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IGF-1, C-Reactive Protein, and Skin Temperature Responses to a Non-Contact Team Sport Activity Circuit in Under-20 Elite Soccer Players

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HIGHLIGHTS

- The responses of the IGF-1/CRP ratio can present a biphasic behavior after a non-contact team sport activity circuit in elite soccer.
- This behavior seems to be influenced by the expression of the IGF-1 isoforms, since CRP did not show changes during the studied period, demonstrating that CRP can be a dependent biomarker on "body contact".
- The Tsk of the LL was presented as a sensitive marker to changes arising from the tissue repair process, although it is not possible to identify which phase of this process is in evidence at the time of analysis, then it should be used as a support method.
- This information can be useful to develop monitoring and intervention strategies throughout the tissue repair process due to the physiological demand imposed by training in an elite soccer season.

Abstract: The aim of the study was to evaluate the responses of insulin-like growth factor 1 (IGF-1), C-reactive protein (CRP), IGF-1/CRP ratio, and skin temperature (Tsk) of the lower limbs (LL) of under-20 elite soccer athletes to a non-contact team sport activity circuit throughout 48h. Thirty elite soccer athletes (19.0 ± 1.0 years, 74.3 ± 7.13 kg, 10.3 ± 2.2 %F, 178.1 ± 6.6 cm, 56.3 ± 3.1 mL.kg⁻¹.min⁻¹) were submitted to a team sport activity circuit with intermittent exercises, jumps, direction shifts, accelerations and decelerations. Plasma concentrations of IGF-1 and CRP were evaluated pre-training session (baseline), and immediately after, and at 3, 24 and 48h following exercise. While the Tsk of the LL was evaluated at baseline, and at 24

and 48h following exercise. Plasma IGF-1 concentrations were greater immediately after, 24 and 48h after the circuit compared to 3h ($p < 0.05$; ES= 0.66, 0.72 and 0.70, respectively). CRP values did not change throughout the study ($p > 0.05$). When verifying the IGF-1/CRP ratio, the values at 3h after the activity circuit were lower than those presented in baseline, and immediately after, 24 and 48h after the exercises ($p < 0.05$; ES= 0.53, 0.65, 0.56 and 0.57, respectively). The hot and neutral zones increased the number of pixels at 24 and 48h after the activity circuit, while the cold zone showed an opposite behavior ($p < 0.05$). Changes in the values of blood biomarkers and Tsk of the LL suggest that there is possibly an active tissue repair process throughout 48h following exercise.

Keywords: Tissue repair; muscle damage; inflammation; soccer; high performance.

INTRODUCTION

Elite soccer is an intermittent sport, characterized by high-intensity actions with eccentric contractions, e.g. jumps, sprints, acceleration and deceleration with changes in direction [1-3]. Such actions are related to muscle damage and consequent inflammatory processes [2,3], which needs to be considered when preparing athletes. Proper preparation involves planning, monitoring and controlling training sessions in order to maximize performance and decrease injury risks over the course of a season [4,5]. In this sense, tools and assessments of the load imposed in training and games, as well as recovery processes are very important [6,7].

Assessments of metabolism and muscle status throughout the phases of the tissue repair process are commonly evaluated in elite soccer through blood biomarkers, e.g. testosterone, cortisol, insulin-like growth factor 1 (IGF-1) and C-reactive protein (CRP) [2,3,8]. In addition, the skin temperature (Tsk) of the lower limbs (LL) has been evaluated by means of infrared thermography (IRT), which is a complementary method in prognoses from multiparameter analyzes [7,9,10].

Plasma concentrations of testosterone and cortisol, as well as the testosterone/cortisol ratio (T/C) have been used as they provide information on muscle metabolism [3,11-13]. However, the responses of these hormones can be affected by personal relationships, rivalry between teams and place of match [12,14,15]. These factors can decrease reliability during data analysis and to influence decision making [16,17], therefore, it is necessary to investigate other blood biomarkers for analysis of muscle metabolism.

IGF-1 has been linked to the anabolic processes of muscle metabolism [12, 18-20]. Recent studies demonstrate that IGF-1 participates in the activation of satellite cells and may represent a key factor in the completion of the inflammation phase [21,22]. However, IGF-1 concentrations in elite soccer were evaluated only chronically, in the face of a competitive season [23]. Thus, acute analysis of IGF-1 responses can generate relevant information for understanding the imposed demand and recovery processes.

Inversely to IGF-1, CRP is understood as a biomarker of a systemic inflammatory process [17, 24-27]. The functionality of CRP to bind to the chromatin of damaged cells contributes to macrophages (LyC6⁺) being able to engulf and phagocytose the damaged structure, without exacerbating the microlesion. This mechanism gives CRP a quick and efficient response to inflammatory conditions, as in the case of the stress imposed by soccer training sessions [21, 27,28]. Thus, it is possible that IGF-1 and CRP are inversely related during the tissue repair process, as is the T/C.

Assessing the IGF-1/CRP ratio can be explained as a function of metabolic signals that these markers could be involved. While one is related to a more catabolic content (CRP), the other is related to a markedly anabolic context (IGF-1). However, due to the complexity and multifactorial nature of the recovery processes, it is interesting that the scientific literature reports analyzes of these markers (IGF-1, CRP and IGF-1/CRP ratio), supported by a complementary method, for example, the IRT, that may increase the robustness of knowledges to load control and injury prevention programs.

Considering the characteristics of soccer and that IGF-1 and CRP are directly related to muscle stress, possibly in metabolic different phases [20,29], it would be important to compare the responses of these blood biomarkers in a timeline with some complementary method after an elite soccer training session. Assessing these issues could present a proposal for a muscle recovery monitoring tool and reduce the risks of bias that are currently presented by the scientific literature for the T/C [30-32]. We hypothesized that the IGF-1 and CRP curves would have an opposite behavior after performing the non-contact team sport activity circuit protocol, and there would be a change in the IGF-1/PCR ratio. In addition, CRP responses would present an increase at 24h after the training session and would not return to similar baseline responses throughout the study period, as well as the Tsk of the LL, because these responses were previously reported following soccer games [9,25,44,45]. However, there aren't similar analyses following training, and this investigation may improve the knowledge about team sport training sessions. The aim of the study was to evaluate the

responses of insulin-like growth factor 1, C-reactive protein, IGF-1/CRP ratio, and skin temperature of the lower limbs of under-20 elite soccer athletes to a non-contact team sport activity circuit throughout 48h.

MATERIAL AND METHODS

Subjects

Thirty under-20 soccer players (19.0 ± 1.0 years; 74.3 ± 7.13 kg; 178.1 ± 6.6 cm, 56.3 ± 3.1 mL.kg⁻¹. min⁻¹) underwent a non-contact team sport activity circuit. Subjects were familiar with the protocol of a circuit, but they did not perform these types of exercises at least 1 month before the study. All participants belong to the under-20 category of an elite Brazilian soccer club. The inclusion criteria were: a) being linked to elite clubs in the first division of Brazilian soccer; b) participating in competitions ruled by the sport's federation; c) no history of kidney disease; d) not being under the effect of drug treatment; e) not using any diuretic and antiseptic; not having fever within 7 days prior to the beginning of the study; f) completing the training session. The athletes signed an informed consent form after being informed about all the procedures and risks of the study. This study was approved by the Ethics Committee on Research with Human Beings of the Federal University of Minas Gerais (CAAE: 34943320.3.0000.5149).

Experimental design

On the first day of the study, sample characterization was performed using body composition measurements and maximum oxygen consumption (VO₂max). Body mass in kilograms and height in centimeters were measured with a digital scale with an attached stadiometer (Filizola®, São Paulo, BR). The fat percentage was obtained by the sum of skin folds (subscapular, tricipital, pectoral, axillary-middle, supriliac, abdominal and thigh) collected with an adipometer with a sensitivity of 0.01 mm (Lange®, Cambridge Scientific Industries, Inc., Cambridge, USA). The value of each skinfold was used for the sum of the folds (Σ folds) for body density calculation [33] and percentage of fat [34]. VO₂max was indirectly assessed using the YoYo Endurance Test level 2 [35].

After sample characterization, on the morning of the second day of the study, the first blood collection took place to analyze baseline concentration of IGF-1 and CRP. At this same time, thermal images were collected. Then, athletes performed a non-contact team sport activity circuit. Immediately (0h) and 3h after session training, the second and third blood samples were taken, respectively. On the third (24h) and fourth day (48h) after the exercise, there was, in this order, a fourth and a fifth blood collection. At these times, thermal images were also captured. Throughout the study period, from sample characterization procedures up to 48h after session training, the subjects remained lodged within the club facilities, where they ate all meals following instructions of a dietary nutritionist.

A non-contact team sport activity circuit

Initially, athletes performed a warm-up and then completed twice a circuit consisting of five stations with intermittent exercises combining jumps, direction shifts, accelerations and decelerations [3]. Length of permanence in each station lasted 3min with a break of 30s rest for station changes. The activities were carried out at maximum speed and athletes were verbally encouraged by coaches. The five stations performed during the training session were as follows: 1) The first station consisted of jumping with both feet together from a 45cm high platform and, in sequence, jumping over another 30cm high obstacle. Soon after, the athlete ran in a straight line at maximum speed a 10m distance demarcated with cones; 2) At the second station, the athlete covered 20m at maximum speed, jumping four 20cm high obstacles with both feet together, arranged in a rectilinear way, with a distance of 1.5m between them; 3) Next station started with the athlete performing the drop jump, then he performed lateral jumps over a 30cm high obstacle, positioned on the sides of the platform, once with the right lower limb and once with the left one. Right after the jump, the athlete covered a 20m path at maximum speed, with direction shifts pre-fixed by cones. At the end of this distance, the athlete makes a sudden brake; 4) The path covered in the fourth station was demarcated with cones. Participants ran 10m in a straight line, turned left and then another 5m, then returned running 10m in a straight line, turned right running 5m and then in a straight line for another 10m, totaling 40m distance with abrupt and fast shifts in direction; 5) At the end of the jumps, the subjects covered 10m at maximum speed until the end of the path (see Figure 1).

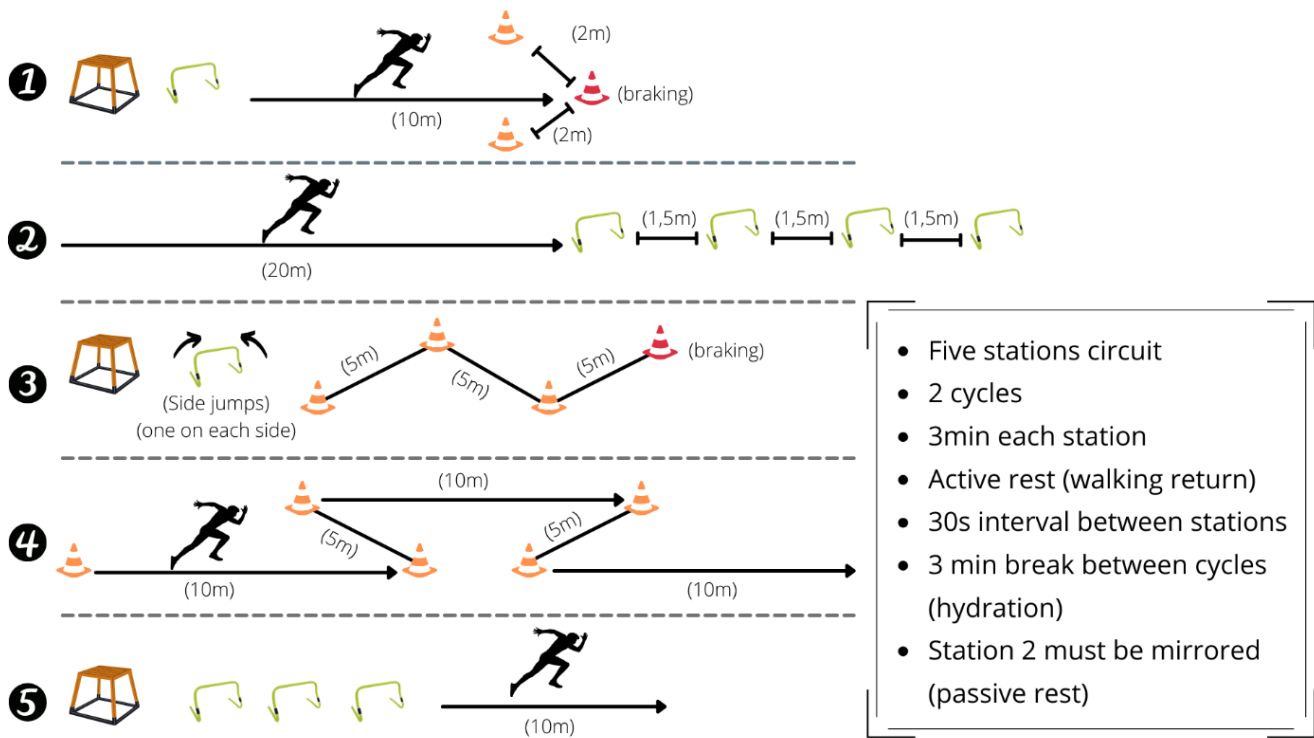


Figure 1. Non-contact team sport activity circuit protocol

Total distances

Analysis of the non-contact team sport activity circuit load was recorded by registering heart rate (HR) and other variables such as total distance covered, average speed and high-intensity actions. These variables were recorded using GPS devices integrated with branded heart rate monitors (MinimaxX S5 units®, Catapult, Orlando, USA) with operational characteristics previously described [36]. Each athlete used the device fixed in the cavity of a specific vest, positioned on the athlete's back. GPS devices were activated at least 20 min in advance for initial recognition of the signals by the device. Data was recorded and analyzed by specific software (OpenField 1.8). The environmental conditions (dry and wet) of all the training session were registered through the digital Thermo-Hygrometer (Instrutherm® HT-260, São Paulo, BR) [3]. Registered temperature was 27°C and the relative humidity was 68%.

CRP and IGF-1 concentration determination

The blood samples (~10mL) were taken by antecubital arm vein, at predetermined periods described above and collected in tubes with separator gel (Venoject II, Terumo Europa®, Leuven, BE) [3]. The serum was isolated by centrifugation (1.500g, 4°C, for 15min). The resulting serum was placed in microtubes (Eppendorf do Brasil Ltda®, São Paulo, Brazil), separated in multiple aliquots and frozen at -80°C [3] for further analyses of the IGF-1 plasma and CRP concentrations analyzes. IGF-1 plasma concentration analyzes were performed using the radioimmunoassay method. The turbidimetry method was used to assess CRP concentrations, using highly sensitive kits (Quantikine®, R&D Systems, Minneapolis, USA). Two nurses and two physicians from the club's medical department performed venipuncture. For statistical analysis, the data of concentrations of IGF-1, CRP and IGF-1/CRP ratio were relativized according to the equation 1.

$$\text{Relativized value} = (\text{moment value}) / (\text{higher individual value}) \times 100 \quad (1)$$

Thermal images capture

The collections were carried out in a specific room, with a 22°C room temperature and a relative humidity of 65% [10]. The emissivity index was adjusted to 0.98, using a black background [37,38]. The thermal imager was kept at a distance of 1.5m from the athlete, who wore only swim trunks [37,38]. Before the capture of thermal images, participants remained 10min inside the collection room to achieve a thermal balance [39]. Two thermal images (posterior and anterior view of lower limbs) were captured using a thermal imager (FLIR®, T1020, Stockholm, SE), with a measurement range of -20 to + 120°C, the accuracy of 1%, sensitivity ≤ 0.02°C, the infrared spectral band from 7.5µm to 13µm, refresh rate of 60Hz, with automatic focus and

FULL HD resolution. Finally, the images were selected and visualized in the specific software (APOLLO® version 1.0, BR) for analysis using the thermopixelgraphy method. This method allows checking the frequency of the number of pixels and their percentage by temperature ranges [40,41], grouping them into three temperature zones: cold ($< 30.99^{\circ}\text{C}$), neutral (31°C to 32.99°C) and hot ($\geq 33^{\circ}\text{C}$) [42].

Statistical Analyses

Data are presented as mean \pm standard deviation. The Shapiro-Wilk test was used to check data for normal distribution. As IGF-1, CRP, IGF-1/CRP ratio and Tsk data did not present normal distribution, time differences were analyzed using the nonparametric Friedman test, and pairwise comparisons were performed using the Dunn's method. To allow a better interpretation of the results, the effect size was calculated using the method for non-parametric data suggested by Field [43]. Effect size was classified as trivial (<0.2), small ($0.2-0.49$), medium ($0.5-0.79$), and large (≥ 0.8). The statistical package used for all the analyses was Statistical Package Social Sciences, 21 version (SPSS Inc®, Chicago, IL). The significance level was at 0.05.

RESULTS

Internal and external load of the non-contact team sport activity circuit are shown in Table 1.

Table 1. Internal and external load of the training session.

| Variables | Mean (DP) | Minimum | Maximum |
|---|---------------|---------|---------|
| Distance (m) | 3.102 (166.2) | 2.702 | 3.377 |
| High intensity running (m) | 160.6 (37.3) | 86.5 | 270 |
| Sprints ($\text{m}\cdot\text{s}^{-2}$) | 20 (5) | 13 | 31 |
| High intensity accelerations (m/s^2) | 61 (7) | 50 | 72 |
| High intensity decelerations (m/s^2) | 52 (9) | 30 | 72 |
| Total high intensity actions ($\text{m}\cdot\text{s}^{-2}$) | 132 (18) | 90 | 175 |
| %HR _{máx} | 79.8 (3.4) | 75.5 | 90.2 |

There was a significant time main effect ($\chi^2 = 24.33$, $p < 0.001$) for IGF-1 blood concentration in response to the non-contact team sport activity circuit (Figure 2). The post hoc analysis showed that IGF-1 concentrations were greater immediately post, 24h and 48h post-circuit in comparison to 3h ($p = 0.003$ and $ES = 0.66$, $p = 0.001$ and $ES = 0.72$, $p = 0.001$ and $ES = 0.70$, respectively). No significant difference was found between the other comparisons ($p > 0.05$).

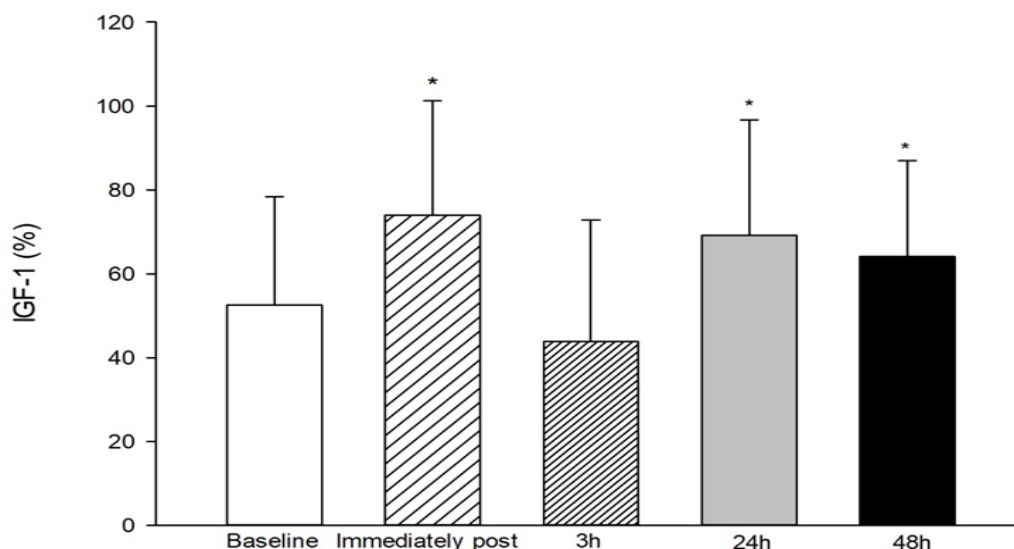


Figure 2. Mean \pm SD of IGF-1 blood concentration pre-session training (*baseline*), immediately post, 3h, 24h and 48h post-circuit. (*) $p < 0.05$ greater than 3h.

No significant time main effect was found for CRP blood concentration in response to the non-contact team sport activity circuit ($\chi^2 = 7.58$; $p > 0.05$; see Figure 3).

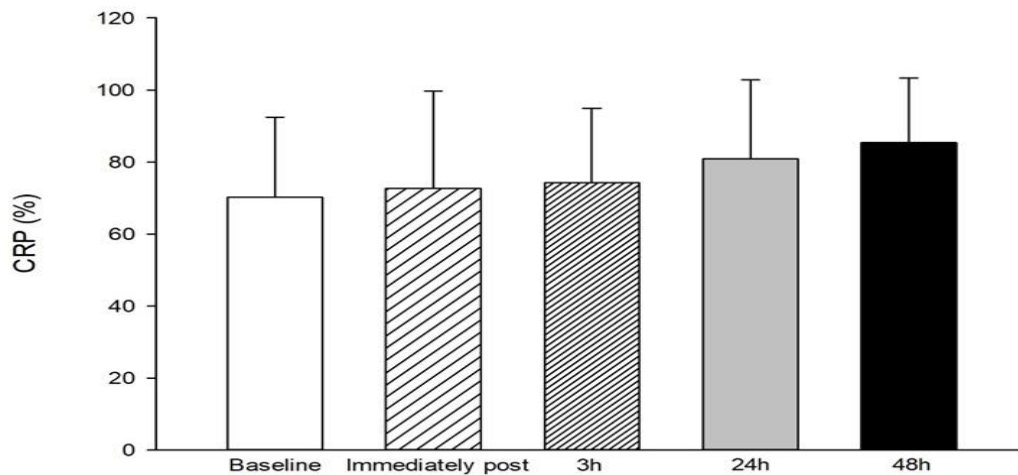


Figure 3. Mean \pm SD of CRP blood concentration pre-session training (baseline), immediately post, 3h, 24h and 48h following post-circuit.

Figure 4 shows IGF-1/CRP ratio pre-circuit and up to 48h after the non-contact team sport activity circuit. A significant time main effect was observed ($\chi^2 = 16.62$, $p = 0.002$). IGF-1/CRP ratio was lower 3h post-circuit in comparison to baseline values ($p = 0.037$ and $ES = 0.53$). In addition, it was greater immediately post, 24h and 48h following the training session in comparison to 3h ($p = 0.004$ and $ES = 0.65$, $p = 0.022$ and $ES = 0.56$, $p = 0.017$ and $ES = 0.57$, respectively). There was no significant difference between the other times ($p > 0.05$).

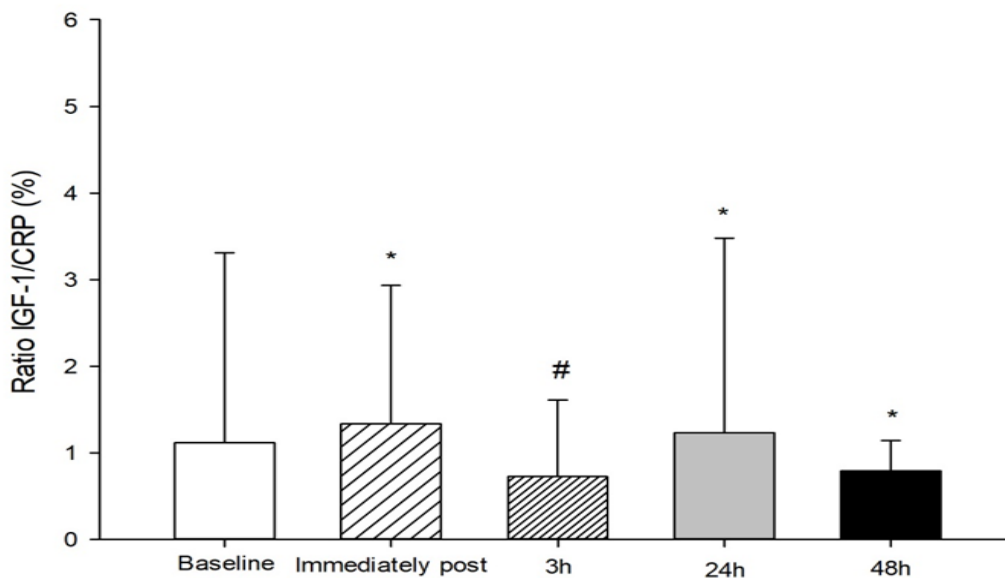


Figure 4. IGF-1/CRP ratio pre-session training (baseline), immediately post, 3h, 24h and 48h post-circuit training. (*) $p < 0.05$, greater than 3h. (#) $p < 0.05$, lower than baseline.

The pixels count in anterior and posterior views of the LL are shown in Table 2. There was a significant time main effect for Tsk of the anterior view for all zones (hot [$\chi^2 = 56.3$, $p < 0.001$], neutral [$\chi^2 = 23.1$, $p < 0.001$] and cold [$\chi^2 = 78.4$, $p < 0.001$]). Hot zone and neutral zone showed greater pixels at 48h after training session when compared to baseline ($p < 0.001$ and $ES = 0.50$, $p = 0.001$ and $ES = 0.42$, respectively) and at 24h values ($p < 0.001$ and $ES = 0.32$, $p < 0.001$; $ES = 0.51$, respectively). However, the cold zone showed lower pixels at 48h post-circuit in comparison to the baseline values ($p < 0.001$, $ES = 0.72$) and at 24h post-training session ($p < 0.001$, $ES = 0.67$).

There was also a significant time main effect for Tsk of the posterior view for all zones (hot [$\chi^2=31.9$, $p<0.001$], neutral [$\chi^2=20.2$, $p<0.001$] and cold [$\chi^2=47.2$, $p<0.001$]). Hot zone and neutral zone showed greater pixels 48h after circuit when compared to baseline ($p<0.001$ and $ES=0.37$, $p=0.001$ and $ES=0.39$, respectively) and at 24h values ($p<0.001$ and $ES=0.37$, $p<0.001$; $ES=0.29$, respectively). Nevertheless, the cold zone showed lower pixels at 48h after the training session in comparison to the baseline values ($p<0.001$, $ES=0.58$) and 24h post-circuit ($p<0.001$, $ES=0.50$).

Table 2. Mean \pm SD of the number of pixels, in anterior and posterior view of lower limbs, separated for temperature zones throughout 48h after a circuit training session.

| | Mean (SD) | | | ES | | |
|-----------------------|-----------|-----------|-------------|-----------------|-----------------|------------|
| | Baseline | 24h | 48h | Baseline vs 24h | Baseline vs 48h | 24h vs 48h |
| Anterior view | | | | | | |
| Could zone | 218 (96) | 203 (74) | 141 (54)*† | 0.05 | 0.72 | 0.67 |
| Neutral zone | 235 (167) | 250 (160) | 289 (133)*† | 0.09 | 0.42 | 0.32 |
| Hot zone | 65 (78) | 72 (85) | 135 (135)*† | 0.01 | 0.50 | 0.51 |
| Posterior view | | | | | | |
| Could zone | 214 (111) | 196 (85) | 141 (84)*† | 0.08 | 0.58 | 0.50 |
| Neutral zone | 235 (167) | 255 (180) | 301 (136)*† | 0.10 | 0.39 | 0.29 |
| Hot zone | 31 (59) | 36 (70) | 53 (80)*† | 0.01 | 0.37 | 0.37 |

(*) $p<0.05$, different from baseline; (†) $p<0.05$ different from 24h

DISCUSSION

In present study, the initial hypotheses were partially supported. The main result of this study is that the IGF-1/CRP ratio showed a biphasic behavior, and probably this was due to the expression of IGF-1, which alters throughout phases of tissue repair. In this case, this is first study that analyzed the IGF-1/CRP ratio after a non-contact team sport activity circuit in elite soccer, thus, the results found cannot be directly compared with previous studies. Surprisingly, CRP concentrations remained unchanged throughout the study, while we waited these levels were modified, mainly in 24h and 48h after exercise, because it has been reported in elite soccer players [25,26,44,45]. Additionally, the results of Tsk of lower limbs reached peak values for the hot and neutral zones at 48h after a non-contact team sport activity circuit., but we waited that these peak values were happen at 24h after exercise, like previously reported [37,42,45].

The tissue repair process can be divided into five phases. Two with catabolic content (degeneration and inflammation) and three with anabolic content (regeneration, maturation and functional recovery [20]. Considering the physiological stress imposed by the training session and the interval between them due to the congested calendar of Brazilian elite soccer, it is believed that, in general, the phases of maturation and functional recovery can be impaired [5,14]. Therefore, it is extremely important to understand how tissue repair phases are related and which mechanisms regulate their activity. IGF-1/CRP ratio showed the higher values immediately post, and 24 and 48h after training compared to 3h. The other analyzed moments did not show changes. As CRP results did not presented changes after training, this fluctuation in IGF-1/CRP ratio curves can have occurred due to IGF-1 concentrations and isoforms [46-49].

The increase in IGF-1 concentrations seems to be related to eccentric actions so that exercise-induced muscle damage is a stimulus for its activation [29, 50]. And then, a biphasic response can be presented acutely (lasts for approximately 3 days) by this biomarker [48]. The biphasic theory of IGF-1 is based on its three isoforms: muscle growth factor (MGF), IGF-1Ea, and IGF-1Eb (this last is expressed only in animal

models) [48]. The studies carried out so far indicate that the physiological mechanisms that act on different actions may be the activation pathways of each isoform, as well as its functionality [46-49].

The MGF is recognized for being activated in response to exercise or stretching, in the muscle tissue itself and acts in an autocrine and paracrine way [48]. While IGF-1Ea is the isoform synthesized in the liver and its main interaction is paracrine [49]. The biphasic theory of IGF-1 shows that the MGF response is different from the IGF-1Ea response [48]. Right after the exercise, the IGF-1 is presented by the MGF isoform and after a day or more, this presentation is made by the IGF-1Ea [48]. The main function of MGF is mitogenic (replenishment of the satellite cell pool, activation, and proliferation for the moment of muscle regeneration), while IGF-1Ea is responsible for the myogenic action (differentiation of proliferated satellite cells) [48,49]. In addition to different functions, different signaling pathways are associated with the responses of these isoforms. In this context, the Ras/Raf-1/MAPK pathway mainly mediates cell proliferation, while the differentiation pathway is stimulated by the PI-3 kinase/p70 s6 kinase [46]. The exact mechanisms that generate the reversal of expression between IGF-1 isoforms are not yet clear. But it is believed that MGF itself opposes the differentiation of precursor cells [48], and then, MGF concentrations decrease, while IGF-1Ea concentrations increase. In the present study, the IGF-1 concentrations were changed, with lower levels at 3h after the activity circuit than at other moments, possibly due to the IGF-1 biphasic responses that were described before.

Surprisingly, CRP showed no changes over the 48h after the training session. CRP has a high concentration during the inflammation phase, due to its functionality to bind to the chromatin of injured cells so that macrophages in the LyC6⁺ isoform can engulf and phagocytose the damaged structure, without exacerbating the muscle damage [27,28]. The results of the present study differ from previous studies that observed the peak of CRP concentrations 24h after a professional soccer match, with a return to baseline values within 48h [26,51]. Mohr [25], observed that the 72h interval was not sufficient for the complete recovery of the athletes, who presented higher CRP concentrations ($p < 0.05$) in a sequence of three games a week. Peake CRP concentrations were at 24h post games, and the CRP concentrations were higher than baseline and control group just at 48h for the second match, which has a lower interval between games, it is reported that this could be an effect of the accumulated load [25].

In contrast, studies that evaluated high-intensity protocols "with body contact" and "without body contact" on markers of degeneration and inflammation, observed an increase in markers of the degeneration phase (creatinine kinase) for both groups, however the increase in values of CRP was only observed for the group "with body contact" [52]. Considering this information, it can be that the elevations in plasma concentrations of CRP are dependent on a certain degree of contact during the exercise performed. This information may be corroborated by a recent study that demonstrated changes in creatine kinase, but not in CRP, in response to a high-intensity "no body contact" protocol in semiprofessional athletes [53].

Different factors can affect CRP concentration and a possible explanation for the divergence of results between studies presented and the current work may be related to methodological differences between the studies. The intensity, duration and characteristics of the stimulus proposed in this study can have been enough to demand a longer period on the degeneration phase and, consequently, have generated a delay in the inflammatory process [3,44,55]. The type of exercise evaluated by the studies was divergent, so it may be that when implementing a different demand, the evaluated responses are also different or maybe, since CRP has a half-life of 19h, it is possible that even at a lower magnitude, there were changes in concentrations between the 3h and 24h moments, and this was not possible to verify. Such reasoning could be reinforced by the findings of Thermography, which reported increases in Tsk at 48 hours after the activity circuit".

About Tsk of the LL, the results of the present study showed that 48h post-circuit, the number of the pixels in the hot zone and in the neutral temperature zone reached the maximum values. These results differ from previous studies that demonstrated that Tsk reaches its peak values 24h after a soccer match and returns to baseline values within 48h [9,37]. Tsk is related to the increased blood supply and movement of subpopulations of the immune system that will participate in the tissue repair process, increasing the energy radiated by the skin [38]. Neutrophils, monocytes (differentiating into macrophages), cytokines, myokines and growth factors are recruited to the micro lesion site [20,54]. Based on this assumption, the increase in Tsk of the lower limbs at 48h, but not in CRP, may have been influenced by the increased movement of other subpopulations involved in the degeneration phase, such as creatine kinase and IGF-1, for example. However, as creatine kinase was not measured, we cannot affirm this finding.

Finally, the effect size of a non-contact team sport activity circuit was greater for Tsk in the anterior in comparison to the posterior view (Table 2). This can be explained by the characteristic of the training session performed (e.g. composed of jumping, high-speed sprinting, plyometrics) which can further overload the anterior musculature of the LL [56,57]. This result shows a relationship of dependence between the tissue repair process and the higher concentration of pixels in the extreme value bands of the lower limbs Tsk. As the present work is the first to evaluate the use of the thermopixelgraphy method after a training session with a non-contact team sport activity circuit in elite soccer players, further studies should be carried out to verify the responses after official matches with different categories, levels, and other modalities.

This study was a multiparameter observational design, composed of a non-contact team sport activity circuit with muscle recovery analysis throughout 48h, seeking to offer greater ecological validity. However, future studies should analyze a period of 72h or more to elucidate the metabolic actions of the tissue repair process over a longer period [20]. Additionally, the intensity of actions developed by high-performance athletes during training is greater than among athletes of other levels and different categories during official matches, which in turn can influence the physiological demand [58,59]. Therefore, the findings of the present study must be applied with caution.

As practical implications, the results of the present study demonstrated that the Tsk of the LL was presented as a sensitive marker to changes arising from the tissue repair process, even if it is not possible to identify which phase of this process is in evidence. While the IGF-1/PCR ratio showed a biphasic behavior due to IGF-1 isoforms. Therefore, Infrared Thermography proved to be a support method for monitoring muscle recovery. In addition, further studies are needed to assess the applicability of the IGF-1/PCR ratio considering different demands (e.g. small games and official matches). This information may be useful for technical staff who are in charge of elite soccer teams that work in monitoring and intervention throughout the tissue repair process due to the physiological demand imposed by training in an elite soccer season.

CONCLUSION

The responses of the IGF-1/CRP ratio can present a biphasic behavior after a non-contact team sport activity circuit in elite soccer. This behavior seems to be influenced by the expression of IGF-1 isoforms, since CRP did not show changes during the studied period, demonstrating that it can be a biomarker dependent on "body contact". The Tsk of the LL was presented as a sensitive marker to changes arising from the tissue repair process, although it is not possible to identify which phase of this process is in evidence at the time of analysis, so it should be used as a support method. This information can be useful to develop monitoring and intervention strategies throughout the tissue repair process due to the physiological demand imposed by training in an elite soccer season.

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