were pooled between control and rugby participants for the ΔO_2 Hb adult and youth Δ HHb for WW and adult and youth squat-stand ΔO_2 Hb and Δ HHb to establish 'cut-off' TE threshold values (Table 3).

Concussion

Individual concussion fNIRS profiles are shown for "Where's Wally" (Fig. 3a and b) and squat-stand (Fig. 4a and b) tasks, with TE thresholds overlaid.

Concussion symptom number and severity reporting are shown in Table 4.

Eight players (five youth, three adult) who sustained a concussion reported symptom severity and number scores via the GSC during the early post-injury period (<25 days) period. Four players (50%) reported symptom severity and six players (75%) reported symptom number changes greater than those suggested to be 'broadly normal' [35] following concussion. All players (100%) had returned to within normative normal ranges within the GRTP period [13].

Discussion

This is the first study to use fNIRS to monitor early postinjury (<25 days) and season long (8 months) changes in brain haemodynamic response utilising fNIRS measurements during activities inducing NVC and dCA as objective markers of brain health in adult and youth rugby players. Results showed no significant group differences between rugby and non-contact sport controls for changes in fNIRS measurements of NVC or dCA. There were, however, small but significant variation in adult and youth fNIRS responses during "Where's Wally" and squat-stand tasks, indicating neuro-cognitive/physiological changes over time. Seven players (87.5%) who sustained a concussion showed altered fNIRS responses that were greater than the TE. Importantly, within the players who sustained a concussion, 75% of players' NVC ΔO_2 Hb (Fig. 3a) and 87.5% of player's ΔO_2 Hb SS (Fig. 4a fNIRS responses had not returned to normal levels within the



Fig. 3 (a) Pre-season and post-concussion change in oxyhaemoglobin (ΔO_2 Hb) for eight players (five youth, three adult). Seven out of eight (87.5%) players show changes greater than Typical Error (TE) during the early post-injury period (<25 days) period. Six players show an increased ΔO_2 Hb response during the "Where's Wally" task and one player shows a decrease. Findings indicate a dysregulated ΔO_2 Hb response following concussion. Six out of eight (75%) players' ΔO_2 Hb profiles had not returned to within 'normal' levels within the GRTP period. R = adult rugby and CR = youth rugby. (b) Pre-season and post-concussion change in deoxyhaemoglobin (Δ HHb) for eight players (five youth, three adult). Seven out of eight (87.5%) players show changes greater than TE during the early post-injury period (<25 days) period. Six players show an increased Δ HHb response during the "Where's Wally" task and one player shows a decrease. Findings indicate a dysregulated ΔP_2 Hb response following concussion. Six out of eight (75%) players' ΔO_2 Hb profiles had not returned to within 'normal' levels within the GRTP period. R = adult rugby and CR = youth rugby. (b) Pre-season and post-concussion change in deoxyhaemoglobin (Δ HHb) for eight players (five youth, three adult). Seven out of eight (87.5%) players show changes greater than TE during the early post-injury period (<25 days) period. Six players show an increased Δ HHb response during the "Where's Wally" task and one player shows a decrease. Findings indicate dysregulated Δ HHb responses following concussion. Five out of seven (71%) players' Δ HHb profiles had not returned to within 'normal' levels within the GRTP period. R = adult rugby and CR = youth rugby

GRTP period. Conversely, all players' (100%) subjective symptom numbers and severity had returned to normal ranges within the same period (Table 4.).

Playing Season

Neurovascular Coupling (NVC): "Where's Wally"

No significant group differences were found for ΔO_2 Hb or Δ HHb between adult or youth rugby and control groups. However, significant differences during the season were seen with youth groups showing a decrease in Δ HHb from mid- to end-season. The "Where's Wally" visual search task is designed to engage the visual and attention mechanisms in the brain [7, 18], resulting in increased neural activity. This requires more oxygen, leading to a process called neurovascular coupling [36]. In healthy individuals, complex visual search tasks induce an increase in O_2 Hb in the PFC [37]. The increase in Δ HHb seen within youth groups at mid-season indicates an altered haemodynamic response, with no change in task performance i.e., no change in number of correct identifications of "Where's Wally" (Supplemental 1) to easily explain this alteration. Jain et al. [11] found that task familiarity significantly influenced PFC activation during dual-task assessments, with adolescents showing reduced O₂Hb as they became more accustomed to repeated trials. This reduction suggesting a shift toward more efficient neural processing with experience. In the present study, the observed variability in Δ HHb among both youth groups over the season may reflect developmental differences in the brain's ability to adapt to repeated cognitive demands, even in the absence of performance changes. While limited comparative studies exist, studies have reported significantly increased post-season Δ HHb and Δ total haemoglobin (Δ tHb) in collegiate rugby players [38] and significantly increased ΔO_2 Hb from pre-to post-season in soccer players [39]. Additionally, average



Fig. 4 (a) Pre-season and post-concussion change in oxyhaemoglobin (ΔO_2 Hb) for eight players (five youth, three adult). Seven (87.5%) players show changes greater than Typical Error (TE) during the early post-injury (<25 days) period. Six players show increased ΔO_2 Hb responses during the squat stand manoeuvre and one player shows a decrease. Findings indicate a dysregulated ΔO_2 Hb response following concussion. Seven out of eight (87.5%) players' ΔO_2 Hb profiles had not returned to within 'normal' levels within the GRTP period. R=adult rugby and CR=youth rugby. (b) Pre-season and post-concussion change in deoxyhaemoglobin (Δ HHb) for eight players (five youth, three adult). Seven (87.5%) players show changes greater than TE during the early post-injury (<25 days) period. Six players show increased Δ HHb responses during the squat stand manoeuvre and one player shows a decrease. Findings indicate dysregulated Δ HHb profiles had not returned to within 'normal' levels within the GRTP period. R=adult rugby and CR=youth rugby and CR=youth rugby.

head impact load was associated with greater neural activation, suggesting that cumulative head impacts altered NVC responses [39]. The present study shows small but significant changes in NVC responses for both youth groups, indicating that the fNIRS changes are not due to playing rugby and accumulative impacts. As such our findings do not support previous literature [38, 39] that suggest changes in brain function in team sports outside of specific concussive events. Youth participants showed a more variable seasonal profile than adults, possibly because the PFC does not fully develop until the age of ~25 years [30], with increases in PFC activity shown during a working memory task across 7 to 22 years [31]. This highlights the importance of developmental considerations when directly measuring concussive biomarkers.

Dynamic Cerebral Autoregulation (dCA): Squat-Stand

A squat-stand manoeuvre was used to acutely activate the dCA mechanism, which provides information on the brain's ability to control and maintain cerebral blood flow (CBF) [40]. Healthy individuals are able to regulate CBF during orthostatic challenge [41] and no significant differences were seen in adult groups. Adult rugby players showed a trend towards a greater ΔO_2 Hb and Δ HHb during the squat-stand task compared to controls, possibly an effect of training [42]. Youth responses were again more variable, with pre-season ΔO_2 Hb and Δ HHb significantly lower than mid and end-season. Additionally, a significant interaction effect showed that youth control subjects produced higher values at mid-season compared to youth rugby players. This is not easily explained, although it must be noted that the ability to perform the squat-stand manoeuvre well was most challenging in this group. The variability in responses between adults and

Table 4	Shows the graded symptom	checklist (GSC) from	n sport concussion	assessment tool	version 5 (SCAT5)	symptom sever	ity and
symptom	number scores during the e	arly post-injury (< 2	5 days) period				

Participant ID	Visit 1 (<i>n</i> =8)		Vis	Visit 2 (<i>n</i> = 8)		Visit 3 (n = 5)		Visit 4 (<i>n</i> = 1)	
	Symptom No.	Symptom Severity	Symptom No.	Symptom Severity	Symptom No.	Symptom Severity	Symptom No.	Symp- tom Severity	
CR13	15*	39*	2	3					
CR15	21*	56*	10*	13	5	5			
CR3	14*	33*	7	10	3	3	1	1	
CR5	9	17*	12*	23*	8	9			
CR7	12*	16*	4	4					
R13	15*	35*	0	0	0	0			
R16	0	0	0	0					
R18	4	6	1	1	0	0			
No. of Days	4 ± 2		1 7+12		19±6		23±0		

Note: n = indicates the number of participants at each visit. Days indicates the mean numbers of days since concussion ± standard deviation

Symptom severity scale = 0-132; 0-14 'broadly normal', 15-22 'above or below average', 23-39 'unusually low or high', 40-132 'extremely low or high'. Symptom number scale = 0-22; 0-19 'broadly normal', 10-13 'above or below average', 14-18 'unusually low or high', 19-22 'extremely low or high'

* Indicates results greater than broadly normal [35]

youths could reflect developmental differences in autoregulatory mechanisms [43, 44], indicating a need for age-specific considerations in assessing and understanding cerebrovascular responses to sport concussion. historical concussion to sub-concussive impacts highlight the need for ongoing cerebrovascular health moni-

toring in rugby and contact sport players.

Scarce comparative data are available. Studies have reported significant acute alterations in dCA in professional [45] and amateur boxers following sparring [46]. Additionally, significantly increased ΔO_2 Hb in the left PFC in contact sport athletes with a history of concussion [28], and reduced capacity to respond to dynamic changes in CBF over a season in professional rugby [47] and contact sport athletes [48] have been observed. Wallis et al. [46], using the same squat-stand manoeuvre as this study, measured middle cerebral artery blood velocity (MCAv) via transcranial Doppler ultrasound, and observed a significant reduction in phase (timing offset between mean arterial pressure and MCAv) post-boxing. The magnitude of the change was associated with the number of head impacts received and not seen following pad sparring without head impacts. Similarly, Wright et al. [48] applied the same squat-stand protocol and reported significant reductions in phase and increases in gain (response amplitude) over a season of contact sports participation. These changes were linked to cumulative sub-concussive exposure, suggesting that repetitive impacts can impair the cerebrovascular pressure-buffering system. The difference and stability of adult dCA responses in the present study may be the result of differing methods to assess CBF responses and/or related to the population. Collectively, our findings, alongside those discussed, underscore the utility of measuring mechanisms that assess the brain's ability to control and regulate cerebral blood flow (CBF). The observed alterations in dCA across different sports contexts ranging from

Concussion

NVC, dCA and GSC

Eight players experienced concussion and were monitored across the return-to-play period and at end-season. Seven out of eight players exhibited $\Delta O_2 Hb$ and Δ HHb responses during NVC and dCA inducing tasks outside normally expected values i.e., TE. Dysregulated NVC appears to manifest as either reduced ΔO_2 Hb or increased ΔO_2Hb during the cognitive task. Reduced ΔO_2 Hb suggests that the brain's ability to increase blood flow to active regions is compromised post-concussion, while increased ΔO_2 Hb may reflect the recruitment of additional neural resources or compensatory mechanisms. Both patterns are considered abnormal or dysregulated responses, indicating disruption of typical NVC. Importantly, these variations highlight the individual nature of NVC responses post-concussion, rather than a singular 'negative' or uniform pattern. Reduced cerebral activation using the same paradigm has been seen in retired rugby players with a history of concussion [49] and concussed youth sport players have shown differing brain responses, including hyperactivation during cognitive testing [50]. Additionally, concussed adolescents have also shown significantly reduced Δ in the difference between O₂Hb and HHb during a visual-cognitive task [51], which concurs with our findings.

Dysregulation was also seen during the squat-stand manoeuvre form the app six players (four youth, two adult) showing increased and one youth player reduced ΔO_2 Hb and Δ HHb. Retired contact sport athletes with

a history of concussion have also shown significantly elevated ΔO_2 Hb in the left PFC [28] and dCA indices in concussed junior athletes, showed reduced phase (timing) [48]. The auto regulatory mechanism that manages CBF in response to changes in blood pressure appears to be compromised post-concussion, with concussed individuals displaying more pronounced responses to orthostatic challenge.

During the same time period, four players reported increased symptom severity and six players increased symptom number scores on the GSC, all greater than 'broadly normal' normative reported scores [35]. All symptoms had resolved within the GRTP period. Currently, resolution of symptoms forms the approach for clinical recovery within most amateur sports, despite evidence showing limited clinical application after sevendays post-injury [1].

Importantly, alterations in fNIRS responses during NVC (six out of eight ΔO_2 Hb; five out of eight Δ HHb) and dCA tasks (seven out of eight ΔO_2 Hb and Δ HHb) had not returned to within normal values within the GRTP period. Seven out of eight players returned to match play following clinical clearance, with four players' NVC (ΔO_2 Hb) and five players' dCA (ΔO_2 Hb) profiles outside of normal expected values at end-season. One player opted to take a prolonged recovery following concussive injury as they only had one game remaining for the season.

Implications

Our study offers critical insights into the neurocognitive/physiological impacts of a rugby season, with significant implications for player health monitoring, and concussion management. Firstly, we highlight the utility of fNIRS as an objective tool for monitoring cerebral oxygenation. The significant changes observed in NVC and dCA-inducing tasks, particularly among players who experienced concussions, underscore the potential of fNIRS measurements as biomarkers for brain health and concussion assessment. This supports a shift towards more objective, continuous monitoring of brain function in contact sports, rather than relying on subjective symptom reporting and clinical assessments [7, 48]. The persistence of altered fNIRS responses in concussed players beyond the GRTP period, despite the resolution of subjective symptoms, calls for a re-evaluation of current return-to-play guidelines. Incorporating objective measures such as fNIRS into return-to-play protocols could enhance the safety and health outcomes of rugby players. Additionally, age-specific considerations in concussion management, with tailored approaches to monitoring, and managing concussions is required.

Strengths and Limitations

This study is among the first to provide both measurements of brain haemodynamic responses across a season and during the GRTP period post- concussion. The use of a portable, non-invasive technology has enabled fieldbased measurements which facilitates understanding of the effects of cumulative sub-concussive and concussive impacts. However, this study is not without limitations. Measurement, interpretation, and analysis of the fNIRS signal has important considerations; limited penetration depth to reach subcortical structures, limited coverage of the whole brain and confounders to the signal including movement artifact and physiological noise [52]. Stringent pre-processing steps, recommended by recent best practice fNIRS filtering approaches [53], were used to counter the influence of signal confounders. Despite widespread use within the sports science literature [33, 34], the use of TE to provide boundaries of accepted variation has not been employed within the context of fNIRS or concussion research previously. Future experimental designs should employ multiple individual pre-season measurements to establish individualised TE thresholds. Finally, the data analysis approaches presented here are limited to global measurements of the PFC. While this approach was appropriate for the aims of this study, more advanced analysis techniques, such as channel-by-channel or hemispheric-specific approaches, could provide greater insight into the spatial localisation of PFC activity. Such methods may be particularly useful for identifying specific brain regions affected by injury or for exploring potential lateralised effects associated with certain tasks. Future studies could incorporate these techniques to enhance the understanding of the neural mechanisms underlying concussion-related changes in PFC oxygenation.

Conclusion

In conclusion, results indicate current return-to-play protocols are limited and underscore the need for an individualised approach to concussion management, utilising objective biomarker tools. Our findings demonstrate the effectiveness of fNIRS as a method for assessing brain haemodynamic response, and advocate for enhanced protocols that include baseline (pre-season) assessment to prioritise the long-term neurological health of players. This has broader applications for contact sports where players are at risk of concussive and sub-concussive impacts. Future research should continue to explore innovative methods for monitoring brain health in athletes, with a focus on developing safer sport practices and improving player welfare.

Abbreviations

- CI Confidence Intervals
- CTE Chronic Traumatic Encephalopathy
- dCA Dynamic Cerebral Autoregulation

fNIRS	Functional Near Infrared Spectroscopy
GHQ	General Health Questionnaire
GRTP	Graduated Return to Play Protocol
HHb	Deoxyhaemoglobin
MCAv	Middle Cerebral Artery blood velocity
mTBI	Mild Traumatic Brain Injury
NVC	Neurovascular Coupling
O ₂ Hb	Oxyhaemoglobin
PFC	Pre-frontal Cortex
RFU	Rugby Football Union
SCAT5	Sport Concussion Assessment Tool Version 5
SD	Standard Deviation
SRC	Sport Related Concussion
SS	Squat Stand
TE	Typical Error
WW	"Where's Wally"

Supplementary Information

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supplementary material i	

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Author Contributions

BJ drafted the initial manuscript, and conceptualised, designed, and acquired the data and is guarantor of the study. JPN, JS, SW, JAP, EH, JM, and JP conceptualised and designed the study and critically revised the manuscript for important intellectual content. MR, JM, CC, JM and TA made substantial contributions to the conception of the study, the acquisition of data, and critically revised the manuscript for important intellectual content. All authors approved the final manuscript as submitted and agree to be accountable for all aspects of the work.

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Data Availability

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethics Approval and Consent To Participate

This study was reviewed and approved by University of Essex Research Ethics Committee (ETH2122-0997), all participants provided their signed informed consent prior to the start of testing, and the research was carried out in accordance with the Declaration of Helsinki (2013).

Consent for Publication

Not applicable.

Competing interests

BJ, MJ, SW, MR, JAP, JP, EH, JM, TA, JS, JPN and CEC declare they have no competing interests.

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