COMPARISON BETWEEN DIFFERENT PERIODIZATION MODELS ON MUSCULAR STRENGTH AND THICKNESS IN A MUSCLE GROUP INCREASING SEQUENCE

EXERCISE AND SPORTS SCIENCES

ORIGINAL ARTICLE

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ABSTRACT

Introduction: Studies comparing periodization models in sequences that begin with small muscle group and progressed toward large muscle group in untrained subjects in resistance training are scarce. Objective: The purpose of this study was to compare the effects of ondulatory periodization and linear periodization models on maximum strength and muscular hypertrophy in a muscle group increasing exercise sequence. Methods: Twenty-nine men with no experience in RT were randomly assigned into three groups: ondulatory periodization (OP, n = 10), linear periodization (LP, n = 13), and control group (CG, n = 9). The individuals performed 1RM tests in four exercises: biceps curl (BC), triceps extension (TE), lat pull down (LPD) and bench press (BP) and evaluations of maximum voluntary isometric contraction (MVIC), muscle thickness of elbow flexors (EF) and elbow extensors (EE) before and after the 12 weeks of training were carried out. The OP group varied in volume and intensity on a daily basis, while LP group varied every four weeks. The CG did not perform ST. A two-way ANOVA with repeated measures and the effect size (ES) were used to analyze muscle thickness, 1RM load improvement in each of the four exercises and the MVIC between groups. Results: The major findings of this study were: 1) OP showed major ES for 1RM of BC and TE and for muscle thickness of EF and EE when compared with LP. 3) The ES data did not show significant differences for BP and LPD which finished the training session. Conclusions: We conclude that both periodization models were efficient at improving strength gains and muscular growth. However, ES data show that OP promotes major gains in strength for exercises that are positioned at the beginning of the session and hypertrophy.

Keywords: resistance training, strength testing, muscle hypertrophy, periodization.

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INTRODUCTION

Periodization of physical training refers to the manipulation of the methodological variables of the physical training divided in logical phases and has the aim to perform specific adjustments for physical performance increase and prevention of overtraining[1]. The use of training periodization in resistance training (RT) has become considerably popular in the last years. Currently, elite athletes, bodybuilders and health clubs goers use physical training periodization in RT with the aim to improve performance[2,3]. Concerning the increase of muscular strength, when RT programs with linear periodization are compared (LP, which performs variations in training volume and intensity in monthly periods) and not periodized programs (which do not present variations in intensity, number of series and repetitions and recovery interval between series and exercises) higher strength gain was observed after performance of RT programs with LP, when compared with not periodized programs[1].

Besides the comparison of LP and not periodized programs, other authors[4-7] evaluated new ways of alternate the RT variables with a periodization model which was named ondulatory periodization (OP). The OP hypothesis is that higher alternance frequency of volume and intensity (performed at every session) provides more frequent stimulus alterations, which cause the neuromuscular system to adapt to each training session and avoid stagnation of the muscular strength increase[6]. Some studies which compared the LP and OP effects demonstrated higher strength, muscular power and local muscle resistance gain, in OP programs when compared with LP programs[4,6,8-10]; however, other studies did not demonstrate significant differences between the two systems[5,7,11]. The higher maximal strength gain found in OP programs is attributed to higher manipulation of training volume and intensity, which allows better stimulus/recovery ratio and prevention of overtraining, which may be caused by the linear intensity increase of the training used in LP[1,4,6,8-10,12-15]. The studies which did not find differences between LP and OP attribute the strength gain to the total volume training and suggest that LP is used in training programs which have the aim a physical performance peak[7,11].

Recent studies[5-7] of the LP and OP models indicate the need for further investigation, especially when the previously described models are compared. Additionally, studies comparing periodization models in individuals not trained in RT are scarce, and few of them verified muscular alterations such muscle volume[5,9]. Another important factor is that all the studies which compared periodization models used sessions with exercise sequences following from large to small muscle group[5,7,9] and the exercises order may interfere in the increase of muscle strength[16]. Thus, no previous study has verified the LP and OP influence in a sequence from small to large muscle group; therefore, the aim of the present study was to compare the OP and LP effect on maximal strength and muscle hypertrophy in a performance sequence of exercises from small to large muscle groups. Our hypothesis is that the OP will be more efficient than the LP in improving the analyzed dependent variables.

MATERIALS AND METHODS

Sample

Thirty-two men, students from the Brazilian Marine corps sergeant's course, were randomly distributed in three groups. All of them were physically active and performed calisthenic and aerobic exercises. All individuals reported not to have experience in RT. The fact the individuals were participating in the formation course of the Brazilian Navy sergeants was important so that the individuals had a similar daily routine during the study period. The first group (OP, n = 10) trained using OP with daily overload and number of repetitions maximum variation (RM). The second group (LP, n = 13) used LP starting between 12 and 15 RM on the first training weeks and ending with 3 to 5 RM on the last four weeks. The third group was control (CG, n = 9) and remained performing regular military physical activity program during 12 weeks, but did not perform the RT program. The comparison of the CG results was important to verify whether the military physical training significantly interfered in the maximum strength and muscle hypertrophy.

The inclusion criteria of the study were: a) to be physically active, but not be experienced in RT; b) to be military and student from the formation course in sergeants during the experimental protocol; c) not to perform any kind of regular physical activity during the study period, except for the RT prescribed and the military physical training; d) not to have any functional limitation for the RT or for performance of the 1RM test; e) not to present any medical condition which could influence on the physical training program; f) not to use any nutritional supplementation during the study period.

All the subjects signed a consent form for participation in the research and were informed about tests and training protocol procedures to be performed during the study period. The study was approved by the Ethics Committee of the Clementino Fraga Filho Hospital, from the Federal University of Rio de Janeiro, RJ, Brazil, under protocol number 014/018.

One-repetition maximum test (1RM)

After two weeks of familiarization with the exercises in the RT session, in a total of four sessions, all participants performed two sessions of 1RM test with interval of 48 to 72 hours between sessions. The 1RM tests were performed for all exercises using alternate order. The evaluated exercises were the same used in the training program: biceps curl, (BC), triceps extension (TE), lat pull down (LPD) and bench press (BP). On the first day, the 1RMtests were performed and 48 to 72 hours later, the tests were repeated to determine the loads reproducibility. The highest load obtained in the tests was used for statistical analysis. None exercise was performed between the 1RM tests to avoid interference in the results. The 1RM test protocol was previously described[17].

In order to minimize the errors during the 1RM tests, the following strategies were adopted: a) standard instructions about the test procedures were given to the participants; b) the participants received instructions about the performance technique of the exercises; c) all individuals received verbal encouragement during the tests; d) weight of all free weight, plaques and bars used was verified on a precision scale. The 1RM load was determined through most of five attempts for each exercise, with interval of five minutes between attempts and 10 minutes between exercises. After the 12 weeks of training, the 1RM test was conducted in a similar way to the pre-training moment with the aim to compare it with the loads obtained in the pre-test.

Maximal voluntary isometric contraction (MVIC)

The MVIC test was previously described[18,19]. Maximal load isometric test was used for the elbow flexors (EF) and elbow extensors (EE). For the EF, the individuals remained seated with right elbow flexed at 90 degrees. For the EE, the individual remained at dorsal decubitus with shoulder and right elbow flexed at 90 degrees. After verbal command, the individual performed the MVIC for eight seconds. The maximal load considered was the maximum value obtained in the test, measured in kilograms-strength. The wrist was wrapped with a band attached to a rigid and non-flexible cable, connected to a strength transducer attached to the ground.

Muscular thickness measurements

Muscle thickness (MT) was evaluated with an ultrasound device (US) model EUB-405, with linear transducer and matrix of 512 elements, with excitement frequency of 7.5 MHz, depth resolution of 65 mm and lateral resolution of 80.3 mm. Gel was used for acoustic attachment and to avoid depression on the skin surface. Circumference of right arm (CIR) was measured at 60% of the arm length (L), defined as the distance between the acromion process of the scapula and the lateral epicondyle of the humerus with the individuals standing and wit harms relaxed along the body. The transducer was transversally attached to the segment to measure the thickness of the primary flexors and extensors of the anterior and posterior arm, respectively[20]. MT was considered as the distance between the interfaces of the muscle tissue with the bone and adipose tissue[20,21], calculated with resources of the device, conditioned to the image choice with the best visualization. The MT measurement was taken two consecutive times and the mean was used for data analysis.

Training protocol

After the 1RM loads were obtained in the BC, TE, LPD and BP exercises, the subjects were randomly divided in one of the three groups (OP, LP or CG). Each of the trained groups (OP and LP) was characterized by the variation in training volume and intensity (table 1).

A physical education teacher experienced in RT supervised all the training sessions. The training program frequency was of two weekly sessions with an interval of 72 hours between sessions, in a total of 24 sessions during a period of 12 weeks, all occurring between seven and eight o'clock in the morning. Data analysis was performed only in the individuals who completed the 24 training sessions.

During the training program, the same exercises performed were used, in the following order for both groups: BC, TE, LPD and BP. The periodization models suggested for the study were applied to all exercises. Whenever the individuals performed more repetitions than the expectation for the series of an exercise, the load was increased for that specific exercise. Before each training session, all subjects performed specific warm-up including 20 repetitions with load of 50% of the one used in the first exercise of the session. During the exercises performance, the subjects

Table 1. Dynamics of the training programs of the experimental groups during 12 weeks of training.

Group	Cycle duration	Type of strength	Series x RM	Recovery
OP	Tuesday	Resistance	2 x 12RM – 15RM	1 min
	Friday	Hypertrophy	3 x 8RM – 10RM	2 min
	Tuesday	Maximum strength	4 x 3RM – 5RM	3 min
LP	Weeks 1 – 4	Resistance	2 x 12RM – 15RM	1 min
	Weeks 5 – 8	Hypertrophy	3 x 8RM – 10RM	2 min
	Weeks 9 – 12	Maximal strength	4 x 3RM – 5RM	3 min

OP = ondulatory periodization; LP = linear periodization; RM = repetition maximum; min = minutes.

were verbally encouraged during all series to perform the series until concentric fail. The technique of the movement used during the 1RM tests was defined as standard so that one repetition was successfully performed. The performance velocity of the repetition was not controlled, but the participants were told to keep velocity at which the performance technique was not altered. Engagement to the training program was of 100%. However, only 90.1% participated in all evaluations. Out of these who did not participate, two were from the LP group and two were from the CG.

Statistical treatment

The reproducibility of the 1RM tests was determined by the intraclass correlation coefficient (ICC). The statistical analysis was initially performed by the Shapiro-Wilk normality test and by the Bartlett test to verify the homocedasticity of the groups. All variables presented normal distribution and homocedasticity. Afterwards, 2 x 3 two-way ANOVA with repeated measures (pre-post x groups) (time [pre-training *versus* 12 weeks of training] x groups [OP *versus* LP *versus* CG]) was used to verify whether there was difference between pre and post-training on the 1RM tests, MVIC and MT between groups. Whenever needed, the analyses were performed using the *Fisher's protected post hoc test for least significant differences* (LSD). The calculation of the effect on the 1RM test, MVIC and MT was performed according to the difference between the pre and post-test means, divided by the standard deviation pre-test[22]. The scale proposed by Rhea[23] was applied for classification of magnitude of the effect size of the maximal strength, MVIC and MT. The *t* test was used to analyze diferences between total load recruited (repetitions x load) and the number of total repetitions (series x repetitions) in both training programs. In all cases, significance level adoted was of < 0.05. The data were analyzed in the Statistica software, version 7.0 (Statasoft, Inc., Tulsa, OK).

RESULTS

Table 2 presents the data of the sample description in the pre-training.

Table 2. Pre-training evaluations (mean ± SD).

Groups	OP	LP	CG	P value
Age (years)	30.5 ± 1.7	29.1 ± 2.9	25.9 ± 3.5	0.06
Height (cm)	173.0 ± 6.5	175.9 ± 7.1	171.0 ± 5	0.26
BW (kg)	81.8 ± 15.4	78.4 ± 9.0	73.9 ± 9.9	0.34
% Fat	17.2 ± 6.1	13.6 ± 3.3	15.3 ± 6.9	0.28
1RMRB (kg)	35.1 ± 5.4	32.6 ± 4.9	34.0 ± 4.1	0.68
1RMBP (kg)	32.7 ± 4.4	36.7 ± 6.2	34.7 ± 3.1	0.18
1RMLPD (kg)	82.5 ± 13.0	86.7 ± 9.4	86.6 ± 11.0	0.47
1RMTE (kg)	70.0 ± 16.1	70.3 ± 13.7	71.6 ± 8.9	0.91
MVICEF (kg)	32.7 ± 3.2	33.3 ± 5.5	29.8 ± 4.4	0.19
MVICEE (kg)	21.6 ± 6.4	21.3 ± 4.3	20.3 ± 2.9	0.76
MTEF (mm)	38.4 ± 4.0	36.8 ± 4.8	36.6 ± 4.6	0.58
MTEE (mm)	36.5 ± 2.1	34.4 ± 5.7	32.5 ± 4.8	0.17

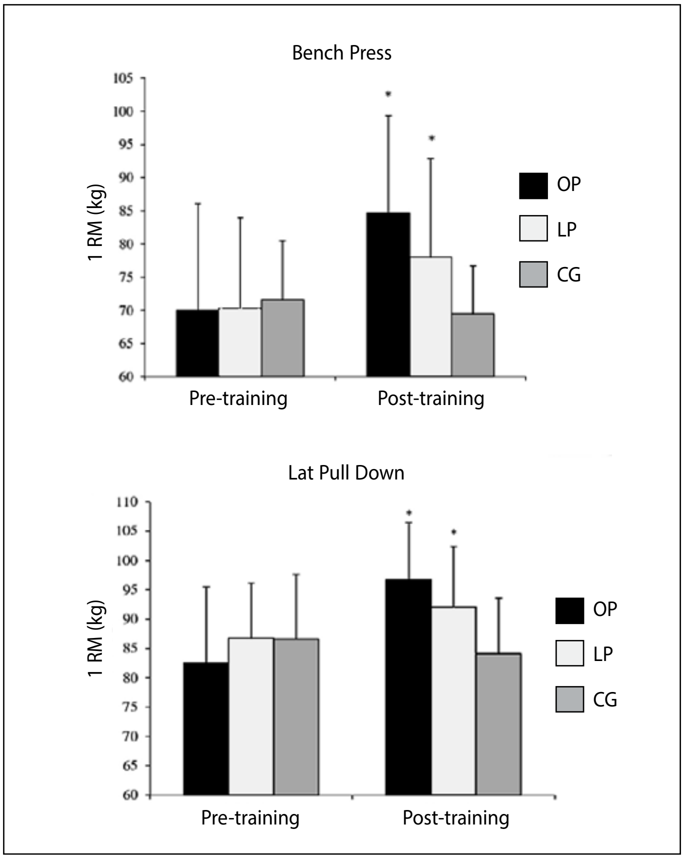
BW=body weight; OP=ondulatory periodization; LP=linear periodization; CG=control group; 1RMBP=1RM test bench press; 1RMLPD=1RM test lat pull down; 1RMTE=1RM test triceps extension; 1RMBC=1RM biceps curl; MVICEF=maximal voluntary isometric contraction elbow flexors; MVICEE=maximal voluntary isometric contraction elbow extensors; MTEF=muscular thickness elbow flexors; MTEE=muscular thickness elbow extensors.

1RM test

The results of the reproducibility of the loads obtained in the 1RM tests demonstrated high ICC for the tests before the OP training (BP r = 0.92, LPD r = 0.90, TE r = 0.97 and BC r = 0.99), LP (BP r = 0.94, LPD r = 0.92, TE r = 0.95 and BC r = 0.95), and CG (BP r = 0.94, LPD r = 0.96, TE r = 0.92 and BC r = 0.93). In the post-training tests high ICC have been also found for the three groups: OP (BP r = 0.96, LPD r = 0.94 TE r = 0.96 and BC r = 0.94), LP (BP r = 0.95, LPD r = 0.94, TE r = 0.93 and BC r = 0.95) and CG (BP r = 0.95, LPD r = 0.95, TE r = 0.94 and BC r = 0.95). After 12 weeks, both training groups presented significant increase concerning the pre-training measurements; however, no significant differences have been found between the training groups. The OP group was higher than the CG in the tests for BC and TE, while the LP group was higher than the CG in the TE test (figures 1 and 2).

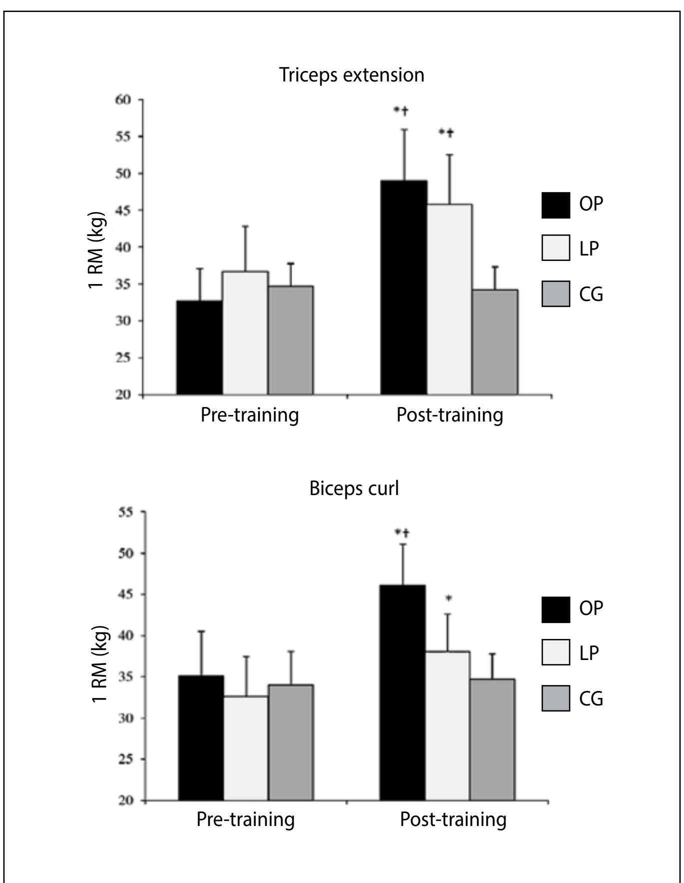
Total repetitions and total recruited load

The total volume of repetitions was similar between the training groups with 2.894 ± 107 and 2.970 ± 111 repetitions for the OP and LP, respectively. No differences have been found for the mean of the total load recruited between OP and LP (369.025 ± 119.611 and 367.820 ± 144.027, respectively).



* = pre and post-training significant difference.

Figure 1. 1RM test (mean and SD) for the BP and LPD pre and psot-training for the ondulatory periodization, linear periodization and control groups.

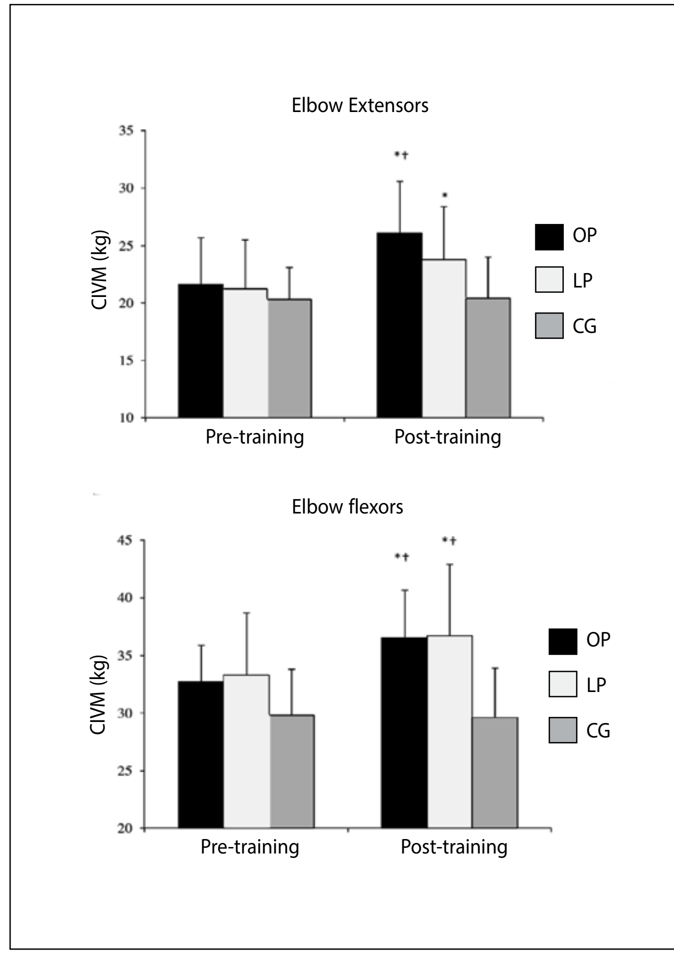


* = significant difference pre and post-training. † = significant difference for the CG.

Figure 2. 1RM test (mean and SD) for the E and BC pre and post-training for the ondulatory periodization, linear periodization and control groups.

Maximal Voluntary Isometric Contraction

After 12 weeks, both training groups obtained significant increase for elbow flexion (EF) an elbow extension (EE). However, no significant differences have been found between the training groups. Both training groups were higher than the CG fo the post-training measurements of EF; however, only OP was higher than the CG for the EE test (figure 3).



* = significant difference pre and post-training. † = significant difference for the CG.

Figure 3. Maximal voluntary isometric contraction (MVIC) for the elbow extensors (EE) and elbow flexors (EF) and (mean and SD) pre and post-training for the ondulatory periodization, linear periodization and control groups.

Muscular thickness (MT)

OP significantly increased the MT of the EF and EE, the LP group significantly increased the MT of EE; however, no significant differences have been found between the training groups (figure 4).

Analysis of the effect size (ES)

In table 3 the ES results after 12 weeks of training for the 1RM test can be seen. The TE and BC exercises demonstrated magnitude "large" when performed in OP (3.70 and 2.04, respectively) and magnitudes "moderate" and "small" when performed in LP (1.47 and 1.12, respectively).

Concerning the MT results, the individuals who performed the OP obtained higher results than the ones obtained by the individuals from the LP group. The OP group obtained magnitude "small" for EE and EF (1.14, 0.80, respectively), since the LP group obtained magnitude "trivial" for EE and EF (0.47 and 0.26, respectively).

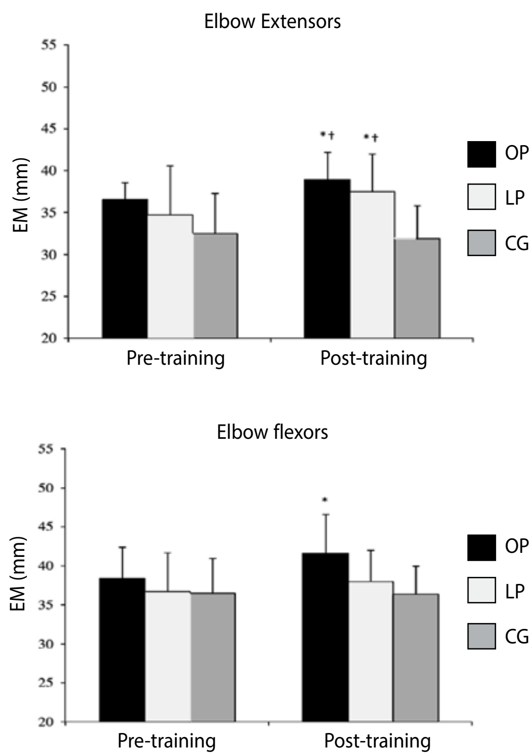
DISCUSSION

The aim of this study ws to evaluate and compare the effect of two periodization models (OP and LP) on the maximum strength and muscle hypertrophy in a RT sequence performed from the smallest to the biggest muscular group. The highest findings of this study were that the OP was more effective in promoting improvement on the maximum dynamic strength in the exercises which started the session and on the muscle hypertrophy of EE and EF. Additionally, both periodization models of the physical training adopted were efficient in promoting increase in maximum strength and military physical training performed by the CG with no addition of RT did not promote alterations in the analyzed parameters.

When the results of the studies in RT are interpreted, we should not observe only the acceptance or rejection of the null hypothesis, but the effect the treatment had on the dependent variable as well, called ES. The calculation of the ES enables us to verify, based on a standard difference of the means, the real effect of the treatment on the independent groups[22]. In order to verify the treatment magnitude through the ES, Rhea[23] suggested an evaluation scale. After the ES was analyzed, the OP model demonstrated higher magnitude in the BC and TE exercises (which started the session) in the 1RM tests, compared with the results obtained by the LP. On the other hand, the LPD and BP exercises (which ended the session) presented similar magnitudes for both training groups, classified as "small".

Our results indicate that the exercises which ended the training session (BP ad LPD), were performed under previous fatigue promoted by the accessory muscles already recruited during the BC and TE performance, for being in the end of the sequence[16,24,25], and consequently thy were less sensitive to the maximum strength increment, which indicates that the B and LPD, besides presenting lower strength increment, have not been influenced by the periodization model adopted either. Thus, higher efficiency of OP is observed in the evolution of maximum dynamic strength in the initial phase of the training for the exercises which started the session and in the muscle hypertrophy of the EE and EF; however, the same results have not been found concerning the MVIC increase. Partially corroborating these results, two studies performed with samples composed of trained men[4,6] verified OP superiority on the maximum strength gain.

In a study similar to ours, Simão *et al.*[9] evaluated untrained individuals divided in three groups: OP, LP and CG, after 12 weeks of RT. The same exercises of the present study were used, but with inverse performance order (BP, LPD, TE, BC). Corroborating the results of the present study, the authors demonstrated OP superiority in the 1RM evolution in the BP and BC. In the present study,



* = significant difference pre and post-training. † = significant difference for the CG.

Figure 4. Measurements of musculr thickness (MT) for the elbow extensors (EE), elbow flexors (EF) and (mean and SD) pre and post-training for the ondulatory periodization, linear periodization and control groups.

Table 3. Effect size (ES) of all post-training variables.

	1RM BC	1RM TE	1RM LPD	1RM BP	MVIC EE	MVIC EF	MT EE	MT EF
Groups	**Post-training**	**Post-training**	**Post-training**	**Post-training**	**Post-training**	**Post-training**	**Post-training**	**Post-training**
OP ES	2.04	3.70	1.08	0.95	1.10	1.18	1.14	0.80
Magnitude	Large	Large	Small	Small	Small	Small	Small	Small
PL TE	1.12	1.47	0.57	0.59	0.60	0.63	0.47	0.26
Magnitude	Small	Moderate	Small	Small	Small	Small	Trivial	Trivial
GC TE	–0.17	–0.16	–0.23	–0.24	0.04	–0.05	–0.02	–0.13
Magnitude	Trivial	Trivial	Trivial	Trivial	Trivial	Trivial	Trivial	Trivial

RM = repetition maximum; BP = bench press; LPD = lat pull down; TE = triceps extension; BC = biceps curl; OP = ondulatory periodization; LP = linear periodization; CG = control group; ES = effect size; MT = muscular thickness; MVIC = maximal voluntary isometric contraction; EE= elbow extensors; EF = elbow flexors.

the OP demonstrated higher magnitude concerning the LP in the BC and TE exercises, which were in the beginning of the sequence. In the study by Simão *et al.*[9], the BP was performed in the beginning of the training session, consequently, it presented sensitivity to the periodization model adopted.; however, the same situation did not occur with the LPD, which was possibly negatively affected for being placed after the BP in the session. Nevertheless, in the study by Simão *et al.*[9], the BC, which was placed in the end of the sequence, presented more remarkable strength increase when performed by the OP. A possible explanation for these divergences of results may be aassociated with the size of the muscular groups involved in each exercise [24].

Previous studies have demonstrated that the exercise order may influence on the development of maximum strength[16,24,25]. Additionally, our results indicate that the RT periodization model does not equally influence on all the exercises in a sequence, since the exercises which start the training session may be boosted by the manipulation of the variables referring to intensity and volume. Dias *et al.*[24] examined the influence of the exercises order in the maximum strength of untrained young men after eight weeks of training. The results demonstrated significant differences in the exercises for small muscle groups (BC and TE) between the different exercises sequences, suggesting that the exercises order may be particularly important during the initial stages of the RT in untrained young men, especially in the exercises for small muscle groups. These results corroborate ours, since the most remarkable increment in maximum strength was observed in the exercises for the small muscle groups, in which the periodization model had more influence. Based on the results by Dias *et al.*[24], a possible explanation for the phenomenon occurred in the present study may be associated with the exercises order, in which the exercises placed in the end of the session presented lower increase in maximum strength magnitudes regardless of the periodization model used.

Concerning the results of the MVIC tests, both training groups demonstrated significant increase in the pre and post-training measurements for the EF and EE and concerning the CG in the EF tests; however, only the OP group was higher in the EE tests, without significant differences between OP and LP. Hartmann *et al.*[13] evaluated sports students and did not find significant differences between the training groups (OP and LP) and the CG for the MVIC performed in the BP exercise. In the present study, the evolution percentage in the MVIC tests was lower than the one found in the 1RM tests for the same muscle groups; however, the OP obtained magnitude "small" for the loads evolution, which demonstrates that the test was sensitive to the adopted training protocol. Nevertheless, both training groups in the study by Hartmann *et al.*[13] presented magnitude "trivial", which evidences that the test was not sensitive to the used protocol. The differences in the results may be aassociated with the strength level of the subjects in the beginning of each study, since in the present study the individuals reported absence of experience in RT; while in the study by Hartmann *et al.*[13], one of the inclusion criteria was to have minimum experience in RT of one year. Thus, it can be concluded that the behavior alterations of the strength-velocity curve in isometric test are not similar in individuals with different strength levels after initial phases of RT[26]

The protocol used in the present study was sufficient to promote adaptations in the MT of the EE, despite not having significant differences between OP and LP. Both training groups presented MT of the EE higher than the CG. However, the same responses were not found for the EF, in which there were no differences between training groups and the CG, with pre and post-training difference only for the OP group. Additionally, the ES data demonstrated that the OP presented higher magnitude in hypertrophy of EE and EF compared with the LP. These results suggest superiority of OP compared with LP in the initial phases of the RT on the muscle hypertrophy gain, indicating that constant alternation of volume and intensity may result in more gain. A possible explanation for MT of EF evolution only in OP may be related to the higher stimulation of the neuromuscular system[6]. The participants of the OP group were submitted to constant variations of recruited overload, number of repetitions performed and different recovery intervals in each session, being less exposed to training load monotony when the intensity and volume are monthly manipulated[4], which may promote stagnation of neuromuscular adaptations[9].

Another important aspect to be analyzed is the sum of the weight recruited and the volume of repetitions performed by each training group during the sessions. No significant differences have been found for volume of repetitions or recruited load. Such fact demonstrates that the method of repetition zones, which uses training by maximum repetitions, did not influence on the volume and work load. Thus, we can state that the differences found between the training groups were due to the periodization model adopted.

The present study has limitations. Among the most important ones we can mention the time of duration of the experiment (12 weeks), which may have not been sufficient to promote optimum responses on the muscle hypertrophy[27]. Additionally, higher weekly frequency, as well as higher exercises volume per muscle group, may have induced to higher responses on the dependent variables analyzed. Future studies should analyze the RT periodization models through longer experimental models and with higher exercises volume per muscle group.

CONCLUSION

The OP and LP models in the RT were efficient in promoting improvement on the variables analyzed during 12 weeks; however, the LP only obtained better responses than the CG in the 1RM test of the TE exercise and MVIC of EF, but did not promote improvement in MT of EF, while in some analyzed parameters, the OP was more efficient when compared with the LP. Additionally, based on the ES data, the OP was more efficient in promoting increase in the 1RM loads of the exercises which started the session and in the MT of the EF and EE. Based on such results, we are able to confirm the initial hypothesis that the OP is more efficient in increasing the strength levels of exercises which start the session and on the muscle hypertrophy in the initial phase of the training when compared with the LP.

All authors have declared there is not any potential conflict of interests concerning this article.

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