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Comparative effect of order based resistance exercises on number of repetitions, rating of perceived exertion and muscle damage biomarkers in men



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ABSTRACT

Objective: The aim of the present study was to compare the effect of orders of resistance exercise, on the number of repetitions performed, rating of perceived exertion and muscle damage biomarkers. *Method:* One week after the 1 repetition maximum (1RM) test, 11 healthy untrained male participants completed two resistance exercise protocols including 4 sets of 4 exercises at 70% 1RM, with 2 min rest intervals between sets, exercises performed until failure and different orders: order A including: hack squat (HS), leg press (LP), leg extension (LE), and leg curl (LC), while order B was opposed to order A (LC, LE, LP, HS).

Results: Increase of CK activity and lactate concentration was the same for order A and order B (P>0.05). The total mean number of repetitions for HS and LC indicated a significant decrease (P<0.05) when they were performed later in each exercise orders; however, no significant difference in the average of total number of exercise repetition was observed for neither resistance exercise orders (order A = 8.59 ± 1.61, order B = 8.78 ± 1.96). Rating of Perceived Exertion (RPE) was not significantly different between the exercise orders; however, the RPE increases for HS and LC when they were the last exercise orders. *Conclusion:* It can be concluded that both of the resistance exercise orders were equally effective in muscle damage parameters (CK, lactate), RPE and the average of the total number of exercise repetitions, although when the exercise session progressed, the number of repetitions performed to volitional failure decreased in last exercise in one single order, and the exercise order can influence performance.

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Efecto comparativo de la secuencia de ejecución de ejercicios de fuerza en el número de repeticiones, el grado de percepción subjetiva del esfuerzo y biomarcadores de daño muscular en hombres

RESUMEN

Objetivo: el propósito del presente estudio fue comparar los efectos de la secuencia de ejercicios de entrenamiento de la fuerza en el número de repeticiones realizadas, la percepción del esfuerzo y en los marcadores de daño muscular.

Método: Una semana despues de la realización de un test de una repetición máxima (1RM), los 11 varones sanos no entrenados participantes, realizaron dos secuencias de ejercicios de fuerza, consistentes en 4 series de 4 ejercicios al 70% de 1RM con dos minutos de reposo entre series, con los ejercicios ejecutados hasta el fallo y distinto orden: la secuencia A consistió: sentadillas hack (HS), prensa de piernas (LP), extensión de piernas (LE), y flexión curl de piernas (LC), mientras que para la secuencia B la secuencia era la opuesta a la A (LC, LE, LP, HS).

Resultados: el incremento de la actividad CK y de la concentración de lactato fue la misma para la secuencia A y la secuencia B (P> 0.05). El total de la media del número de repeticiones para HS y LC mostraron un descenso significativo (P< 0.05) cuando se realizaron posteriormente en cada secuencia de ejercicios; no

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Palavras-chave: Ordem dos exercícios Percepção subjetiva de esforço Dano muscular Número de repetições obstante, no se observó una diferencia significativa, en el recuento del número total de repeticiones, de cada una de las secuencias de ejercicios de fuerza (secuencia A= 8.59 ± 1.61 , secuencia B= 8.78 ± 1.96). el grado de percepción del esfuerzo (RPE) no presentó cambios significativos entre las secuencias de ejercicios; no obstante, la RPE aumenta para HS y LC cuando este fue la última secuencia de ejercicio. *Conclusion:* se puede concluir que ambas secuencias de ejercicios de fuerza fueron igualmente de efectivos en los parámetros de daño muscular (CK, lactate), RPE y el recuento del total del número de repeticiones del ejercicio, aunque cuando la sesión de ejercicio en sólo uno de las secuencias, por lo que la secuencia de ejercicios puede influir en el rendimiento

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Efeito comparativo da sequência de execução dos exercícios resistidos no número de repetições, percepção subjetiva de esforço e biomarcadores de danos musculares em homens

RESUMO

Objetivo: O objetivo do presente estudo foi comparar os efeitos da ordem de exercícios resistidos sobre o número de repetições, percepção subjetiva de esforço e biomarcadores de danos musculares. *Método:* Uma semana após o teste de uma repetição máxima (1 RM), 11 homens saudáveis não treinados completaram duas ordens de exercícios resistidos, incluindo 4 séries de 4 exercícios a 70% de 1RM, com

intervalos de 2 minutos entre as séries com exercícios realizados até a falha e de ordem distintas: ordem A incluiu: agachamento *hack* (HS), leg press (LP), extensão da perna (LE) e flexão de perna (LC), por outro lado, a ordem B ocorreu de forma oposta à ordem A (LC, LE, LP, HS).

Resultados: Ocorreram aumentos da atividade de CK e concentração de lactato tanto para a ordem A e B (P> 0.05). A média do número total de repetições para HS e LC indicou uma diminuição significativa (P <0.05), quando foram realizadas posteriormente em cada ordem de exercícios; no entanto, não houve diferença significativa na média do número total de repetições nos exercícios resistidos (Ordem A = 8.59 ± 1.61 , B = 8.78 ± 1.96). A percepção subjetiva de esforço (RPE) não foi significativamente diferente entre as ordens de exercícios; contudo, a percepção subjetiva de esforço aumentou para HS e LC, quando estes exercícios foram realizados por último.

Conclusão: Ambas as ordens de exercícios resistidos foram igualmente eficientes nos parâmetros de danos musculares (CK, lactato), percepção subjetiva de esforço, assim como na média do número total de repetições, contudo à medida que a sessão de exercício progredia, o número de repetições realizadas até a falha diminui no último exercício em uma única ordem e a ordem dos exercícios pode influenciar no desempenho.

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Introduction

Resistance training, known as strength or weight training is a modality of exercise that has grown in popularity over the past two decades and has been recommended by many major health organizations¹. The key factor of successful resistance training at any level of fitness or age is the appropriate program design¹. American College of Sports Medicine (ACSM)² has indicated the main variables to acute program for resistance training: exercise choice, number of sets, resistance used, rest period length, and the order of exercise. Although most of these variables have been extensively studied over the past 2 decades^{3–5}, exercise order has been studied less frequently in scientifically controlled investigations⁴.

It has been indicated that the exercise order is an important variable which affect both acute responses and chronic adaptations to resistance training programs and this may have a vital impact on the quality of the constituent exercise performed within a training session^{4–7}. Traditional exercise order recommends that large muscle group or multi joint exercises generally is performed before small muscle group or single joint exercises, because this exercise order may result in the greatest long-term strength gains^{1–7}.

Sforzo and Touey⁷ reported that applying small muscle group exercises before large muscle group in trained men, resulted in significantly less total force production (repetitions × resistance) in training session. Speculation based on these findings suggested that, the large muscle groups, should be requested, before the small muscle group, in a training situation because it may be necessary for optimal strength gains⁷.

Few studies^{8–10} have examined the influence of exercise order on the number of repetition and rating of perceived exertion (RPE) when the repetitions are performed until voluntary exhaustion. These studies demonstrated that performing both large and small muscle groups at the end of a training session resulted in significantly fewer repetitions or less strength gains compared to the same time when exercise was performed earlier in a workout sequence. It is usually recommended that the major goal exercises should be placed first in a training session in order to perform these exercises with maximal intensity. However, they did not find any significant difference in the rating of perceived exertion (RPE) between two different exercise orders (large to small, small to large), suggesting that exercise order does not influence the sense of effort at the end of the session^{8–10}. Additionally, to our knowledge, no study has investigated the effect of exercise order with an intensity of 70% 1RM on the number of repetitions and RPE in lower-body muscles in untrained men.

Serum Creatine Kinase (CK) and lactate concentrations have been used as indicators of muscle damage after resistance exercise and may indicate the status of the muscle cell membranes^{11–14}. Previous acute studies^{11–13}, indicated that highly fatiguing resistance exercise protocols, that involve moderate intensity sets, performed with full repetition maximums (i.e., voluntary exhaustion) may induce significant microtrauma to muscle fibers. To our knowledge, the effect of the exercise order with moderate intensity on muscle performance is unclear. Therefore, the purpose of the present study was to compare the effect of acute bouts of resistance exercise order on the number of repetitions, RPE and muscle damage indices in untrained men.

Method

Experimental design

To compare the influence of exercise order on the number of repetitions, RPE, lactate and the CK activity of untrained men, the subjects performed two exercise sessions separated by 72 h of rest using a counterbalanced crossover design. One session (order A) began with exercises that involved large muscle groups and progressed to exercises that involved small muscle groups (i.e. hack squat (HS), leg press (LP), leg extension (LE) and leg curl (LC)); while the other session (order B) utilized the opposite exercise order; LC, LE, LP and HS. All exercises in both orders were performed for 4 sets to voluntarily fatigue by using the predetermined 70% of 1 repetition maximum (1RM) for each of the exercises. A minimum of a 2-minute rest interval, of passive recovery, took place between each set. The number of repetitions was recorded for each set of each exercise for both orders. The RPE measured and recorded after end each exercise order by OMNI scale¹⁹. The CK activity was measured before exercise and 24 and 48 h post exercise. The lactate concentration was assessed pre and immediately post exercise (within 2 min). All evaluations were conducted in the same period of the day (11 am).

Subjects

Eleven healthy and untrained voluntary males who were selected from Guilan University participated in the present study. Inclusion criteria consisted of the following: (a) not to have medical conditions that might be aggravated by participation and (b) not to use nutritional supplements that may enhance performance (i.e., creatine).

All subjects were informed of the risks and benefits of the experiment and signed an informed consent form before participating in the study and were asked not to participate in any resistance exercise other than that prescribed as part of the current study. The Institutional Review Board of the University approved the research protocol.

Anthropometric measurements

Age, weight, height, body mass index, and the body fat percentage were measured and are reported in Table 1. Body fat percentage was assessed using body composition analyzer (InBody3.0, South Korea) according to the manufacturer's protocol.

Table 1

Descriptive characteristics of the subjects (values are Mean \pm standard deviation).

| Variables | $Mean \pm SD$ | | |
|--------------------------------------|------------------|--|--|
| Height (cm) | 175.5 ± 6.41 | | |
| Body weight (kg) | 66.7 ± 8.74 | | |
| Body mass index (kg m ²) | 20.86 ± 1.82 | | |
| Body fat (%) | 11.82 ± 2.66 | | |
| Age (years) | 21.54 ± 2.38 | | |

Strength testing

Two familiarization sessions were designed to habituate subjects with the testing procedures and laboratory environment. The main aim of these sessions was to familiarize subjects with different resistance exercises using weight-training machines. During the familiarization sessions, it was ensured that all subjects performed the correct technique for all exercises before taking part in the main testing trials. After familiarization, the one repetition maximum (1RM) for each exercise was performed on two non-consecutive days for all the exercises using a counterbalanced order¹⁶. The intraclass correlation coefficients was for HS, r=0.94; LP, r=0.92; LE, r = 0.96; LC, r = 0.96. Additionally, paired t-test showed no significant difference between the two occasions when the 1RM tests were performed. To minimize the possibility of injury, short bouts of general and specific warm-up were performed before determination of the 1RM. The general warm up consisted of 5 min low intensity treadmill running and performing lower body flexible movements, while the specific warm-up encompassed 15 repetitions with estimated 40% similar to the actual exercises utilized in the main experiment. After 3 min of rest, each subject had a maximum of 1RM attempts of each exercise with 2-to 5-minute rest intervals between attempts. After the 1RM load in a specific exercise was determined, an interval not shorter than 10 min was allowed before the 1RM determination of the next exercise (9). The standardization of range of motion and movement of the exercises was conducted according to the descriptions of Brown and Weir¹⁷. Each greatest load lifted over the 2-day period was considered the 1RM load and used to calculate 70% resistances for each exercise.

Exercise sessions

Subjects participated in two sessions that were composed of the same exercises performed in two different exercise orders. The subjects were instructed not to engage in any strenuous exercise for the 72 h period preceding the exercise tests and both tests were performed at the same time in the morning on separate days.

Order A began with exercises for large muscle groups and progressed toward exercises for small-muscle groups. The exercise order of order A was hack squat (HS), leg press (LP), leg extension (LE) and leg curl (LC). Order B began with exercises for small-muscle groups and progressed toward exercises for large-muscle groups. The exercise sequence for order B was LC, LE, LP and HS. Warmup before each exercise sequence consisted of 15 repetitions of the first exercise of the session (HS for order A and CL for order B) at 40% of the 1RM load. A 3-minute rest interval was allowed after the warm-up before subjects performed the assigned exercise order. Both exercise orders consisted of 4 sets of each exercise (70% of 1RM load) and recovery times between the different exercise stations were set 2 min. During the exercise sessions, subjects were verbally encouraged to perform all sets to concentric failure. The total number of repetitions for each set of every exercise was determined. Immediately after completion of the fourth set of each exercise and exercise sequences, the OMNI Scale was used to assess the RPE with emphasis on local fatigue^{18,19}.

Collection of blood samples

Venous blood samples (5 cc) were drawn by antecubital venipuncture before the session, immediately after the session (within 2 min) and at the 24 and 48 h after exercise. The blood was immediately centrifuged at 1500 RCF for 10 min at 4 °C, and the plasma was separated and stored in Eppendorf tubes at -70 °C for subsequent use.

Biochemical analysis

Creatine Kinase (CK): plasma level of Creatine Kinase (CK) was measured using an assay kit (Parsazmon, Iran) according to the manufacturer's instruction. Briefly, buffer A and B were mixed with a 1:1 ratio to prepare a volume of $500 \,\mu$ L. Twenty micro liters of the serum samples were added to the mixed buffers, and the absorbance value was measured at 340 nm using an analyzer (Technicon RA-1000-USA) set at 37 °C. The mixture of buffer A and B was used blank for the assay.

Blood lactate: blood lactate concentration was tested from blood samples by enzymatic clirometric method, using the Technicon RA-1000 USA Analyzer with Elitech kit (made in France) according to the manufacturer's instruction.

Statistics

Data were analyzed using SPSS 19.0J (SPSS Japan, Tokyo, Japan) with advanced modules. Kolmogorov-Smirnov test was used to normality of data. Homogeneity of the sample was tested using Levine's test. All variables presented normal distribution and homogeneity. Two-way analysis of variance repeated measures, was used to compare the differences in the mean number of repetitions per exercise, and the repetitions per set between orders and also for the comparison of CK and lactate concentrations, between order A and order B, at different time points. One-way ANOVA was used to evaluate differences on the number of repetitions completed of each set of exercises separately for different exercise order and also for the comparison of CK and lactate in different times of examination. A Tukey post hoc test was performed where indicated. The RPE at the end of each exercise and exercise sequences was analyzed by a Wilcoxon test. P < 0.05 was considered to be statistically significant.

Results

CK levels in order A increased significantly 24 hours post the exercise (P < 0.05). Also, CK levels remained high at 48 hours post exercise (P < 0.05). A similar pattern was observed for CK in order B. There were no significant differences between corresponding CK values of two orders (P > 0.05) (Fig. 1). Differences in lactate concentrations for order A and order B are presented in Fig. 2. Within order A and order B, significant differences in lactate concentrations were demonstrated between pre and immediately post exercise sessions (P < 0.05). When the lactate concentrations were compared between order A and order B, no significant differences were demonstrated at immediately post exercise (P > 0.05). The







Fig. 2. Blood lactate concentrations order A and order B. Values are mean \pm SD.* Significance difference to pre test (*P*<0.05).

Table 2

Number of repetitions per set in both exercise orders (Mean \pm SD).

| | HS | LP | LX | LC |
|------------|--------------------------|-----------------|-----------------|-------------------------|
| Order A | | | | |
| First set | $11.36 \pm 2.69^{*}$ | 9.09 ± 3.33 | 10.09 ± 2.21 | $6.00 \pm 1.34^{*}$ |
| Second set | $10.45 \pm 2.80^\dagger$ | 8.27 ± 2.41 | 10.00 ± 3.34 | $6.45 \pm 2.01^\dagger$ |
| Third set | 10.00 ± 2.48 | $8.0~9\pm2.66$ | 9.09 ± 2.38 | 6.09 ± 2.16 |
| Forth set | $9.36 \pm 2.80^{\P}$ | 7.45 ± 2.38 | 8.36 ± 2.70 | $6.00 \pm 1.34^{\P}$ |
| Order B | | | | |
| First set | 8.09 ± 1.75 | 8.68 ± 3 | 10.81 ± 2.22 | 11.90 ± 3.30 |
| Second set | 7.09 ± 1.86 | 7.54 ± 2.58 | 10.09 ± 2.70 | 11.18 ± 2.44 |
| Third set | 6.45 ± 1.75 | 6.81 ± 2.82 | 9.81 ± 2.99 | 10.81 ± 1.53 |
| Forth set | 6.00 ± 1.67 | 6.72 ± 1.42 | 8.76 ± 2.10 | 10.36 ± 2.16 |

HS, hack squat; LP, leg press; LE, leg extension; LC, leg curl.

* Significant difference when compared to the first set of order B.

[†] Significant difference when compared to the second set of order B.

[§] Significant difference when compared to the third set of order B.

[¶]Significant difference when compared to the fourth set of order B.

comparison of sets between order A and the corresponding order B sets presented differences for 4 sets of HS (P < 0.05) and LC (P < 0.05). The LP and LE presented no significant difference between orders (P > 0.05) (Table 2). In relation to the total average of repetitions developed in each exercise per orders, differences were observed in HS exercise and LC (Fig. 3). There were no differences in the average of total number of exercise repetition for both resistance exercise orders (order $A = 8 \pm 1.61$, order $B = 8.78 \pm 1.96$). With regard to the OMNI scales, no differences were identified between order A



Fig. 3. Mean number of repetitions per exercise in both exercise orders. Values are mean \pm SD. HS, hack squat; LP, leg press; LE, leg extension; LC, leg curl*. Significance difference found when compared to order A.

Table 3

Rating of perceived exertion (RPE) per exercise in both exercise orders (Median).

| | | HS | LP | LE | LC |
|---------|-------------|----|----|----|----|
| Order A | Mean 4 sets | 8 | 9 | 8 | 9 |
| Order B | Mean 4 sets | 9 | 9 | 8 | 8 |

HS, hack squat; LP, leg press; LE, leg extension; LC, leg curl.

(median = 8) and order B (median = 8). Increases in RPE for HS (order A: 8 and order B: 9) and LC (order A: 9 and order B: 8) were observed when those were performed later in the sequences (Table 3).

Discussion

This study examined the effects of different resistance exercise orders on markers of muscle damage (CK, lactate), number of repetitions and rating perceived exertion (RPE) in the untrained men. One of the primary findings of this study was that CK and lactate concentration increase as a result of resistance exercise, regardless of exercise order. According to these findings, Bellezza et al.¹⁵ have reported that there were no significant differences in lactate production in response to resistance exercise order in untrained men. These authors also reported that the average of total number of repetitions was greater from small to large condition¹². Our study showed no significant difference in the average total number of repetitions between large to small (8.24 ± 1.35) and small to large (8.96 ± 1.48) conditions. It appears that the difference in the study of Bellezza et al.¹⁵ is because the subjects did not perform full repetition maximum sets (10RM), on the contrary, in our study, all sets were performed with full repetition maximums until reaching a volitional fatigue for both exercise orders.

Similar to previous studies in which each exercise of the orders was performed to concentric failure in all sets^{5,10}, the present study indicated that exercise order does affect the number of repetitions to volitional fatigue in small muscle group (single-joint) and large muscle group (multi-joint) when they precede exercise for the same general body part. For example, in a multi-joint exercise, the HS total mean number of repetitions decreased (33.6%) in order B when it was preceded by the LC, LE, and LP exercises. Similarly, the total mean number of repetitions performed in a single-joint exercise (LC) decreased (42.3%) in order A when it was preceded by the HS, LP, and LE exercises (Fig. 3). The pattern of a significant decrease in the total mean number of repetitions in four sets indicated that multi-joint and single-joint exercises performance were negatively impacted when performed later in sequence. This decrease in the number of repetition may be as the result of increasing fatigue as the exercise session progresses.

Simao et al.¹⁵ examined the effect of exercise order on the total repetition performance in trained women. The exercise sessions consisted of three sets of each exercise with 80% of 1RM, with 2 min rest intervals between sets and exercises. One of the training sessions began with large muscle group exercise and progressed to exercises that involved small muscle groups (i.e. bench press, shoulder press, triceps extension, LP, LE, LC), while the other session progressed in the opposite order and lower body exercises being performed first in both sessions. Figueiredo et al.¹⁰ compared the effect of exercise order on local muscular endurance (the number of repetitions) in trained women. Two exercise sessions consisted of four sets for each five exercises with 60% 1RM. 2 min rest were applied between sets and exercises. The exercise sequence utilized was bench press, lat pull down, shoulder press, biceps curl and triceps extension in one session, and the exact opposite order in the second exercise sessions (triceps extension, biceps curl, shoulder press, lat pull down and bench press). The results of both studies^{5,10} indicated significantly fewer total repetitions for exercises performed later in the sequence, regardless of whether the exercise involved relatively large or small muscle group. These authors suggested that exercise is considered of primary importance to meet individual needs and movement patterns, and then it should be performed early in training session. Our study differs from the two mentioned studies in which the researcher^{5,10} did not separate the effect of exercise orders on muscle damage indices. They analyzed only the effect of exercise order on the number of repetitions and RPE. However, there are 3 methodological differences between the present study and the above mentioned studies: (a) resistance utilized, (b) the subject population (men vs. women), (c) fitness level (untrained vs. trained). Despite these differences, the studies agree that when exercise for the same body part precedes another exercise of the same body part, performance decreased. Furthermore, our data demonstrated no significant changes for middle exercises of each sequence (i.e. LP, LE) and number of repetitions in the fourth set of each exercise within each order (Table 2).

Recent evidence suggests that OMNI scales provide a conveniently subjective estimate of resistance exercise intensity which can be used reliably to determine RPE^{17–19}. These scales have also been used to evaluate the level of local fatigue or strain and/or discomfort during the resistance exercise session^{17–19}. The 2 min rest interval may emphasize anaerobic glycolysis to a greater extent of the lactate production that is associated with the accumulation of H⁺ that lowers the pH of intracellular fluid¹². The resulting effect is the afferent feedback from muscle chemoreceptors and nociceptors that associates with an increase in the perception of exertion. The central nervous system responds to the increase in RPE by increasing pulmonary ventilation and motor unit recruitment¹⁹. Therefore, we chose to use this scale at the end of each exercise in the orders as a measure of localized muscle fatigue. Our data indicated significant increases in the RPE median after the performance of four sets of HS (order B) and LC (order A) when this exercises were performed later in the orders. These increases are probably to the increased accumulation of H⁺ at the end of training session. In support of the RPE results, the current study also demonstrated a significant increase in blood lactate immediately after exercise session. In addition, significant differences were not found in perceived exertion scale between training orders, an observation which is confirmed by previous studies^{4–10}. The significant difference in RPE scales happens just when a submaximal effort is done at a predetermined percentage of 1RM¹⁷. Therefore, in the present study we can conclude that the lack of significant difference in RPE between exercise orders may be due to the fact that each exercise of the orders was performed in voluntary exhaustion in all sets. Based on our knowledge, the present study is the first one that compares training intensity and muscle damage parameters in response to the two different exercise orders in lower body at untrained men. In the present study the training intensity of 70% 1RM has been utilized, which has not been used at this level in the previous studies^{4–10}. The training intensity used in this study would be deemed by many to be of moderate intensity¹⁵. Therefore, this training intensity can provide health benefits for our sample, which consisted of college-aged men who were physically active and recreationally fit. Future studies are needed to compare the influence of exercise orders with higher-intensity on muscle damage biomarkers and training intensity parameter in lower and upper body exercises, as well as the effect of exercise order on oxidative damage markers. The results of the current study indicate that muscle damage, as indicated by CK and lactate concentrations and RPE was almost the same when resistance exercise orders were performed to voluntary exhaustion in untrained men with 70% 1RM. In addition, this study showed that in both, large and small muscle group exercises, the maximum number of repetitions performed in the last exercise of the order decreases in lower- body exercises.

The present study found that, the evaluation muscle damage biomarkers and RPE, in response to acute resistance exercise in lower-body, was independent of the order of exercise. If the purpose is to induce greater strength and hypertrophy, the exercises or movements most important to the objective of the training session should be performed at the beginning of the session.

Conflict of interest

The authors declare to have no conflict of interest.

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