

## The Effects of Repetitive Running on Middle-Distance Runners' Muscle Damage Level

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### Abstract

The aim of this study was to examine the effect of repetitive running on middle distance athletes' muscle damage. There were 24 female middle-distance athletes who have attended training regularly voluntarily participated in this study. Participants were randomly divided two groups which were experimental (n=12) and control (n=12) groups. Experimental group participants' mean age, height and weight were ( $20.90\pm1.05$  year,  $160.67\pm3.20$  cm,  $57.20\pm3.07$ kg, respectively). Control group participants' mean age, height and weight were ( $21.13\pm.64$  year,  $164.88\pm4.52$ cm,  $58.13\pm3.48$ kg, respectively). Participants' CK, LDH, Urea, Uric Acid, Creatine, HDL, LDL, Triglyceride, Total Cholesterol, Glucose, ALT and AST values were collected before and after 8 weeks repetitive running training. Data were analyzed with SPSS 24.0 package programme. As a result, it showed that LDH, uric acid and creatinine values increased significantly in both groups post-test values. There was no statistically significant difference in pre-post test, CK, Urea, ALT, AST, Glucose, HDL, LDL, Triglyceride and Total Cholesterol values Repetitive running training positively increased LDH levels, while positively decreasing creatine and uric acid levels. As a conclusion, 8 weeks repetitive running training positively affected middle distance athletes' LDH, creatine and urea levels. Applied training program did not affect other values which are used for determining muscle damage. **Keywords:** Repetitive running, Middle distance runners, Muscle damage.

#### Orta Mesafe Koşucularında Tekrarlı Koşuların Kas Hasarına Etkisi

### Öz

Bu çalışmanın amacı, tekrarlı koşuların orta mesafe sporcularının kas hasarı üzerindeki etkisini incelemektir. Bu çalışmaya düzenli olarak gönüllü olarak antrenmanlara katılan 24 kadın orta mesafe sporcusu katılmıştır. Katılımcılar rastgele olarak deney (n=12) ve kontrol (n=12) grupları olmak üzere iki gruba ayrıldı. Deney grubu katılımcılarının yaş, boy ve kilo ortalamaları (sırasıyla 20.90±1.05 yıl, 160.67±3.20 cm, 57.20±3.07kg) idi. Kontrol grubu katılımcılarının yaş, boy ve kilo ortalamaları (sırasıyla 21.13±.64 yıl, 164.88±4.52cm, 58.13±3.48kg) idi. Sporcuların kas hasarını belirlemek için iki grup ön ve son test deney tasarımı kullanılmıştır. Katılımcıların CK, LDH, Üre, Ürik Asit, Kreatin, HDL, LDL, Trigliserit, Toplam Kolesterol, Glikoz, ALT ve AST değerleri 8 haftalık tekrarlı koşu antrenmanı öncesi ve sonrasında toplandı. Veriler SPSS 24.0 paket programı ile analiz edilmiştir. Sonuç olarak, LDH, ürik asit ve kreatin değerlerinin her iki grupta da son test değerlerinin önemli ölçüde arttığını gösterdi. Ön-son test, CK, Üre, ALT, AST, Glikoz, HDL, LDL, Trigliserit ve Toplam Kolesterol değerlerinde ise istatistiksel açıdan anlamlı fark yoktu. Tekrarlayan koşu antrenmanları, LDH düzeylerini olumlu yönde artırırken, kreatin ve ürik asit düzeylerini olumlu yönde azalttı. Sonuç olarak, 8 haftalık tekrarlı koşu antrenmanı orta mesafe sporcularının LDH, kreatin ve üre düzeylerini olumlu yönde etkilemiştir. Uygulanan antrenman programı, kas hasarını belirlemede kullanılan diğer değerleri etkilemedi.

Anahtar Kelimeler: Tekrarlı koşu, Orta mesafe koşucuları, Kas hasarı.

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# INTRODUCTION

Short distance sprints mainly require speed whereas middle distance running relies on a combination of speed and endurance in athletics (Dahl et al., 2020). Physiological profile, including aerobic capacity and anaerobic ability, is essential for success and distinguishes middle-distance runners from long-distance runners and sprinters (Reilly et al., 2005). Performance depends not only on metabolism, but also on the amount of oxygen consumed during running at a given speed. Studies showed that mid-distance performance is more dependent on aerobic capacity than on anaerobic capacity (Weyand et al., 1994). Repetitive runs consist of runs with minimum recovery time and short-term rest, and are reported to be a determinant of the aerobic fitness level of athletes (Wong et al., 2012).

Muscle damage is defined as metabolic and chemical events that occur in the muscles after intense exercise (Clarkson & Hubal, 2002). Loss of function in the muscles, decrease in calcium values, weakness and pain in the muscles as a result of intense exercises are the most prominent features that indicate the presence of muscle damage (Tiidus, 2008). High-intensity exercise causes the breakdown of the muscle membrane and entrance of certain muscle enzymes into the bloodstream. As a result, an increase in intracellular protein values and in blood plasma levels are observed (Bailey et al., 2010). Production of free radicals in the body is affected by energy requirements during exercise, oxygen consumption, and differences in mechanical loads on soft tissue (Park, 2006). To determine exercise-induced muscle damage, pre- and post-exercise blood serum levels of enzymes in muscle metabolism are examined (Moflehi et al., 2012). Creatine Kinase (CK), Lactate Dehydrogenase (LDH) and Lactic Acid (LK) are the most commonly used enzymes in order to determine the presence of muscle damage in skeletal muscle (Nie et al., 2011). In addition, intense and vigorous exercises cause changes in liver and kidney functions (Tiidus, 2008). Studies showed that urea, uric acid and creatinine concentration are the markers in determining fatigue (Morris et al., 2000).

Middle-distance runners' muscle damage and lipid metabolism after running (800 and 1500 m) are directly dependent on running distance (Lippi et al., 2019). However, this seems to be a notable disadvantage. When the literature is searched in this respect, there are few studies examining the effects of repetitive runs on middle-distance runners' muscle damage level. The purpose of this study was to examine the effects of repetitive run workouts on middle-distance runners' muscle damage level.

### METHODS

### Participant

There were 24 female athletes attending in national level competitions voluntarily accepted to participate in the study. Participants were randomly divided into two groups as control and experimental. Mean age, height and weight of the control group were  $21.13\pm0.64$  years;  $164.88\pm4.52$  cm;  $58.13\pm3.48$  kg, respectively. Experimental group participants' mean age, height and weight were  $20.90\pm1.05$  years;  $160.67\pm3.20$  cm;  $57.20\pm3.07$  kg, respectively. Participants in this study were physically healthy, had no anterior cruciate ligament, meniscus, ankle injury and orthopedic problems. At the beginning of the study, participants were

informed about the benefits and risks of working out. During the work out process, participants not attending more than two sessions or having chronic illness were excluded from the study. It was controlled and ensured that participants did not use any drugs or ergogenic aids that would affect their performance.

### Measurement

Body weight and height of the athletes were measured with a stadiometer (SECA-Mod. 220, Seca GmbH & Co. KG., Hamburg, Germany). Participants were asked not to eat 2 hours before the measurements, not to consume caffeine until 24 hours before, not to use any medication, and to avoid vigorous exercise. Blood samples were taken into shaped tubes by a specialist physician at the beginning and end of the study. The blood samples which were taken from participants were inverted 3-5 times to prevent hemolysis and after waiting 20 minutes, they were centrifuged at 3500 rpm for 5 minutes. The samples were stored in the -80° cold chain until the analysis. Blood samples were analyzed in the Biochemistry Laboratory of Karamanoğlu Mehmetbey University Training and Research Hospital. To analyze blood parameters, which were ALT (MAK052), AST (ERMAD457IFCC-1VL), LDL (LP2-2MG), HDL (L8039-10MG), Triglyceride (TR0100-1KT), Total Cholesterol (C8667-500MG), Blood glucose (MAK181), Creatin Kinase (10127566001), Lactate (Lactate Sguat Strips) and Urea (MAK006), Sigma Aldrich kits on a Cobas 8000 analyzer (Modular Systems, Roche Diagnostic, Germany) were used to measure.

### **Training Program**

Participants' performed warm-up and dynamic stretching exercises for 5 minutes before proceeding to the main phase of the workout. They completed the training by performing a static stretching exercise for 5 minutes during the cool-down phase. No ergogenic supplement was given to the athletes during the workout and they were provided to consume water for dehydration (Workout program Table 1). During the workout program, the target heart rate of the participants was aimed to be between 50-70%. The workout program was applied for 8 weeks and 3 days a week. The workout time was between 10:00-12:00 a.m. In each workout session, repetitive running exercise was performed for 25-60 minutes. Participants' target heart rates were determined by calculating the heart rate reserve according to the Karvonen method. The control group was informed not to do any chronic exercise during the study and they were provided to follow the schedule.

	Workout Duration	Workout Intensity	Workout Frequency
Week 1	25 min.	50 %	3 days/week
Week 2	30 min.	50 %	3 days/week
Week 3	35 min.	60 %	3 days/week
Week 4	40 min.	60 %	3 days/week
Week 5	45 min.	60 %	3 days/week
Week 6	50 min.	70 %	3 days/week
Week 7	55 min.	70 %	3 days/week
Week 8	60 min.	70 %	3 days/week

Table 1. The Training programme of the Study

### **Research Publication Ethics**

Ethical approval for the research was obtained from the Ethics Committee of the of Karamanoğlu Mehmetbey University, Faculty of Medicine on 20.01.2021 with the decision number 2021/5.

### Statistical analysis

In the analysis of the data, firstly, descriptive results (means and standard deviations) of muscle damage, liver and kidney functions (Triglyceride, Cholesterol, HDL, LDL, Glucose, ALT, AST, CK, Uric Acid, Urea and Creatine were presented. Second, since this study was preposttest experimental design and the existence of the difference between the post-test values of the control group and the experimental group was important, a Mixed ANOVA analyze was used. Before the statistical calculations, assumptions of Mixed ANOVA which were homogeneity of variance and normality were checked. Homogeneity of variance was checked by levene's test value which should be non-significant. Results showed that assumption of homogeneity of variance was not violated ( $p \ge 0.05$ ). Normality was checked by skewness-kurtosis values and histogram. Results of normality indicated that collected data had normal distribution. Hence, assumptions ensured that Mixed ANOVA could be used for statistical calculation of the collected data. Cohen's d calculation was used to report the effect sizes of group differences. The effect size was decided based on the following criteria: d=0.2 (small effect), d=0.5 (medium effect), and d=0.8 (large effect).

### RESULTS

In this study, we examined the effects of repetitive run workouts on middle-distance runners' muscle damage levels. Pre and posttest results (Triglyceride, Cholesterol, HDL, LDL, Glucose, ALT, AST, CK, Uric acid, Urea and Creatine) were calculated and presented in tables.

Laboratory (DD) _	Gre	Б	р	d	
Laboratory (KK) -	Control Experimental				- r
CK (<145 U/L)	190.88±40.52	211.22±100.83	0.86	0.37	0.26
Uric Acid (3.5 – 7.2 mg/dl)	5.70±0.78	4.24±.65*	6.72	0.02	2.03
Urea (7.9 – 21 mg/dL)	29.38±7.67	29.33±5.48	1.56	0.23	0.01
Creatine (0.66 – 1.09 mg/dl)	1.08±0.10	0.78±0.12*	47.77	0.00	2.72
Lactate (m/mol)	8.14±1.59*	6.35±2.31*	5.49	0.01	1.88

Table 2. Between Group Lactate, Uric Acid, CK, Urea and Creatine Values of the Participants

\*: p<0.05

According to Table 2, a statistically significant difference was found between lactate post-test and pre-test values ( $F_{(1,15)}=1.46$ , d=0.25,  $p\leq0.05$ ). Uric acid findings showed a significant difference between the post-test and pre-test values of the participants in both groups ( $F_{(1.15)}=18.63$ , d=3.04,  $p\leq0.05$ ). A statistically significant difference was found between pre and post-test Creatine findings ( $F_{(1.15)}=14.31$ , d=2.28,  $p\leq0.05$ ). There were no significant difference between pre and post-test findings of CK ( $F_{(1.15)}=3.48$ , d=0.50,  $p\leq0.05$ ) and Urea ( $F_{(1.15)}=1.46$ , d=0.25,  $p\geq0.05$ ).

Laboratory(DD)	Test	Group		Б	n	d
Laborator $y(\mathbf{K}\mathbf{K})$	Test -	Control	Experimental	— г	р	u
СК	Pre Test	148.50±32.12	175.44±69.61	2 49	0.04	0.50
(<145 U/L)	Post Test	$190.88 {\pm} 40.52$	211.22±100.83	- 3.40		
Uric Acid	Pre Test	$3.75 \pm 0.46$	4.27±0.41	19 62	0.00	3.04
(3.5 – 7.2 mg/dl)	Post Test	5.70±0.78*	4.24±0.65	- 18.05		5.04
Urea	Pre Test	23.88±6.03	29.00±6.69	1 46	0.25	0.80
(7.9 - 21  mg/dL)	Post Test	29.38±7.67	29.33±5.48	- 1.40		
Creatine	Pre Test	$0.84{\pm}0.11$	$0.72 \pm 0.07$	- 14.31	0.00	2.28
(0.66 – 1.09 mg/dl)	Post Test	$1.08 \pm 0.10*$	0.78±0.12			
Lactate	Pre Test	$2.08 \pm 0.89$	2.56±0.69	- 1.16	0.02	2.22
(m/mol)	Post Test	8.14±1.59*	6.35±2.31*			2.22

\*p<0.05

Table 3 showed that there was a significant difference between the uric acid post-test values  $(F_{(1.15)}=6.72, p\le 0.05)$ . Follow-up test indicated that the uric acid values of the control group were significantly higher than the experimental group. Effect size was determined as large effect (d=2.03). When the creatine post-test values were examined, a statistically significant difference was found between the experimental and control groups  $(F_{(1.15)}=47.77, p\ge 0.05)$ . According to the findings, the creatine value of the control group was significantly higher than the experimental group. Effects size was calculated as 2.72 and this was a large effect. A significant difference was determined between the two groups according to the lactate post-test values  $(F_{(1.15)}=5.49, p\ge 0.05)$ . Follow-up test demonstrated that experimental group participants had significantly lower lactate value than those in control group. Effect size was calculated as large effect with d= 1.88. Results of CK  $(F_{(1.15)}=0.86, d=0.26, p\ge 0.05)$  and Urea  $(F_{(1.15)}=1.56, d=0.23, p\ge 0.05)$  showed that there were no significant post-test difference between two groups.

**Table 4.** Between group Triglyceride, Cholesterol, HDL, LDL, Glucose, ALT and AST values of the participants

	Gro	Б		d	
Laboratory(KK)	Control Experimental		F		р
Triglyceride (35-150 mg/dl)	$69.38\pm26.60$	$52.89\pm23.04$	0.94	0.35	0.15
Cholesterol (120-200 mg/dL)	$138.50 \pm 28.48$	$148.67\pm21.59$	0.04	0.85	0.11
HDL (<160 mg/dL)	$47.00\pm4.69$	$60.56\pm17.14$	4.22	0.06	0.79
LDL (<130 mg/dl)	$77.63\pm27.45$	$77.44 \pm 18.06$	0.25	0.63	0.17
Glucose (75-106 mg/dL)	$86.25 \pm 13.21$	$79.56\pm14.05$	1.09	0.31	0.49
ALT (7-52 U/L)	26.13 ± 13.68	14.11 ± 2.52	0.60	0.45	1.22
AST (12-39 U/L)	31.25 ± 15.91	21.11 ± 5.84	0.59	0.45	1.11

Table 4 indicated that there were no significant difference between pre and post-test findings of Triglyceride  $F_{(1.15)}=2.02$ , d=0.23,  $p\geq0.05$ ), Cholesterol ( $F_{(1.15)}=3.25$ , d=0.40,  $p\geq0.05$ ), HDL ( $F_{(1.15)}=1.77$ , d=0.19,  $p\geq0.05$ ), LDL ( $F_{(1.15)}=0.02$ , d=0.12,  $p\geq0.05$ ), Glucose ( $F_{(1.15)}=2.65$ , d=0.56,  $p\geq0.05$ ), ALT ( $F_{(1.15)}=2.04$ , d=0.52,  $p\geq0.05$ ) and AST ( $F_{(1.15)}=3.04$ , d=0.84,  $p\geq0.05$ ) values.

**Table 5.** Within group Triglyceride, Cholesterol, HDL, LDL, Glucose, ALT and AST values of the participants

Laboratory (DD)	Group			F	n	d
	Test —	Control	Experimental	Г	þ	u
Triglyceride	Pre-Test	77.00±39.37	71.78 ±29.62	- 2.02	0.18	0.23
(35-150 mg/dl)	Post-Test	69.38±26.60	52.89 ±23.04			
Cholesterol	Pre-Test	158.50±31.33	152.11±8.34		0.09	0.40
(120-200 mg/dL)	Post-Test	138.50±28.48	148.67±21.59	- 3.25		
HDL	Pre-Test	57.25±19.38	63.11±9.02	- 1.7	0.20	0.19
(<160 mg/dL)	Post-Test	47.00±4.69	60.56±17.14			
LDL	Pre-Test	81.38±13.70	75.33±8.89	- 0.02	0.90	0.12
(<130 mg/dl)	Post-Test	77.63±27.45	77.44±18.06			
Glucose	Pre-Test	90.75±11.78	86.89±12.04	- 2.65	0.12	0.56
(75-106 mg/dL)	Post-Test	86.25±13.21	79.56±14.05			
ALT	Pre-Test	12.00±3.25	19.67±9.39	- 2.04	0.17	0.52
(7-52 U/L)	Post-Test	26.13±13.68	14.11±2.52			
AST	Pre-Test	17.75±2.60	23.33±6.58	- 3.04	0.10	0.94
(12-39 U/L)	Post-Test	31.25±15.91	21.11±5.84			0.84

According to table 5, indicated that there were no significant difference between pre and posttest findings of triglyceride ( $F_{(1.15)}=0.94$ , d=0.15, p $\ge 0.05$ ), Cholesterol ( $F_{(1.15)}=0.04$ , d=0.11, p $\ge 0.05$ ), HDL ( $F_{(1.15)}=4.22$ , d= 0.79, p $\ge 0.05$ ), LDL ( $F_{(1.15)}=0.25$ , d=0.17, p $\ge 0.05$ ), Glucose ( $F_{(1.15)}=1.09$ , d=0.49, p $\ge 0.05$ ), ALT ( $F_{(1.15)}=0.60$ , d= 1.22, p $\ge 0.05$ ) and AST ( $F_{(1.15)}=0.59$ , d=1.11, p $\ge 0.05$ ) values.

### DISCUSSION

The most important finding of this study was that repeated running workout for eight weeks affected a wide range of biochemical variables, therefore, it did not cause serious muscle damage in athletes and positively changed their kidney and liver functionsIt was determined that there was a statistically significant difference between lactate, Uric acid, Creatine and CK pretest and posttest values to determine muscle damage,  $[(F(1.15)=1.46, d=0.25, p\leq) 0.05)$ ,  $(F(1.15))=18.63, d=3.04, p\leq0.05), (F(1.15)=14.31, d=2.28, p\leq0.05), (F(1.15)=3.48, d=0.50, p \leq0.05)]$  respectively. However, there was no difference between the pre-test and post-test values for Urea ( $F_{(1.15)}=1.46, d=0.25, p\geq0.05$ ). It is known that workout at different loads has an effect on muscle damage in middle-distance running, and eccentric contraction during exercise causes more muscle damage than other types of contractions. Moreover, maximum oxygen uptake, stride length, power, peak speed, VO<sub>2</sub> Max, thigh length and aerobic capacity are important determinants of performance in running (Dahl et al., 2020). Ensuring the continuity of both speed and power concepts in middle-distance runners turns out as muscle power, loss of function and muscular fatigue (Clarkson & Hubal, 2002).

Literature indicated that creatine kinase levels, which were evaluated to determine the biochemical responses that occurred with changes in training load, were associated with muscle damage caused by specific workout (Freitas et al., 2014). Serum CK level may rise from damage to muscle tissue as a result of intense and prolonged workout (Brancaccio et al., 2007). The highest serum CK activities have been detected after very long duration competitions such as marathon running or triathlon. It was determined that maximal and sub-maximal trainings applied to middle-long distance athletes and sedentary individuals significantly increased the serum CK value in athletes, not for sedentary individuals (Mohamed et al., 2016). In another study, it was stated that pre-competition serum CK levels in master athletes were higher when the marathon was completed and 24 hours later (Kratz et al., 2002). In addition, they determined that eight-week aerobic training applied to male middle-distance runners determined a significant increase in serum CK values (Proira et al., 2015). Studies indicated that there was a breakpoint in serum CK release of 300-500 IU/l after exercise and enzyme levels were correlated with individual muscle characteristics (Totsuka et al., 2002). Furthermore, there are significant gender-related differences in resting serum CK levels and have lower values in women than in men (Fu et al., 2002). Daily workout can induce persistent serum CK increase, and CK levels are higher in athletes at rest and after exercise. In our study, it was determined that there was an increase in the serum CK level after the workout ( $p \le 0.05$ ) and the findings were in parallel with the literature. On the other hand, results showed that the repetitive running training had an effect on the increase in the serum CK level (ES, d=0.50). Studies on the detection and evaluation of kidney functions (lactate, uric acid, urea and creatine) due to muscle damage in middle-distance runners have been investigated in literature (Brancaccio et al., 2007; Siahpoosh & Nesaei, 2016). Dehydration is well known to be associated with acute renal dysfunction, however there is no long-term effects on kidney function (Roncal-Jimenez et al., 2015). Studies showed that chronic kidney disease, especially in athletes, is due to dehydration, heat stress and increased workout load (Heung et al., 2016; Hsu & Hsu, 2016). Endurance athletes (Middle distance runners) are exposed to similar conditions (dehydration, heat stress and increased workout load). As a result, higher eccentric muscle contractions lead to a higher metabolic workload. Rojas-Valverde et al. (2019) reported that serum creatine and bun values in athletes increased acutely during workouts applied on

different loads. In different studies in literature, it was determined that marathon running increased the serum creatine value for kidney function (Boulter et al., 2011). According to findings, kidney function has been determined as an important factor in endurance athletes performing long-term activities at moderate and high intensity workouts (Rojas-Valverde et al., 2019).

To determine liver functions in our study, no statistically significant difference was found between the pre-test and post-test results of Triglyceride, Cholesterol, HDL, LDL, Glucose, ALT and AST  $F_{(1.15)}=2.02$ , d=0.23,  $p\geq0.05$ ) ( $F_{(1.15)}=3.25$ , d=0.40,  $p\geq0.05$ ) ( $F_{(1.15)}=1.77$ , d=0.19,  $p\geq0.05$ ) (F<sub>(1,15)</sub>=0.02, d=.12, p>0.05) (F<sub>(1,15)</sub>= 2.65, d=0.56,  $p\geq0.05$ ), (F<sub>(1,15)</sub>=2.04, d=0.52,  $p\geq0.05$ ) and (F<sub>(1.15)</sub>=3.04, d=0.84,  $p\geq0.05$ ), respectively. It is known that liver aminotransferases are associated with high triglycerides and low cholesterol in the general population, while AST and ALT are associated with insulin resistance and liver fat accumulation in muscle and liver (Clark et al., 2003). Studies in literature have showed a clear relationship between ALT and AST and body weight (Westerbacka et al., 2004). ALT is mainly found in the liver, and AST is substantially found in muscle in addition to the liver, and its amount may increase in case of muscle damage during exercise (Nie et al., 2011). The increase in liver function tests had a greater effect of muscle damage than liver damage (Tirabassi et al., 2018). Arakawa et al. (2016) stated that ultra-marathoners' both AST and ALT values increased on the second day after the race, AST values were prolonged until the fifth day, but ALT values remained in the normal range. This is the reason of muscle damage. Hence, it is difficult to directly determine the role of exercise on liver condition from only circulating AST and ALT levels (Karstoft et al., 2013).

As a conclusion, this study indicated that eight-week repetitive running workload had positive effects on muscle damage, liver and kidney functions. Overall, it was found that the workload program had an effect on muscle damage, liver and kidney function, with small increases in the indicating parameters, but this had non-significant side effects. It was determined that endurance training did not pose any significant health risks at the biochemical level, and low-level health risks might occur when multi-phase workout or competition was performed. Considering individual differences, performance changes can be evaluated by observing the level and frequency of loading on middle distance runners.

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**Researchers' Contribution Statement:** Idea/Concept: RS; Design: RS; Control/Supervision: RS; Data Collection and/or Processing: MD; Analysis and/or Interpretation: MD; Literature Review: MD; Writing the Article: RS, MD; References and Fundings: MD; Materials: MD, RS.

### **Ethics Committee**

Name of Board: Karamanoğlu Mehmetbey University of Medicine Faculty Ethics Committee Date: 20.01.2021 Issue No: 01-2021/5

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