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Pre-Season Training Affects Negatively the Immunological Parameters and Creatine Kinase but Not Power Performance in Young Soccer Players

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Abstract

The training content, volume and intensity are determinants on the training load understand, since it can lead to a chronic fatigue status that affect biochemical, immunological and performance variables. This study aimed to evaluate changes in the creatine kinase (CK) activity, white blood cells subpopulations and lower-limb power performance of U19 soccer players in accordance of the training content during pre-season period. Fourteen U19 soccer players (18.40 ± 0.88 years, 179.52 ± 6.96 cm, 73.70 ± 7.22 kg) were evaluated in two moments: M1 - at the beginning of pre-season; M2 - at the final of pre-season (beginning of the 11^{th} week) with performance collections and blood analyses white blood cells (WBC) subpopulations and CK to compare to the reference interval (RI) and the Reference Change Value ($RCV_{95\%}$). Weekly all training content was registered and transferred to the researcher. We use the countermovement jump (CMJ) test to measure the lower limbs power. Daily it was collected the rate of perceived exertion (RPE) of each player for load calculations. Paired Student t-test was used for the immune variables, and the Wilcoxon non-parametric test was used to the CK sample. The significance level selected was $\alpha \leq 0.05$. Effects sizes were calculated with the Cohen's d coefficient. There was a specific endurance predominance (28 sessions; 2075 min), followed by general and specific strength (26 sessions; 1480 min). Significant lymphocytes decreases and significant CK increases, but not below and above to the reference intervals, respectively. One athlete to the lymphocytes and three athletes to the CK were above of the $RCV_{95\%}$. No changes in the power performance were finding. The data indicate that the content training applied to the athletes on the preseason decreased the immune response for one athlete, increase the muscle damage for three athletes and maintain the muscle power.

Keywords

Soccer, Training content, Training load, Immunological parameters, Muscle damage, Power performance

Introduction

In soccer training, the volume and intensity are important components of the load. The training load is divided in external and internal load. External load is the training process and content prescribed by coaches expressed in time units, and internal load is the adaptation process of training from stimulus applied [1,2]. To improve the adaptation process it is important to recover properly after a training stimulus [2,3]. When volume and intensity of training increase substantially

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by the changing in the training content, that is, the training exercises or activities by mean of which is intended achieve certain objectives that are directly linked to the load changes in the training schedule, the performance can decrease and athletes are affect by acute fatigue [4,5]. The decrement of performance is due to a muscle damage and additionally start an inflammatory response to regenerate the tissue and make a positive adaptation [6].

On the other hand, a continuous increase in the volume and intensity training variables, with an insufficient recovery periods can lead to a chronic fatigue status that affect biochemical, performance, psychological, physiological and immune variables arising by a severe tissue damage [7]. Regarding to the biochemical field, some proteins in the muscle cells, like creatine kinase (CK), are release to the bloodstream, being considered as a marker of muscle functionality [8] when muscle damage is evidenced.

Also, in the last years has been increased the number of studies about immune response of subjects that was submitted in a systematized training process. These studies presuppose that regular physical exercise can decrease risk of upper respiratory tract infections (URTIs) [9-12]. On the other hand, excessive training load without adequate recovery can lead to a decrement on the immune response of the athletes, increasing the risk of URTIs suggested by the researches as a "J" hypothesis curve [13-15].

The soccer is an intermittent modality with high and low activities during a match, depending on various factors like technical, tactical, psychological and physical [16]. As for the physical component, players run an average of 10-12 km by match, with high intensity actions executed each 72 seconds with duration of 2-4 seconds each one [17] demonstrating that the game has a lot of concentric and eccentric muscle actions that can be in relation with the muscle damage incidence [18,19]. Given this, the soccer training programs need vary in accordance with the annual planning to give and maintain the player's conditioning. Frequently, pre-season is the more training load period of the year with the athletes training until twice daily [20]. For this reason, the pre-season is consider the period of the training that the athletes are more exposed to excessive training load and, therefore, to an increased risk of muscle damage, that can lead to a greater chances of falls in the immune system and the performance.

Thus, the aim of this study was to evaluate the changes in the CK concentrations, white blood cells subpopulations and lower limb power performance of U19 soccer players in accordance of the training content and load during the pre-season period.

Methods

Experimental approach to the problem

This study was a one-group pretest-posttest design because there are some problems with intervening protocols in a team since the team management would not allow randomization because of the analyzed period has been an important tournament preparation period. Besides this, a control group was neither feasible nor appropriate, as such to find another group with the same volume and quantity and that would accept the blood analysis at the same training period in our country is very hard. So, because this, the study it can only be a descriptive and thus report changes in the analyzed variables [21].

In this work, we analyzed the changes on the immunological, CK, and performance parameters when soccer athletes are submitted by continuous 10-week high-intensity and volume training. So, this research was conducted during São Paulo Junior Cup pre-season period, an important championship of amateur Brazilian soccer players. We divided the analysis between two moments: M1 - at the beginning of the pre-season; M2 - at the final of pre-season (beginning of the 11^{th} week). The pre-season period was realized during the October/November months.

We submitted the athletes at two days of collections at the two moments (M1 and M2), where the first was to blood samples and the second for the performance (countermovement jump test - CMJ). Blood samples was collected eight hours fasting at morning (07:00 a.m.) 48 hours after the last session training. One day after this we realized the CMJ before the session training of the day. Following this measures of M1, during all weeks until the M2 the training content was register diary and transferred to the researchers weekly with training load and intensity. For this training variables, diary all RPEs of the training sessions was collected and registered by coaches. Also, diary the URTIs relates are noted and archived to be collected weekly by the researches. So, every week the researches had the athletes self-related upper respiratory tract infections by the symptoms.

Informed written consent was obtained from the athletes and their parents (when the subject was not greater than 18-years) before the testing procedures and this study was approved by the University Ethics Committee.

Subjects

Fourteen U19 soccer players participated of this study (18.40 ± 0.88 years; 179.52 ± 6.96 cm; 73.70 ± 7.22 kg). All the athletes had an average experience of three years in soccer. Before the pre-season period the subjects passed by a transitory period of three weeks of resting and they

Table 1: Physical abilities trained and main features of the training content over 10-week pre-season analyzed training period.

Physical Capacities	Exercises Description
General Strength	- Resistance exercises (with weight) for lower and upper limbs and abdomen, 2×/sem; - Until the 5th week: upper limbs: 3 × 8-12RM; 45"- 1min of rest; lower limbs: 3-5 × 6-12RM; 45s-1min of rest; abdomen: 3 × 15Rep; 45"- 1min of rest; - Weeks 6-10: upper limbs: 3 × 6-10RM; 45s-1min of rest; lower-limbs: 2-3 × 6-10RM; 45"- 1min of rest + plyometric;
Special Strength	- Exercises circuit performed at soccer field without the ball, consisted by strength weightlifted exercises for lower limbs, maximal speed exercises, reactive plyometric exercises, strength upper limbs exercises, acyclic displacement maximal speed with and without ball; - Each athlete performed two passages at the circuit, with 30" execution/20" of rest and 5min of recovery between the passages. Total of 50 stimulus; - Energetic systems solicited was: ATP-CP + Glycolytic + Aerobic (Lactic and alactic anaerobic system/ Aerobic);
Specific Endurance	- Technical/Tactical exercises performed at soccer field with ball: Defensive and offensive organizations and transitions; - Friendly matches and training games: 11 vs. 11; - Energetic systems solicited was: ATP-CP + Glycolytic + Aerobic (Lactic and alactic anaerobic system/ Aerobic);
Speed	- Technical exercises performed at soccer field with ball: Isolated and/or chained elements; integrated defensive and offensive technical elements; - Anaerobic energetic systems requested: ATP-CP (Alactic Anaerobic);

would start the training period for the championship after these three weeks.

Blood collection

At the morning before the first session of training at the M1 and 48 h after the last training session of the M2 period, the blood samples were collected by a eight hours fasting at 07:00 p.m. in both times of the experimental. Antecubital vein 14 mL blood was collected and stored in two Vacutainer® tubes (Becton Dickinson LTd, UK) being one to CK analysis (10 mL) with gel separator (Vacuette® - Greiner Bio-one) and another with EDTA/K3 anticoagulant for White Blood Cells (WBC) analysis (4 mL). The CK (U/L) was analyzed by spectrophotometer UV® (Biospectro, SP-220, Brasil). The WBC cells was made by automatic cells counter (BX Micros 60 - CT; ABX Diagnostics, Irvine, CA®).

Upper respiratory tract infections questionnaire

Weekly the researches receive the URTIs self-related collected by the athlete's coaches accordingly with the procedures used by Tsai, et al. [22]. The participants were asked to fill out a health checklist each day and at the weekend the coaches gave the information to the researches. URTI was recorded when the athletes reported two or more flu symptoms, such as fever, headache, sore throat, sneezing, stuffy nose, nasal discharge, cough, malaise and chilliness, for at least two consecutive days.

Performance of lower limb power

The countermovement jump (CMJ) with upper limbs help was made to verify the performance of lower limbs power by the jumping eight one day after the blood collection using a contact mat (Jump Test Fit®) with results presented by specific software (Jump Test Pro 2.1®). The athletes realized a ten-minute warm-up before the test that consisted of running exercises like jogging, jumping change of direction, coordinative and speed exercises. They didn't do stretches exercises before the test so as not to take the risk of interference in the test. So, each player performed three maximal power jumps and the greater value was used to the data analysis. Since that the club's athletes investigated had familiarity with this technique and due to this test be common in the training athletes routine, the researches decided use it in the study to find some interference of the training content in the power performance together with the blood markers. So, all the athletes had familiarity with the test procedures.

Content training

The training sessions happened diary with a mean duration of 94 minutes. All the content training applied was register and transferred for the researchers to monitor what physical abilities were trained by this period, as well as the diary training volume. For us understanding we standardized four physical abilities: General strength, special strength, specific endurance and speed. The Table 1 shows what was compounded each capacity.

Training load variables

To measure the session training intensity we used the RPE values based on the general training session context, like present in Table 2 [4]. The training load was calculated by the product of session RPE and it volume. Furthermore, mean training weekly load and total weekly training load was calculated. Also we used the training monotony and strain by the following steps:

Monotony = ($\overline{X}$ *Load*) ÷ ($\overline{S}$ load standard deviation)

- Strain = (Monotony) ÷ (Total Weekly Training Load)

Statistical Analysis

A normality of Shapiro-Wilks data analysis was made to verify if the data follow a parametrical standard. To verify the changes in immunological and performance parameters we applied the paired Student t-test. For the CK analysis we used the Wilcoxon non-parametric test. For the training content number of sessions and volume comparisons we used the Qui-Square for contingency samples. The significance value fixed was $\alpha \leq 0.05$. The Cohen's d was used to the effect size and the values were classified like "small" to 0.2 until 0.5 values; "medium" effect with the values between 0.5 to 0.8; and ≥ 0.8 like a "large" effect. The blood markers were classified by the Reference Intervals (RI) and the Reference Change Values ($RCV_{95\%}$) accordingly Nunes, et al. procedures [23] when a significant differences between the moments was found. After this, the outliers was punctuated to compare with the $RCV_{95\%}$ values. The BioEstat 5.0® and the Microsoft Excel® 2010 were used to the statistical analysis.

Results

The Table 3 show the number of sessions in absolute and relative values and total absolute and relative time of the content training at the pre-season, distributed by general and special strength, specific endurance and speed. At the analyzed period, we observed that the specific endurance content was the most frequent training content at the 10-week analyzed period.

The Table 4 shows the RPE values, mean load, mean volume, total load, monotony and strain determined over the 10-week pre-season period and the Table 5 in turn, shows the predominant physical abilities, weekly total volume and physical abilities' training volume, percentual of the training time of the predominant ability, RPE, mean load, total load, monotony and strain, observed at the 10-week training over the pre-season period. From this data, we verify that average RPE of the sessions was "hard", with a total volume of 4325 min of training and a total load of the 25939 UA. Of the 10-week analyzed, seven was predominantly specific endurance, and only three was general and special strength.

The Table 6 shows the Reference Intervals, 90% Confidence Interval, subjects, mean, standard deviation, t-test, Wilcoxon test, p-value and Effect Size (Cohen's d) values of the blood markers of the athletes' pre and post training period. The results showed that the Lymphocytes (t_{lym} = 2.46; p = 0.0284; Cohen's d = 1.23 - "large") and the CK (z_{CK} = 2.04; p = 0.0413; Cohen's d = -0.64 - "medium") decrease and increase significantly, respectively.

As we found these differences, we classified the data like presented in the Table 7 and Table 8 with the mean, standard deviation and coefficient of intra-individual lymphocytes and CK variation (Δ%) between the two moments evaluated. The bold results represent the outli-

Table 2: RPE scale modified by Foster, et al.

Rating	Verbal Description
0	Rest
1	Really Easy
2	Easy
3	Moderate
4	Sort of Hard Hard
5	Hard
6	-
7	Really Hard
8	-
9	Really, Really Hard
10	Maximal

Table 4: Intensity values (RPE), mean load, mean volume, total load, and total training volume determined over the pre-season.

Variables	Values
Mean RPE (UA)	6.00 ± 1.47
Mean Volume (min)	88.27 ± 27.47
Total Volume (min)	4325 ± 51.55
Mean total load (UA)	2593.99 ± 547.47
Total load (UA)	25939
Monotony (UA)	1.10 ± 0.16
Strain (UA)	2919.55 ± 989.65

Table 3: Sessions number and total absolute and relative time of content training applied to each physical ability during pre-season period.

Physical Abilities	Sessions		Time (min)	
	Absolute	Relative (%)	Absolute	Relative (%)
Specific Endurance	28*	43.75*	2075*	50.61*
Strength (general and special)	26	40.63	1480	36.10
Speed	10	15.63	545	13.29
Total	64	100	4275	100

*p ≤ 0.05.

Table 5: Predominant physical abilities, weekly total volume, physical abilities' training volume, percentual of the training time of the predominant ability, intensity (RPE), mean load, total load, monotony and strain observed at 10-week training analyzed over the pre-season.

Variables	Weeks									
	1	2	3	4	5	6	7	8	9	10
Predominant Capacity	Strength	Specific Endurance	Strength	Specific Endurance	Specific Endurance	Strength	Specific Endurance	Specific Endurance	Specific Endurance	Specific Endurance
TV of training at the week (min)	470	485	515	380	380	430	360	440	300	340
PATV at the week (min)	215	250	260	230	240	195	160	200	200	250
Percentual value (%)	46%	52%	50%	61%	63%	45%	44%	45%	67%	74%
RPE ($\bar{X}$ ± DP)	6.69 ± 1.20	4.85 ± 2.74	6.41 ± 1.33	5.90 ± 1.04	7.03 ± 1.55	6.00 ± 1.01	5.50 ± 0.58	6.53 ± 1.28	5.98 ± 1.05	5.44 ± 1.85
Weekly Mean Load ($\bar{X}$ ± DP)	463.56 ± 378.63	341.49 ± 326.53	483.93 ± 403.07	364.70 ± 308.93	279.86 ± 353.86	414.76 ± 344.46	275.55 ± 187.14	461.42 ± 364.44	351.96 ± 293.55	268.46 ± 296.40
Total Load (UA)	3244.92 ± 606.98	2390.42 ± 433.90	3387.50 ± 606.89	2552.92 ± 677.31	1959.00 ± 211.00	2903.33 ± 799.77	1928.85 ± 257.78	3229.97 ± 413.14	2463.75 ± 313.87	1879.22 ± 531.42
Monotony (UA)	1.22 ± 0.39	1.05 ± 0.50	1.20 ± 0.62	1.18 ± 0.28	0.79 ± 0.27	1.20 ± 0.34	0.96 ± 0.60	1.27 ± 0.62	1.20 ± 0.32	0.91 ± 0.29
Strain (UA)	3972.83 ± 1999.16	2499.93 ± 1506.03	4067.07 ± 2608.18	3013.85 ± 1381.72	1549.33 ± 577.62	3495.85 ± 1376.71	1851.02 ± 1320.63	4089.52 ± 1730.47	2954.07 ± 1018.37	1702.08 ± 711.26

TV: Total Volume; PATV: Physical Ability Training Volume.

Table 6: Reference intervals, 90% Confidence Interval, Subjects, Mean, Standard Deviations, t-test, Wilcoxon test, p-value and Effect Size (Cohen's d) values of the blood markers of the athlete's pre and post training period.

Analysis	Reference Intervals	90% Confidence Interval		Subjects	Mean	Standard Deviation	t-test/Wilcoxon test*	p value	Effect Size
	2.5th - 97.5th	2.5th	97.5th	(n)	Pre - Post	Pre - Post			*Cohen's d*
Leucocytes ($10^3/mm^3$)	4.5 - 10.1	4.2 - 4.7	9.7 - 10.4	14	6.85 - 6.42	1.70 - 1.19	1.09	0.2916	0.43 (small)
Neutrophils ($10^3/mm^3$)	1.8 - 6.7	1.7 - 2.0	6.5 - 7.0	14	3.99 - 3.59	1.90 - 1.04	0.58	0.5681	0.27 (small)
Lymphocytes ($10^3/mm^3$)	1.2 - 3.3	1.2 - 1.3	3.15 - 3.4	14	2.58 - 2.08	0.39 - 0.42	2.46	0.0284†	1.23 (large)
CK	< 1309	-	-	14	211 - 316	105 - 199	2.04	0.0413†	0.64 (medium)

*Non-parametric test for CK analysis; †Significant differences between the moments ($p \leq 0.05$).

Table 7: Mean, standard deviation and coefficient of intra-individual lymphocytes variation (∆%) between the two moments evaluated. The bold results represent the outliers' values (above of the $RCV_{95\%}$).

Athletes	1st Result	2nd Result	Individual Mean	Standard Deviation	∆%
1	1.93	2.01	1.97	0.06	4.15
2	**3.03**	**1.73**	**2.38**	**0.92**	**-42.9**
3	3.27	2.74	3.01	0.37	-16.2
4	2.62	2.20	2.41	0.30	-16.0
5	2.22	1.93	2.08	0.21	-13.0
6	2.35	1.56	1.96	0.56	-33.4
7	2.04	1.60	1.82	0.31	-21.5
8	2.69	2.11	2.40	0.41	-21.5
9	2.39	1.65	2.02	0.52	-30.9
10	2.97	2.71	2.84	0.18	-8.7
11	2.60	2.29	2.45	0.22	11.9
12	2.80	2.68	2.74	0.08	-4.29
13	2.84	2.27	2.56	0.40	-20.0
14	2.37	1.65	2.01	0.51	-30.38
Mean	**2.58**	**2.08**	-	-	**-19.08**
Standard Deviation	**0.39**	**0.42**	-	-	-

Table 8: Mean, standard deviation and coefficient of intra-individual CK variation (∆%) between the two moments evaluated. The bold results represent the outlier's values (above of the $RCV_{95\%}$).

Athletes	1st Result	2nd Result	Individual Mean	Standard Deviation	∆%
1	**175**	**579**	**377**	**285.6**	**230.8**
2	**346**	**803**	**575**	**323.5**	**132**
3	205	115	160	63.6	-43.9
4	**88**	**272**	**180**	**130.1**	**209**
5	181	216	199	24.7	19.3
6	313	316	315	2.1	0.9
7	172	156	164	11.3	-9.3
8	67	137	102	49.5	104.8
9	447	285	366	114.5	-36.2
10	183	379	281	138.5	107.1
11	129	215	172	60.1	66.6
12	162	223	193	43.1	37.6
13	307	549	428	171.1	78.8
14	175	177	176	1.4	1.1
Mean	**211**	**316**	-	-	**64.2**
Standard Deviation	**105**	**199**	-	-	-

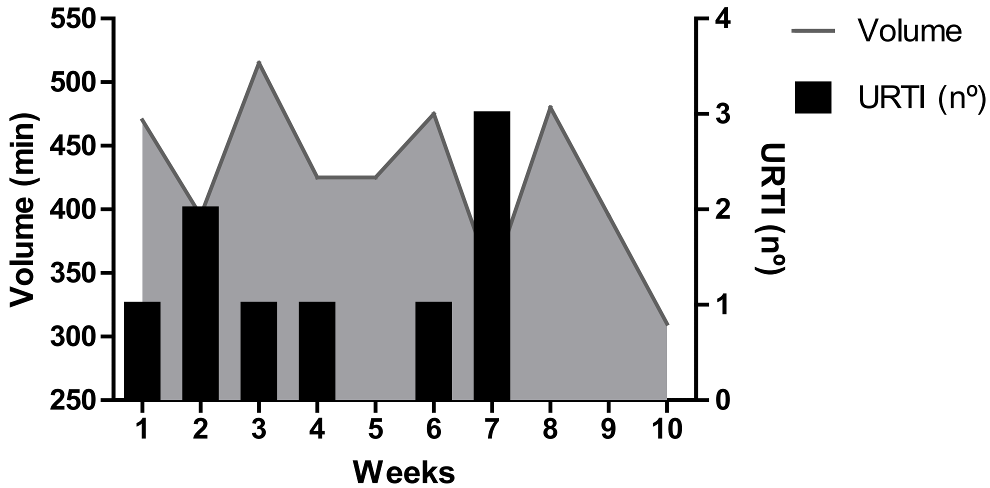

Figure 1: Number of self-related URTIs and training volume over the 10-week pre-season analyzed period.

ers' values (above of the $RCV_{95\%}$ values). To the lymphocytes, accordingly Nunes, et al. [23], the $RCV_{95\%}$ value is ± 40.5%. The CK, in another way, is ± 119%. Only one athlete presented major values than the $RCV_{95\%}$ values for lymphocytes and three athletes for CK.

The lower limb power did not change at the analyzed period of the pre-season, accordingly the results showed at mean ± standard deviation: pre = 53.7 ± 9.6 cm; post = 55.6 ± 9.14 cm (t_{CMJ} = -2.17; p = 0.0548; Cohen's d = -0.2; "small").

Finally, in the Figure 1 there is the informative graphic with the data of number of self-related URTIs and the variation of the volume training over the 10-week pre-season period. In this graphic, we use a visual comparison between the data to find some relationship between the variables. At the seventh week we had three self-related symptoms of URTIs that leaded to a necessary of volume decrease by the coaches.

Discussion

The aim of this study was to evaluate the changes in the CK concentrations, white blood cells subpopulations and lower limb power performance of U19 soccer players as a result of the applied training content and loads during the pre-season period. About the WBC cells the data showed a significantly lymphocytes subpopulation decrease between the two moments but even so within the RI. One athlete presented an $RCV_{95\%}$ major than 40.5% what can demonstrate that this subject could be in risk of URTI. Also, the CK values were within the RI, but three athletes presented the $RCV_{95\%}$ major than 119% and therefore, they could be affected by a muscle injury.

According to Jastrzebski, et al. [24] applied training loads have an important influence on the player's physical capacity. These authors applied in four periods of their season training loads and abilities that predominates the aerobic and aerobic-anaerobic performances finding worsening in the 5-m sprints (1.11 ± 0.03 vs. 1.13 ± 0.05 s; p < 0.04) and increase in the 150-m shuttle run (32.09 ± 0.98 vs. 31.67 ± 0.66 s; p < 0.05) at the end in comparison with the start of the season, demonstrating that the training content can modulate the answer of the physicals abilities of the athletes over the season. This physical abilities adjustment reacts in proportion of the stimulus type, training load and the training content that is the external training load.

Besides that, in the training intensity patterns (Table 2) we verified that the mean RPE corresponded to the "hard" classification according to the Foster scale [4]. This information when related to the total volume during the analyzed pre-season period gave the load training values and this, in turn, was can be able to change the immunological parameters and muscle injury risk. This is according to recent studies that punctuate the pre-season like the most volume and intensity period of the soccer training. For example, Jeong, et al. [25] verified that the major percentuals of maximal heart rate (HR_{max}) between 90-100% (4 ± 3 sessions) and 80-90% (14 ± 4 sessions; p < 0.05) values were found in the pre-season training period when compared with the season period.

On the other hand, it has been reported by the literature that high training loads periods can be falls in immunological variables [13-15,26]. We reported significant decreases in lymphocytes concentrations (2.58 ± 0.39 vs. 2.08 ± 0.42 · $10^3/mm^3$; p < 0.05). Heisterberg, et al. [27] watching the changing in concentrations of different blood markers during the training season of nineteen Danish soccer players (26.3 ± 1.1 years; 183 ± 1 cm) that trained 5-8 times per week, with duration of 1.5-2 hours per session training, verified that the leucocytes concentrations was less at the end of season when compared with the start of eight weeks pre-season (5.3 ± 0.2 vs. 5.8 ± 0.3 · 10^{-9}/L; p < 0.05). In addition, the lymphocytes concentrations were less at the final of pre-season when compared of the start moment of the season (1.84 ± 0.12 vs. 2.08 ± 0.13 10^{-9}/L).

Recently, Horn, et al. [28] in a longitudinal study of ten years with male and female athletes of different sports verified that the meanly changes in the immunity of the athletes, like the leucocytes and subsets, is linked to the predominant energetic metabolism of the sport. Thus, the authors found that modalities like triathlon and cycling were those who had the lowest neutrophils concentrations (< 2 × 10^9/L) in relation to the other sports. In contrast, modalities like canoeing showed the lowest lymphocytes concentrations (< 1 × 10^9/L). The authors classified the sports with metabolism according to a scale like Likert in that exercises physiologists gave their opinions. So, activities like triathlon were classified as aerobics, while team sports like volleyball were classified as anaerobic.

This study shows that when the volume in the individuals sports wherein the endurance ability predominates, there is a trend to falls in the immune system variables. Particularly, although soccer be a team sports, players run distances between 10-12 km during a match, alternating with high intensity activities [16,29]. Thus, it is required the endurance training which can affect the concentration of white blood cells like lymphocytes. Reflects of this drop in immunological variables can be an increase involvement in a URTI, that can remove the athlete for the training routines by some days.

According to our data, there was a predominance of the specific endurance content with 50.61% of the training total time (Table 3). In addition, this ability was predominant in seven of the ten weeks of pre-season period. Frequently high volume training methods are used to increase the endurance ability and, how this variable predominates in the 10-week of our pre-season, it can be relationship with decrease in the lymphocytes concentration found in our study. Also, after six weeks of high training volume, it was necessary decrease it since that the number of self-related URTIs was increased (Figure 1), possibly because the constant high training volume and consequently, high loads. So, for these athletes, although some authors say that there is no evidence of the "J curve" hypothesis in high level athlete, the data seems demonstrate that it was possible change the immunity parameters. This happens due to the fact that our athletes were U19 soccer players and only for olympic athletes the "J curve" don't apply [30].

On the other hand, soccer also is characterized by muscle eccentric actions as jumps, accelerations and unforeseen changes of directions, can leading to an increase in muscle damage and consequently increase the CK blood concentrations [19,27,31]. Besides, for the athlete be able to perform this actions it is important that he train the strength ability. We verified that the strength training was the second that predominates in the training content, with 36.10% of the total time. To increase this ability it was used resistance exercises, run speeds and plyometric training that evolves muscle eccentric and concentric actions, which is capable of increase the muscle damage [32,33], explained by the increase in the CK values. Thorpe and Sunderland [18] found relationships between the number of sprints (> 18 km/h) performed in a soccer match with the change percentage of CK ($r = 0.86$; $p = 0.014$) demonstrating that the greater the high intensity running actions, greater the muscle damage and consequently the CK concentrations. So, for three athletes this training content and loads were capable to increase their CK activity above than the $RCV_{95\%}$, demonstrating that it is necessary to check if the training loads are in accordance with the individual characteristics of the athletes.

Another tool that can be a good performance indicator is a test that represents an intense muscle action like a CMJ. According to Paulsen, et al. [32] a 20% decrease in the performance values with an increase in CK values can be worrying to the athletes, demonstrating a possible structural damage that can lead an injury. However, our data do not demonstrate any alterations in the lower-limbs power performance concomitantly with CK increments. Despite this, there is one athlete that showed significant alterations in $RCV_{95\%}$ lymphocytes and CK, respectively (Table 7 and Table 8 - subject number 2). So, although they did not increase their jump test, these data can suggest us that it was possible which he could be an injury.

So, accordingly with this research we need to pay attention when programming the trainings periods. All because the loads and the training content acting together will modulate the outcomes in the physical abilities and in the immune and neuromuscular systems. So, if the desire is increase the athletes' physical abilities without prejudice in their recuperation capacity, it is important to look at an excessive endurance and strength training contents, once this research demonstrated that this type of content may leads in risk of URTIs and muscle injury without positive effects (increase) in the power performance.

Practical Applications

These findings can contribute to the training organizations providing information about the selection and application of the training loads. From this data, the coaches may know that when the specific endurance predominates in the content training with a "hard" training intensity and the load as found in this study can lead a decrements in the immunological parameters, specifically at the lymphocytes subpopulations, increases in the CK concentrations and without any changes in the lower-limbs power performance.

Therefore, this study shows the possible importance of applied compensation speed and power training stimulus that can increase these abilities in the subjects. The lack of a control group is a limitation of this study. Because this, some results can only be descriptive and a little comparisons can be made. But, more studies with more detailed training content report needs to be done for major understanding of the results of it combinations.

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