

## A randomized controlled trial: effectiveness of 10-week Nordic Hamstring exercise training and subsequent detraining in healthy young men

*Randomize kontrollü bir çalışma: sağlıklı genç erkeklerde 10 haftalık Nordic Hamstring egzersiz eğitimi ve onu izleyen egzersizi bırakma süreçlerinin etkileri*

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### Abstract

**Purpose:** Nordic Hamstring Exercise (NHE) is used to increase hamstring muscle strength, prevent injuries. We aimed to reveal the acute, long-term effects of NHE, followed by detraining on demographic measurements, flexibility, anaerobic power, damage, fatigue, oxidative stress, blood lactate levels.

**Material and method:** A total of 40 sedentary healthy male participated the experiments. 20 of them underwent 10 weeks of progressive NHE followed by detraining. Muscle architecture was determined by ultrasonography, strain ratio by elastography. Anaerobic power was assessed by standing long jump, vertical jump, flexibility by sit reach tests. Creatine kinase activity, oxidant/antioxidant parameters were measured from venous blood by commercial kits.

**Results:** NHE allowed subjects to lose weight, which was reversed by detraining of 5 weeks. Exercise caused an increase in knee angles that wasn't affected by detraining. 10-week NHE caused a partially reversed increase in anaerobic performance upon detraining. NHE resulted in increment of biceps femoris long head area, pennation angle which were reversed by detraining of 10-weeks. Blood lactate, muscle pain, fatigue were increased after each exercise session. NHE didn't change oxidative stress but, 5-week detraining resulted in an increase in total oxidant capacity, oxidative stress index. Detraining of 10 weeks caused a reduction of these parameters.

**Conclusions:** It has been observed that most of the gains obtained with 10-week progressive NHE are reversed with 5-week detraining. These results may guide the selection of the exercise type to increase performance and muscle strength.

**Key words:** Nordic Hamstring Exercise (NHE), detraining, anaerobic power, oxidative stress, creatine kinase.

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### Öz

**Amaç:** Nordic Hamstring Egzersizi (NHE) hamstring kas kuvvetini arttırmak ve yaralanmaları önlemek amacıyla kullanılmaktadır. NHE'nin akut, uzun süreli ve egzersizi bırakma (detraining) süreçlerinde demografik veriler, esneklik, anaerobik güç, kas hasarı, yorgunluk, oksidatif stres, kan laktat seviyeleri üzerindeki etkilerinin ortaya konması amaçlanmıştır.

**Gereç ve yöntem:** 40 sağlıklı, sedanter erkek gönüllü birey deneylere katılmıştır. 20 deneğe 10 haftalık ilerleyici NHE ve takiben detraining uygulanmıştır. Kas mimarisi ultrasonografik ölçümle ve kas sertliği strain elastografi ile belirlenmiştir. Anaerobik güç, çift ayak durarak uzun atlama ve dikey sıçrama ile esneklik oturuşan testleriyle değerlendirilmiştir. Kreatin kinaz aktivitesi, oksidan/antioksidan parametreler venöz kandan ticari kitleler aracılığıyla ölçülmüştür.

**Bulgular:** NHE, deneklerin kilo vermesini sağlamış; egzersizin 5 hafta boyunca bırakılması ile verilen kilolar geri alınmıştır. Egzersiz deneklerin gonyometre ile ölçülen diz açılarında artışa neden olmuş; bu artış egzersizi bırakma ile geri dönmemiştir. 10 haftalık NHE anaerobik performansta egzersizin bırakılmasıyla kısmen geri dönen artışa sebep olmuştur. NHE, biceps femoris uzun başı alanı ve pennasyon açısından 10 haftalık egzersizi bırakma ile geri dönen bir artış oluşturmuştur. Her egzersiz seansını takiben bireylerin kan laktat düzeyinde, kas ağrısında ve yorgunlukta artış tespit edilmiştir. NHE oksidan/antioksidan parametrelerde değişiklik oluşturmazken; 5 haftalık egzersizi bırakma ile total oksidan kapasite (TOK) ve oksidatif stres indeksi (OSI)'nde artışa sebep olmuştur. 10 haftalık egzersizi bırakma süreci bu parametrelerin yeniden azalmasına neden olmuştur.

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**Sonuç:** 10 haftalık ilerleyici NHE ile elde edilen kazanımların çoğunun 5 haftalık detraining ile geri döndüğü gözlenmiştir. Verilerimiz kas gücü ve performansı arttırmak için seçilebilecek egzersiz tipi hakkında yol gösterici olabilir.

**Anahtar kelimeler:** Nordic Hamstring Egzersizi (NHE), detraining, anaerobik güç, oksidatif stres, kreatin kinaz.

Özdamar HC, Kılıç Erkek O, Akkaya HE, Kılıç Toprak E, Bor Küçükkatay ZM. Randomize kontrollü bir çalışma: sağlıklı genç erkeklerde 10 haftalık Nordic Hamstring egzersiz eğitimi ve onu izleyen egzersiz bırakma sürelerinin etkileri. Pam Tıp Derg 2022;15:756-771.

## Introduction

Hamstring strain injuries are the most common injuries in sportsmen, such as athletes, American football, rugby, baseball, cricket and football players [1]. Nordic Hamstring Exercise (NHE) is a widely used type of exercise that doesn't require special equipment and consists of eccentric muscle contractions on the floor in which body weight is used as resistance [1, 2]. Hamstring muscle activation in both joints during NHE requires more maximal eccentric muscle strength than traditional hamstring muscle exercises [3]. Hamstring muscle responses of to eccentric muscle strength training are multifactorial. They contain changes in muscle structure such as increase in motor unit discharge [4], hypertrophy, lengthening of muscle fascicles and decrease in pennation angle (PA) [5, 6].

Eccentric muscle contraction and strenuous exercise may also cause inflammation and muscle damage [7]. With exercise-induced muscle damage, there is a shift towards decreased glucose uptake and insulin sensitivity, impaired glycogen synthesis and non-oxidative metabolism [8]. Detraining, which may be defined as the process of giving up exercise partially or completely is a common issue especially in sedentary individuals. Although oxidative responses to different types of exercise have been studied [9-11], up to our knowledge there is no study in the literature investigating possible alterations in oxidative stress parameters, creatine kinase (CK) activity and blood lactate concentrations in response to NHE and detraining. This study was carried out to elucidate the effect of NHE and subsequent detraining on these parameters. Data obtained from the results of the study was predicted to provide important information to exercise physiologists, physiotherapists and trainers in the selection of exercise type to be performed. In addition, learning which of the possible post-NHE gains will be lost after the detraining

process may be encouraging for continuity to exercise.

## Materials and methods

### Participants

The study protocol was approved by the Medical Ethics Committee of the participating university and the study was conducted in accordance with the Declaration of Helsinki. Participants were selected from volunteer, healthy, male, sedentary (active), students aged 18-30. The subjects were randomly divided into 2 groups as the exercise (n=20) and age, gender matched control group (n=20). Each individual was given detailed information including risks, written informed consent was obtained from all patients, inconvenience that might be encountered.

Exclusion criteria for the participants were; having BMI  $\geq 30$  kg/m<sup>2</sup>, history of hamstring and/or anterior cruciate ligament injury during the last year, presence of any known disease, having a history of trunk-hip, lower extremity injuries, using alcohol and/or drugs, smoking and the vegetarian type of diet.

### Nordic Hamstring exercise and detraining protocols

A standard period of exercise and exercise test was performed by the students at the exercise laboratory of Health Services Vocational School, Karamanoğlu Mehmetbey University. 3 sessions of NHE familiarization trials were applied to the participants for 1 week before starting the exercise sessions. The subjects took a position with their arms bent at the elbow joints and their palms facing forward at the level of the shoulder joints. A partner supported them by holding their ankles [6]. The participants descended slowly and gradually to the ground using their hamstring muscles, while the trunk and hips remained stable [3]. Later, they used their upper body and arms to return to the starting position. To standardize

the hamstring eccentric torque in the initial phase, the time for the individual to land on the ground was determined as 4 seconds. Subjects were asked to land as long as possible using their hamstrings [3], thereby the hamstring eccentric torque raised as they approached the ground. In order to warm up, pedaling at a pace of 5 minutes on a stationary bicycle and static stretching exercises for the hamstring muscles for 30 seconds, 3 repetitions were applied [12]. 1-minute rest periods were implemented between sets. Participants were given a 10-week progressive NHE training [3], followed by detraining of 5/10 weeks during which, the

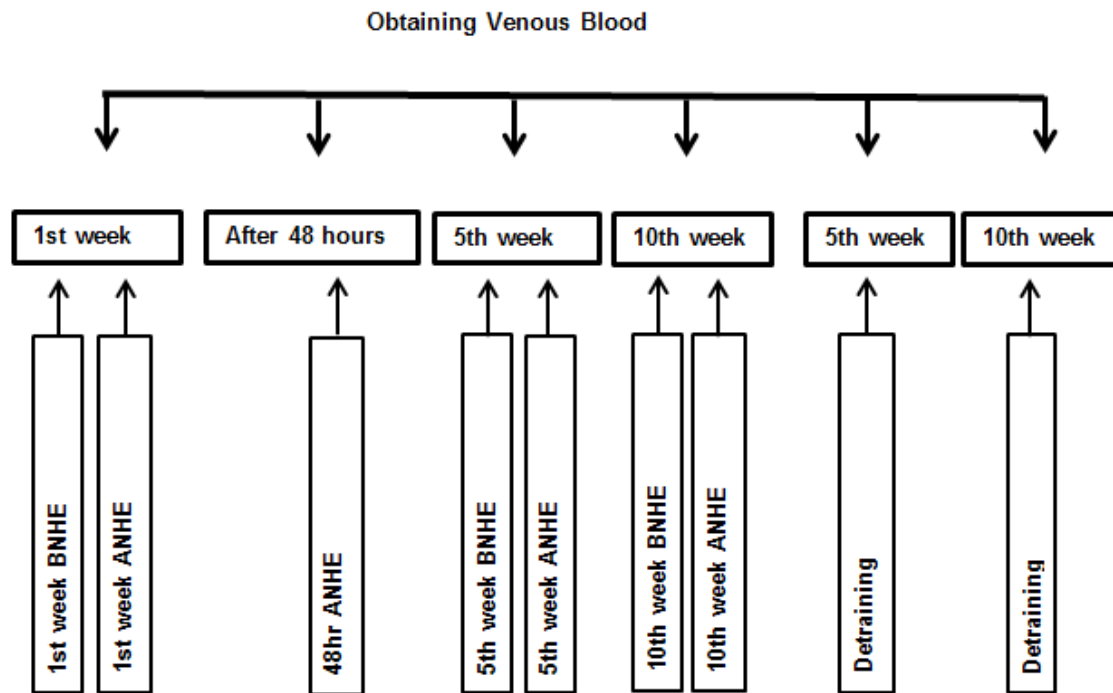
subjects were asked to return their past life routines (Table 1).

**Venous blood samples**

10 ml of blood from antecubital veins was obtained (1 ml at 48 hours following acute exercise) as demonstrated in (Figure 1). Serum was used for oxidative stress parameters, whereas plasma was used for the determination of CK activity. Blood samples were centrifuged at 3000 rpm for 10 minutes to obtain plasma. Plasma samples were stored at -80°C. Samples were appropriately transported to the Physiology Laboratory of Pamukkale University.

**Table 1.** NHE protocol

Weeks	Sessions per week	Sets and repetitions
1	1	2x5
2	2	2x6
3	3	3x6-8
4	3	3x8-10
5-10	3	3x12-10-8



**Figure 1.** Venous blood collection times from NHE group  
 BNHE: Before Nordic Hamstring Exercise, ANHE: After Nordic Hamstring Exercise

**Demographic assessments**

Body composition of the subjects was measured by a body composition analyzer (Jawon Gaia 359 Plus, South Korea).

**Flexibility measurements**

Hamstring flexibility measurements were

performed by a physiotherapist using a goniometer. While the subjects were in the supine position, the hip and knee were placed in 90° positions, and the knee was passively brought to extension until hamstring muscle tension was felt [13]. The individuals without bending their knees, moved their fingers along the measuring table as far as possible. The test

was repeated 3 times and the highest value was recorded in "cm" [14].

### Assessment of anaerobic power tests

Standing long jump test was performed with a fixed meter (m) on the ground. Each subject stood behind the meter while their feet slightly apart. Later, they attempted with two feet take-off to jump as far as possible, swinging their arms and bending knees to provide forward drive and land on both their feet without falling backwards. The participants performed the test 3 times with 3-minute break between repetitions. The best value gained was recorded in "cm" [15].

For the vertical jump test, subjects stood with the dominant side facing the wall. Hand fingertips reach heights were determined. Then, subjects jumped vertically as high as possible. This height was marked. The jump height was calculated by subtracting this height from the reach height. The test was performed three times and best scores were recorded. Anaerobic power calculation was made according the following formula [16];

$$P = \sqrt{4.9} \cdot (W) \cdot \sqrt{D}$$

P=Power (kg.m/s)

W=Body Weight (kg)

D=Jumping Distance (m)

$\sqrt{4.9}$ =Constant value (s)

### Assessment of muscle architecture

#### Ultrasonography and elastography

Biceps Femoris Long Head (BFLH), being the most commonly injured hamstring muscle [17] was examined ultrasonographically (Aplio 500 platinum Ultrasound Device Toshiba-Canon Medical Systems Corporation, Japan) using a linear probe. The frequency range of the ultrasound device was 5-14 megahertz. The radiologist was double-blind to the study. Individuals were prone on a standard treatment table. Measurements were applied to the dominant leg. BFLH was viewed cross-sectionally and longitudinally from its distal to proximal musculotendinous junction. Cross-sectional images were acquired along the length of the muscle from the most distal cross-sectional image that could be traced and measured, which is just proximal to the musculotendinous

junction to the gluteal fold. Longitudinal images were also recorded. BFLH fascicle length, area, PA were determined (Figure 2).

Ultrasound elastography is based on the principle that tissue compression creates tension within the tissue [18]. The applied pressure is marked by an indicator on the screen. Blue color indicated hard, red color soft, green and yellow indicated medium-hard tissues. The strain is less in hard, more in soft tissues [19]. Therefore, tissue stiffness can be estimated by measuring the tissue tension caused by compression [20]. A semi-quantitative measurement value called Strain ratio can be obtained by proportioning the region of interest values of a reference tissue (usually subcutaneous fat tissue) to the tissue to be evaluated [18, 19, 21]. The most swollen region of the hamstring muscle was assessed. Strain indices for hamstring muscle were measured by the ratio of hamstring muscle (T) and subcutaneous adipose tissue (R). Strain ratio value (R/T) reflecting the muscle tension was calculated automatically (Figure 3). Measurements were obtained 5 times; the mean values were recorded.

### Measurements of muscle damage

#### Creatine kinase (CK) activity measurements

CK activities were investigated using the Biovision Creatinine Colorimetric/Fluorometric Assay Kit using plasma. The working principle of the kit may be explained as follows; Creatine kinase converts creatine into phosphocreatine and ADP. The generated phosphocreatine and ADP reacts with CK Enzyme Mix to form an intermediate, which reduces a colorless Probe to a colored product with strong absorbance at 450 nm.

#### Blood lactate measurements

Blood lactate concentrations were determined from capillary blood taken from fingertips via a lactate analyzer (Cera Chek Lactate Plus, Korea).

#### Pain assesment

A 10-cm visual analog scale (VAS) was used to record the highest grade of muscle soreness sensed by each individual.

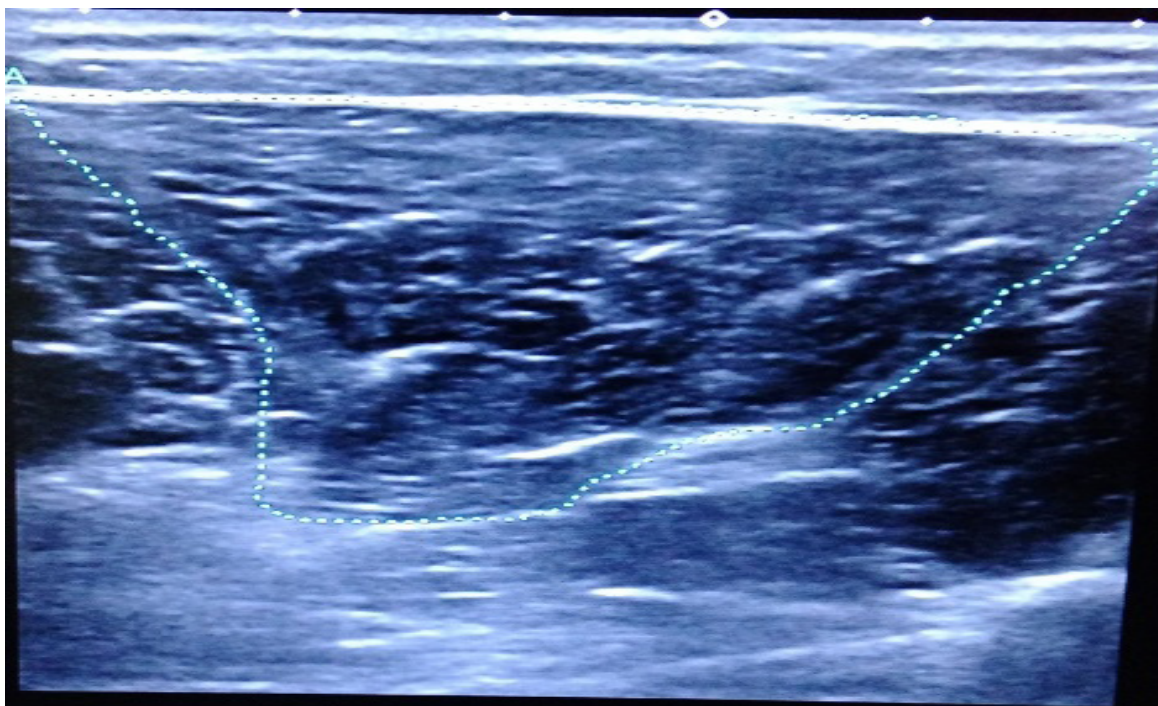


Figure 2. BFLH area measurement USG

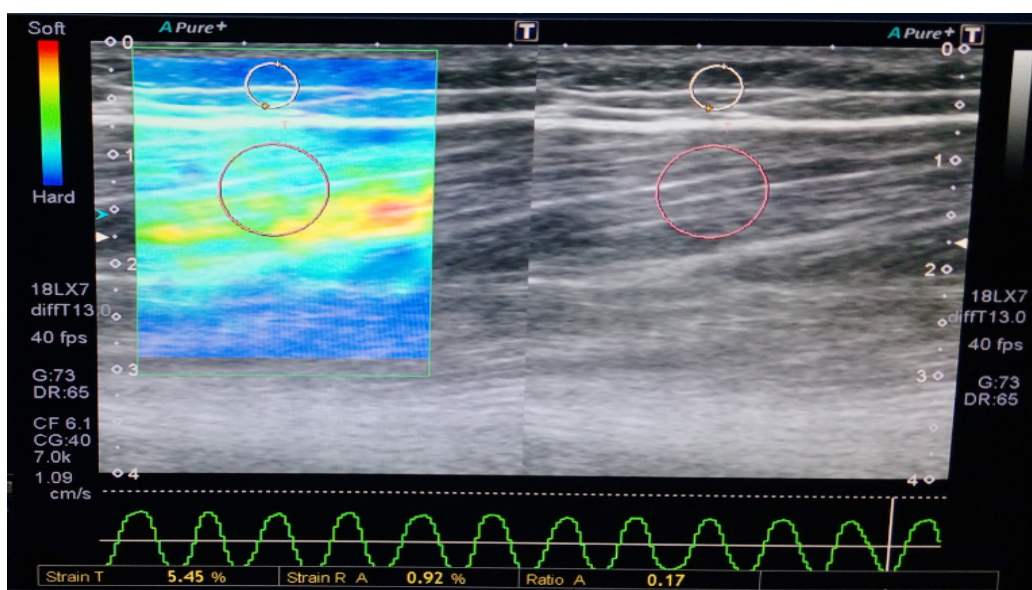


Figure 3. Strain elastography imaging

### Fatigue assessment

The fatigue values felt by exercise groups were marked according to the Borg perceived fatigue scale classification.

### Determination of Total Oxidant Capacity (TOC), Total Antioxidant Capacity (TAC) and Oxidative Stress Index (OSI)

Serum TOC and TAC were measured via an automated colorimetric method through a commercial kit (RelAssay Diagnostic, Turkey) [22, 23]. OSI was calculated according to the

following formula [24].  $OSI = \frac{TOC (\mu\text{mol H}_2\text{O}_2 \text{ Eqv/L})}{TAC (\mu\text{mol Trolox Eqv/L})} \times 100$ .

### Statistical analyses

Given that the difference between groups would have a large effect size ( $d=1.08$ ) a power analysis was performed before the study. Accordingly, when at least 32 people were included in the study ( $n=16$ ), that would result in 90% power with 95% confidence level. Data were analyzed with SPSS 25.0 (IBM SPSS Statistics 25 software (Armonk, NY: IBM Corp.))

package program. Continuous variables were expressed as mean  $\pm$  Standard Error (SE) and categorical variables were demonstrated as numbers and percentages. The suitability of the data for normal distribution was examined with the Shapiro-Wilk test. Considering that there may be loss of participants, 4 more individuals were added to each group (n=20). For independent groups comparisons, when parametric test assumptions were provided we used Independent samples t test and when parametric test assumptions were not provided we used Mann-Whitney U test. For dependent group comparisons; if parametric test conditions were satisfied Paired Samples t test and Repeated Measures ANOVA (post hoc: Bonferroni method) were used; and if parametric test conditions were not satisfied Wilcoxon signed rank test and Friedman test (post hoc: Dunn test) were used.  $P < 0.05$  was considered as statistically significant.

## Results

All subjects completed exercise protocols without problems. There was no difference between control and exercise groups before Nordic Hamstring exercise (BNHE) measurements. Since the exercise intensity reached maximum level in the 5<sup>th</sup> week, 5<sup>th</sup> week measurements were made before the maximum exercise, ie BNHE. 10<sup>th</sup> week measurements were obtained after Nordic Hamstring exercise (ANHE).

## Demographics characteristic

The exercise group lost weight at a statistically significant level on the 10<sup>th</sup> week measurements compared to BNHE ( $p=0.02$ ). The 5-week detraining process caused individuals to gain statistically significant weight compared to the 10<sup>th</sup> week ( $p=0.002$ ) and BNHE ( $p=0.009$ ) groups. Additionally, detraining for 5 weeks, caused increment in TBW compared to BNHE ( $p=0.029$ ), raise in BMI compared to BNHE ( $p=0.015$ ) and 10<sup>th</sup> week group ( $p=0.001$ ). PBF of detraining group was higher than that of 10<sup>th</sup> week group ( $p=0.041$ ). 10<sup>th</sup> week detraining measurements couldn't be performed reliably due to a problem with the analyzer. Demographic characteristics of participants are demonstrated in (Table 2).

## Flexibility measurements

NHE of 10 weeks caused a statistically significant increase in right ( $p=0.019$ ) and left knee ( $p=0.015$ ) angles compared to BNHE. Detraining did not cause any alteration in flexibility measurements (Table 3).

## Anaerobic performance assessments

NHE of 10 weeks led to increment in standing long jump test compared to BNHE ( $p=0.0001$ ) and the 5<sup>th</sup> week NHE group ( $p=0.0001$ ). Vertical Jump test scores of NHE of 10-week group was higher than the 5-week NHE group ( $p=0.009$ ). Detraining of 5 ( $p=0.0001$ ) and 10 weeks ( $p=0.0002$ ) yielded decrement in standing long jump test compared to 10-week NHE group (Table 4).

**Table 2.** Demographic data of the participants

	Control		Exercise		Detraining
	1 <sup>st</sup> week mean $\pm$ SE	BNHE mean $\pm$ SE	5 <sup>th</sup> week mean $\pm$ SE	10 <sup>th</sup> week mean $\pm$ SE	5 <sup>th</sup> week mean $\pm$ SE
WEIGHT (kg)	74 $\pm$ 2.33	73.14 $\pm$ 2.81	72.98 $\pm$ 2.59	72.64 $\pm$ 2.5'	74.1 $\pm$ 2.49 <sup>*,##</sup>
LBM (kg)	59.74 $\pm$ 1.56	59.86 $\pm$ 1.66	60.17 $\pm$ 1.6	60.53 $\pm$ 1.59	61.03 $\pm$ 1.62
SLM (kg)	55.43 $\pm$ 1.42	55.62 $\pm$ 1.51	55.91 $\pm$ 1.46	56.3 $\pm$ 1.46	56.72 $\pm$ 1.49
TBW (kg)	43.01 $\pm$ 1.12	43.11 $\pm$ 1.19	43.32 $\pm$ 1.15	43.59 $\pm$ 1.14	43.94 $\pm$ 1.17'
BMI (kg/m <sup>2</sup> )	23.59 $\pm$ 0.51	23.08 $\pm$ 0.67	23.05 $\pm$ 0.63	22.94 $\pm$ 0.64	23.41 $\pm$ 0.62 <sup>*,##</sup>
PBF (%)	18.95 $\pm$ 0.88	17.49 $\pm$ 1.17	16.99 $\pm$ 1.06	16.07 $\pm$ 1.06	17.19 $\pm$ 1.05 <sup>#</sup>
BMR (kcal)	1653 $\pm$ 28.79	1657.95 $\pm$ 28.81	1662.1 $\pm$ 28.11	1666.4 $\pm$ 27.84	1669.45 $\pm$ 29.74

BNHE: Before Nordic Hamstring Exercise, LBM: Lean Body Mass, SLM: Soft Lean Mass

TBW: Total Body Water, BMI: Body Mass Index, PBF: Percent Body Fat

BMR: Basal Metabolic Rate, mean  $\pm$  Standard Error, n=20

\* $p < 0.05$  difference from BNHE; '  $p < 0.01$  difference from BNHE

# $p < 0.05$  difference from 10<sup>th</sup> week; ## $p < 0.01$  difference from 10<sup>th</sup> week

**Table 3.** Flexibility measurements of the subjects

	Control		Exercise		Detraining	
	1 <sup>st</sup> week	BNHE	5 <sup>th</sup> week	10 <sup>th</sup> week	5 <sup>th</sup> week	10 <sup>th</sup> week
	Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE
<b>Sit-Reach Test (cm)</b>	4.5±1.82	8.45±2.05	10.2±2.21	11.13±2.13	9.48±2.35	9.47±2.38
<b>Right Knee Angle</b>	16.1±1.3	17.55±1.73	15.1±1.48	11.2±1.26*	13.7±1.33	10.37±1.59
<b>Left Knee Angle</b>	13.95±1.2	17.75±1.58	12.6±1.12	10.25±1.1*	14.6±0.32	11±1.45

BNHE: Before Nordic Hamstring Exercise, mean ± Standard Error, n=20  
\*p<0.05 difference from BNHE

**Table 4.** Anaerobic performance test assessment of participants

	Control		Exercise		Detraining	
	1 <sup>st</sup> week	BNHE	5 <sup>th</sup> week	10 <sup>th</sup> week	5 <sup>th</sup> week	10 <sup>th</sup> week
	Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE
<b>Standing long jump test (cm)</b>	146.4±4.35	141.1±4.2	141.48±3.62	159.85±3.04*,**	143.2±2.59 <sup>#</sup>	145.08±2.88 <sup>#</sup>
<b>Vertical Jump test (kg.m/s)</b>	1022.2±36.38	1015.76±37.96	1009.77±37.52	1056.64±38.12**	1052.64±37.34	1036.2±40.14

BNHE: Before Nordic Hamstring Exercise, mean ± Standard Error, n=20  
\*p<0.01 difference from BNHE; \*\*p<0.01 difference from 5<sup>th</sup> week exercise  
<sup>#</sup>p<0.01 difference from 10<sup>th</sup> week exercise

### Ultrasonography measurements and strain elastography of the participants

Table 5 demonstrates that PA of the detraining for 10-week group was less than that of the 9<sup>th</sup> week exercise group ( $p=0.0001$ ). In the exercise group, the PA measurement values at the 9<sup>th</sup> week were statistically significantly higher than the 1<sup>st</sup> week values ( $p=0.002$ ). Detraining for 10 weeks resulted in decrement of BFLH muscle area compared to exercise 9<sup>th</sup> week group ( $p=0.001$ ). Exercising for 9 weeks caused a statistically significant increment in BFLH area ( $p=0.006$ ).

### Muscle damage assessments

#### Creatin kinase activity

There was no statistically significant difference in subjects' plasma CK activities (Figure 4).

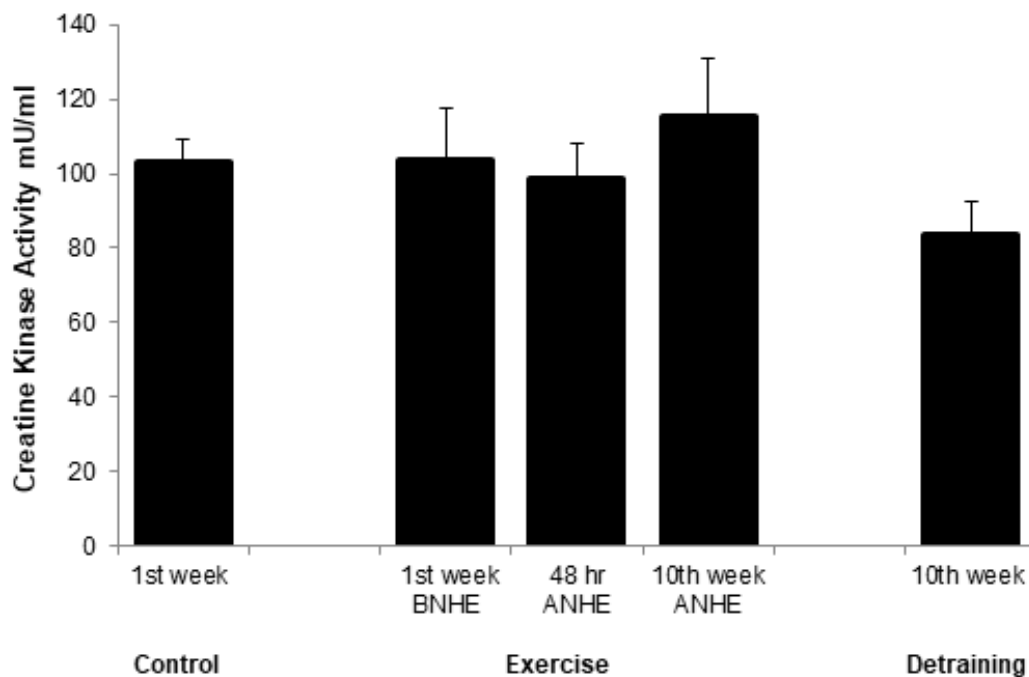
#### Blood lactate concentrations

Figure 5 shows that blood lactate levels of ANHE groups were statistically significantly higher than corresponding BNHE values ( $p=0.001$ ,  $p=0.002$ , respectively). When BNHE values were compared with each other, the 10<sup>th</sup> week BNHE blood lactate concentration was observed to be higher than that of the 1<sup>st</sup> week ( $p=0.027$ ).

**Table 5.** Ultrasonography and strain elastography measurements of the participants

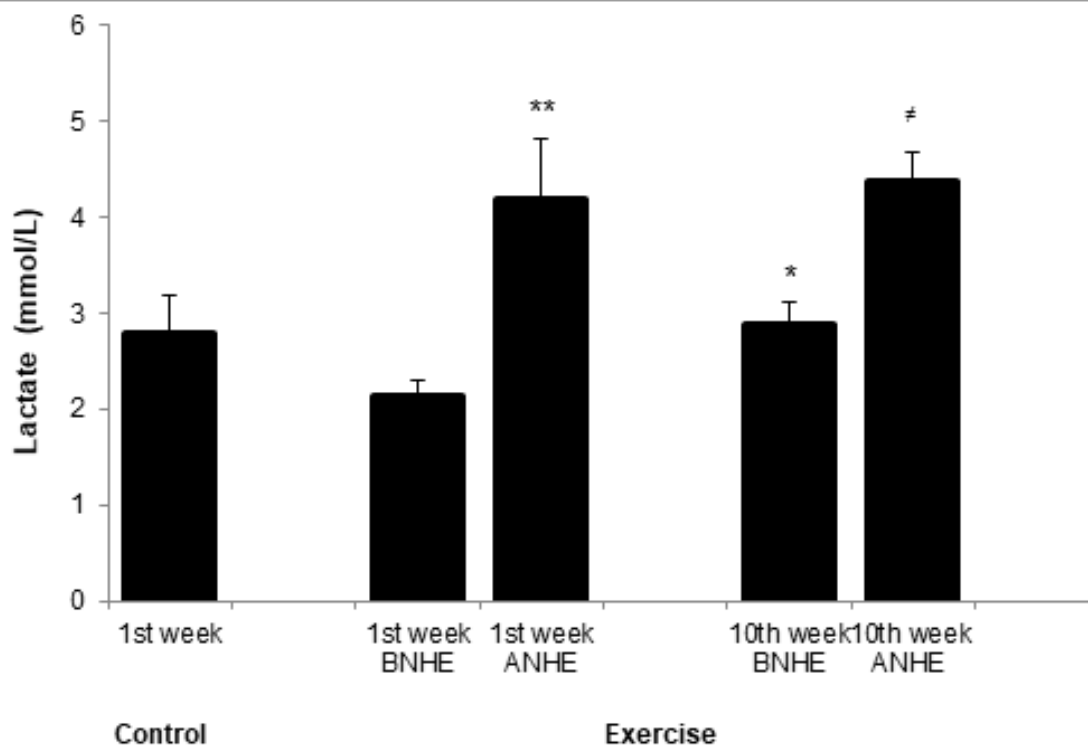
	Control		Exercise		Detraining	
	1 <sup>st</sup> week	BNHE	9 <sup>th</sup> week	10 <sup>th</sup> week	10 <sup>th</sup> week	10 <sup>th</sup> week
	Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE
<b>BFLH fascicle length (cm)</b>	9.34±0.36	8.68±0.26	8.89±0.26	8.97±0.14		
<b>BFLH Area (cm<sup>2</sup>)</b>	11.36±0.38	11.29±0.4	12.34±0.42*	11.19±0.43 <sup>#</sup>		
<b>PA</b>	11.8±0.51	12.85±0.53	15.4±0.6*	12.35±0.47 <sup>#</sup>		
<b>BFLH strain ratio</b>	0.20±0.002	0.23±0.03	0.25±0.03	0.15±0.01**		

BNHE: Before Nordic Hamstring Exercise, mean ± Standard Error, n=19  
\*p<0.01 difference from BNHE; \*\*p<0.05 difference from 9<sup>th</sup> week group  
<sup>#</sup>p<0.01 difference from 9<sup>th</sup> week group



**Figure 4.** Plasma CK activity values

BNHE: Before Nordic Hamstring Exercise, ANHE: After Nordic Hamstring Exercise  
 mean ± Standard Error, n=13



**Figure 5.** Blood lactate concentrations

BNHE: Before Nordic Hamstring Exercise, ANHE: After Nordic Hamstring Exercise  
 mean ± Standard Error, n=18

\* $p < 0.05$  difference from 1<sup>st</sup> week BNHE; \*\* $p < 0.01$  difference from 1<sup>st</sup> week BNHE

# $p < 0.01$  difference from 10<sup>th</sup> week BNHE



**Visual analog scale of exercise groups**

Soreness assessments are demonstrated in Figure 6. In all weeks, participants stated that they felt more pain following exercise sessions compared to BNHE ( $p=0.0001$ ,  $p=0.0001$ ,  $p=0.0001$ , respectively).

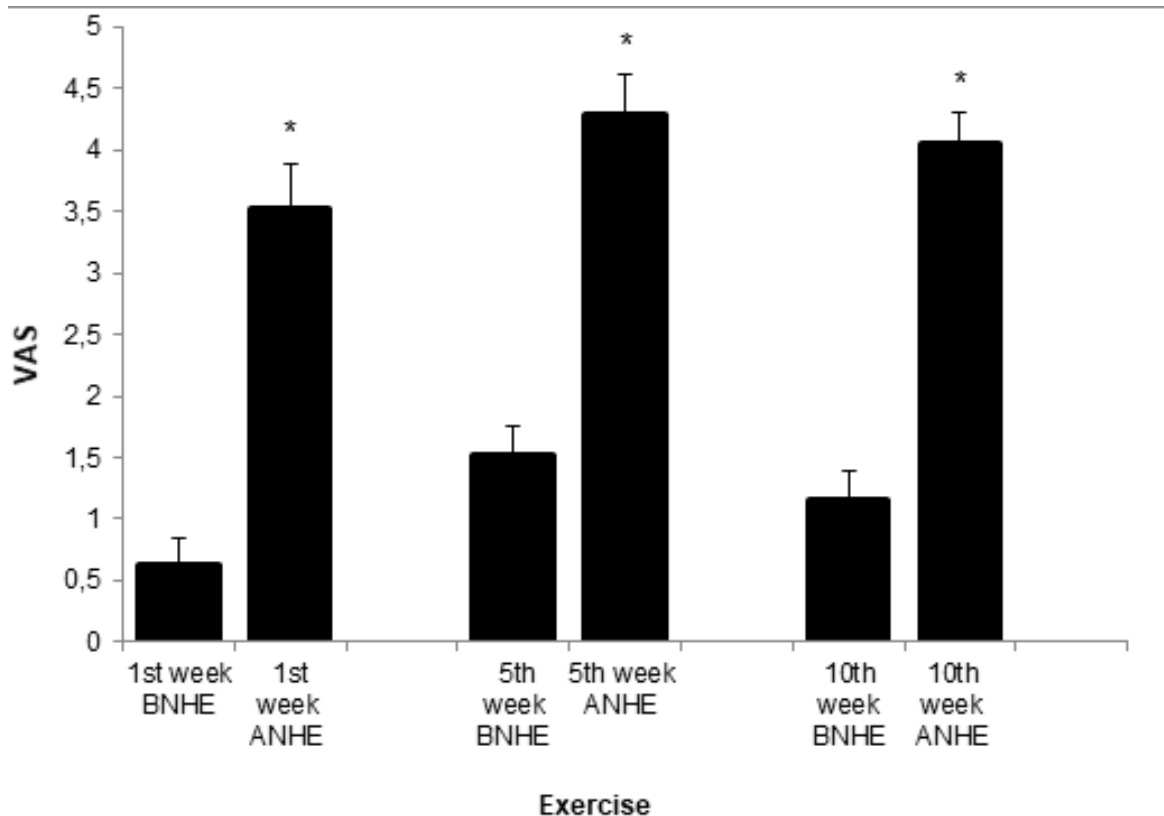
**Fatigue assessments of exercise groups**

Fatigue levels expressed according to Borg scale following exercise sessions, were found to be significantly higher in all groups compared to corresponding BNHE ( $p=0.002$ ,  $p=0.0001$ ,  $p=0.0001$ , respectively). The subjects stated that they felt more fatigue in the 5<sup>th</sup> week BNHE compared to the 1<sup>st</sup> week BNHE ( $p=0.006$ ). Additionally, the Borg ratio perceived fatigue values of the 5<sup>th</sup> and 10<sup>th</sup> week ANHE groups were higher than the 1<sup>st</sup> week ANHE value ( $p=0.022$ ,  $p=0.0001$  respectively), (Figure 7).

**Total Oxidant Capacity (TOC), Total Antioxidant Capacity (TAC) and Oxidative Stress Index (OSI)**

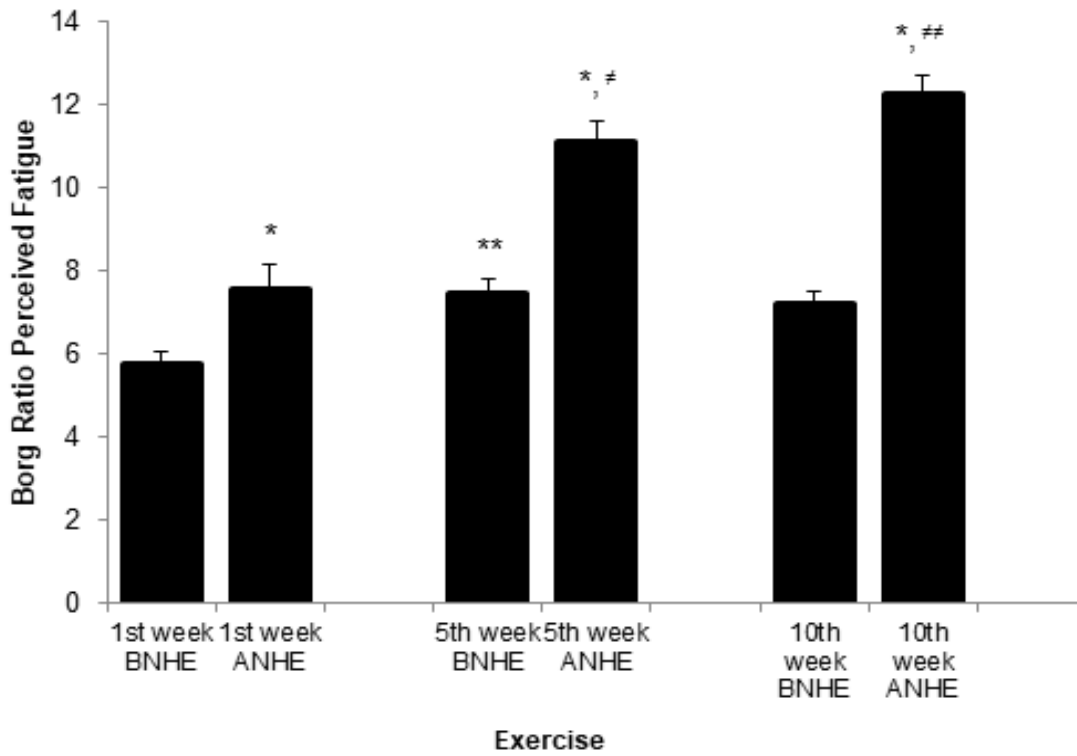
Figure 8 shows TOC measurements of individuals. Detraining for 5 weeks caused a statistically significant increase in TOC values compared to the 5<sup>th</sup> week BNHE ( $p=0.048$ ), ANHE ( $p=0.029$ ), 10<sup>th</sup> week BNHE ( $p=0.001$ ) and ANHE ( $p=0.029$ ) values. Detraining for 10 weeks caused a statistically significant decrease in TOC compared to the 5<sup>th</sup> week detraining group ( $p=0.001$ ).

Figure 9 demonstrates that no statistically significant alteration was observed in TAC. As shown in Figure 10, detraining OSI measurements at the 5<sup>th</sup> week were found to be statistically significantly higher than the 10<sup>th</sup> week BNHE group ( $p=0.037$ ). Quitting exercise for 10 weeks caused decrement in OSI values compared to detraining of 5 weeks ( $p=0.001$ ).



**Figure 6.** VAS of the subjects

BNHE: Before Nordic Hamstring Exercise, ANHE: After Nordic Hamstring Exercise  
 mean ± Standard Error, n=20  
 \* $p<0.01$  difference from BNHE



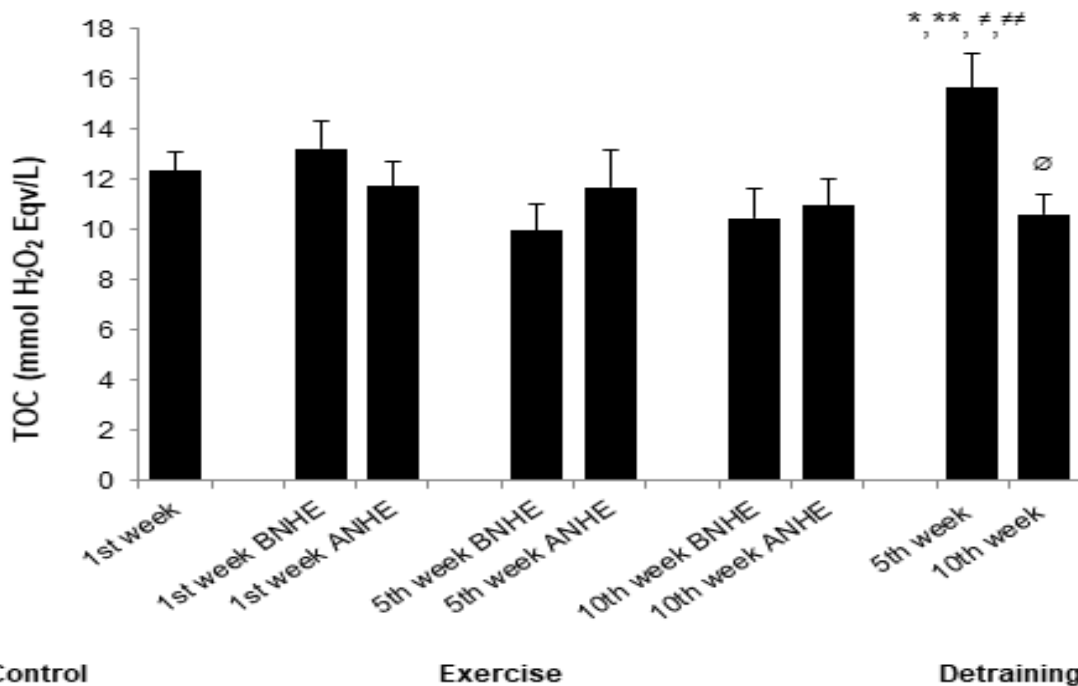
**Figure 7.** Borg ratio perceived fatigue assessment of the participants

BNHE: Before Nordic Hamstring Exercise, ANHE: After Nordic Hamstring Exercise  
 mean ± Standard Error, n=20

\*p<0.01 difference from BNHE, \*\*p<0.01 difference from 1<sup>st</sup> week BNHE

#p<0.05 difference from the 1<sup>st</sup> week ANHE group

##p<0.01 difference from the ANHE group in the 1<sup>st</sup> week



**Figure 8.** Serum TOC values

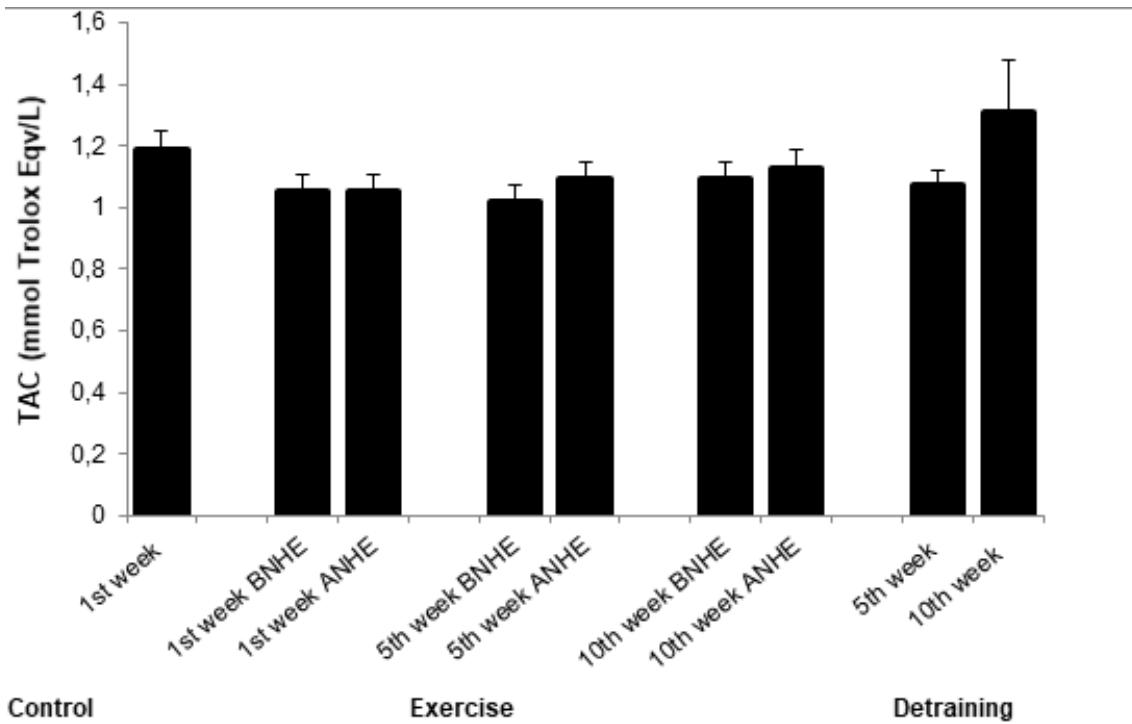
TOC: Total Oxidant Capacity, BNHE: Before Nordic Hamstring Exercise

ANHE: After Nordic Hamstring Exercise, mean ± Standard Error, n=17

°p<0.05 difference from 5<sup>th</sup> week BNHE, \*\*p<0.05 difference from 5<sup>th</sup> week ANHE group

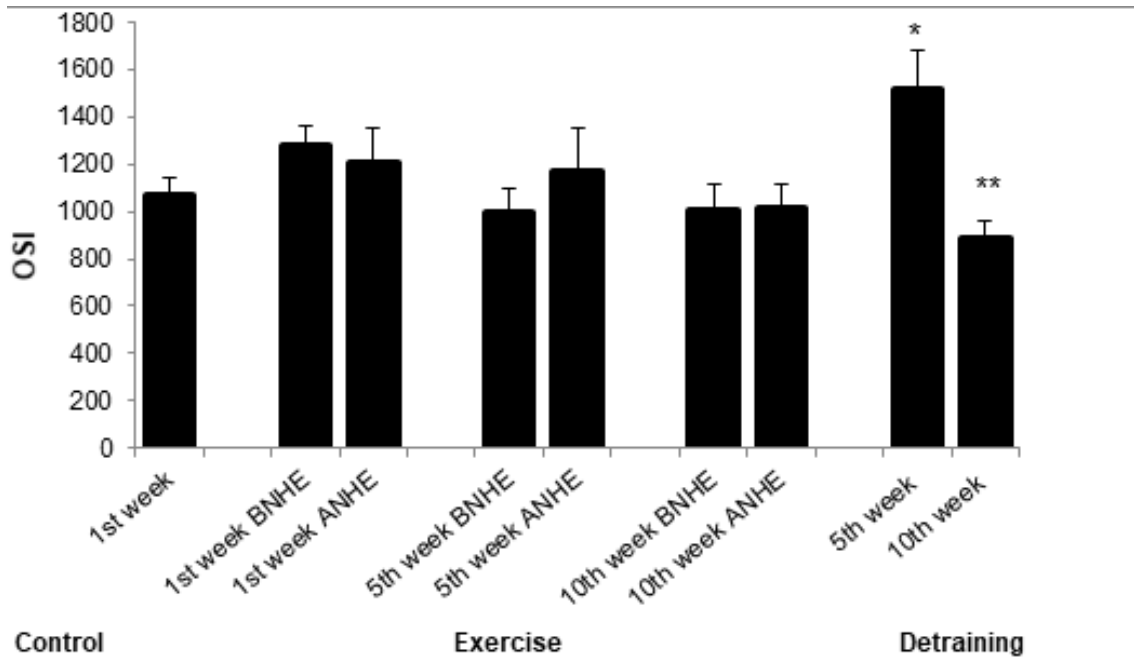
#p<0.01 difference from 10<sup>th</sup> week BNHE, ##p<0.05 difference from 10<sup>th</sup> week ANHE

°p<0.01 difference from 5<sup>th</sup> week detraining group



**Figure 9.** Serum TAC values

BNHE: Before Nordic Hamstring Exercise, ANHE: After Nordic Hamstring Exercise  
 mean  $\pm$  Standard Error, n=17



**Figure 10.** Serum OSI values

BNHE: Before Nordic Hamstring Exercise, ANHE: After Nordic Hamstring Exercise  
 mean  $\pm$  Standard Error, n=17

\* $p < 0.05$  difference from the 10<sup>th</sup> week BNHE

\*\* $p < 0.01$  difference from the 5<sup>th</sup> week detraining group

## Discussion

Effects of 10-week progressive NHE training, which is a simple but effective method used for the development of hamstring muscle strength, on demographic characters, flexibility, anaerobic parameters, USG and strain elastography determinations, muscle damage, pain and fatigue assessments as well as oxidative stress were examined. The alterations in these parameters in response to two separate detraining processes (5 and 10 weeks) were also investigated. Compliance with the exercise regimens is usually low; many people upon risk of injury begin exercise programs but generally give up some time later. In this case, even if a reduction in the risk of injury and/or illness has been detected in response to exercise, there isn't enough information about how long these positive alterations can be preserved after cessation of exercise (detraining).

Basal values of exercise groups were similar to the control. 10 weeks of progressive NHE caused sedentary, active individuals to lose weight, and an increase in their thigh circumference. In addition to regaining the weight lost, 5 weeks of detraining caused an increment in the subjects' total body water (TBW); body mass index (BMI); percent body fat (PBF); arm, leg, trunk lean body mass (LBM) and a decrement in right thigh circumference. Right leg was dominant in most students. 10 weeks of NHE produced an increase in both knee angles; which were not affected by detraining. At the end of the 10<sup>th</sup> week, the individuals' anaerobic performance tests were increased while, the positive alteration in standing long jump test was reversed in response to detraining. BFLH area and PA were increased by NHE; the change in these parameters were reversed by detraining of 10 weeks. Although NHE didn't cause any change in BFLH strain ratio, cessation of the exercise yielded this parameter to fall below basal values. While the exercise protocol applied didn't alter oxidative stress, detraining for 5 weeks caused increment in TOC and OSI. In response to detraining for 10 weeks, these two parameters decreased again.

In the literature no weight loss in response to NHE was observed in sedentary and recreationally active individuals [25]. No dietary advice within the scope of our study was implemented. However, most of the students'

staying in dormitories may have contributed to their weight loss. Any study examining the change in TBW, PBF and LBM following NHE has been found. Although these parameters did not change during NHE, they increased in response to detraining for 5 weeks which may be explained by the weight gain of individuals during detraining.

Eccentric training has been incorporated with increased muscle flexibility and falled risk of hamstring strain injury in sports [26-28]. Our data, demonstrated that NHE for 10 weeks increases the flexibility of the individuals evaluated by the knee angle, but does not alter sit-reach test. Knee angle measurement examines regional hamstring flexibility, while sit-reach test determines lumbar extensor, hip extensor, and hamstring flexibility [29, 14]. Thus, our findings have shown that NHE mainly increases hamstring flexibility. Detraining did not reverse the hamstring resilience gain. How long hamstring flexibility will be maintained in response to quitting NHE may be tested by studies involving longer detraining processes. Performance in sports is mainly related to concepts such as anaerobic explosive power, flexibility, agility, and speed [30]. As expected, progressive NHE applied for 10 weeks improved anaerobic performance. The gains obtained in the standing long jump test were reversed with detraining, while the ones in vertical jump test didn't change. Our detraining protocols didn't also reverse the hamstring flexibility parameters mentioned above. Flexibility is a measurement of physical fitness and anaerobic performance [30]. The vertical jump response to detraining might be determined by the unchanged alterations in knee angles. Standing long jump test requires arm activity in addition to the lower extremity. Detraining for 5 weeks was not enough to reverse the alterations in vertical jump test and flexibility.

Adaptability of the structure and function of the hamstring muscle in response to a variety of exercises is important in reducing hamstring injuries [2]. Isolated eccentric resistance training causes more neural and morphological adaptations than isolated concentric training [31]. BFLH fascicle length, strain ratio, PA, and cross-sectional area are usually determined to assess muscle architecture [21, 2]. Results of the studies investigating muscle architecture in response to NHE vary depending on training

status of the subjects, NHE being performed against an external load or with body weight, exercise intensity and duration [32, 33]. The range of motion and speed during NHE varied among participants. These differences may have influenced architectural adaptations of the muscle [12, 34].

The potential benefits of long-term eccentric exercise are clinically important in healthy populations [35]. However, eccentric strenuous and acute exercise can yield deep muscle structural and functional deterioration [36]. Duhig et al. [37] demonstrated no statistically significant alteration in CK levels following 5 weeks of NHE. Abaïdia et al. [38] applied an exercise protocol using an isokinetic dynamometer consisting of 5 sets and 15 repetitions to physically active men and found that, CK levels increased in 24 hours following exercise but returned to normal at the 48<sup>th</sup> hour. In the current study, CK activity, but not level was investigated and a progressive NHE program was applied. Acute exercise-related CK activity was measured at the 48<sup>th</sup> hour following the exercise in the 1<sup>st</sup> week, whereas the effect of long-term exercise on plasma CK activity in the 10<sup>th</sup> week was examined immediately after the exercise. No change in CK activity was found, supporting Abaïdia et al. [38] acute exercise findings. In the first session, NHE of 2 sets and 5 repetitions was probably not enough to cause muscle damage, thus alter CK activity. The familiarization process might also have contributed to this result. The fact that CK activity was unchanged in the 10<sup>th</sup> week, may be explained by adaptation to long-term exercise. The detraining processes applied, was demonstrated not to cause any significant alteration in CK activity for the first time in literature. Blood lactate concentrations of sedentary active subjects were measured immediately after NHE in the 1<sup>st</sup> and 10<sup>th</sup> weeks and were found to be increased, as expected. Acute increase in blood lactate concentration was also detected despite 10 weeks of adaptation to progressive NHE. No study in literature was found that measured blood lactate levels in response to NHE.

Eccentric exercise is known to cause acute and delayed onset muscle soreness [39]. We questioned VAS was to sedentary active subjects just after the 1<sup>st</sup>, 5<sup>th</sup> and 10<sup>th</sup> week exercise sessions, and a significant increase

has been found in all of them compared to their BNHEs. 1<sup>st</sup> week VAS data reflect the effect of acute, low intensity exercise. The intensity of NHE achieved the highest level in the 5<sup>th</sup> week, and the 5<sup>th</sup> week data shows the soreness in response to this exercise intensity. Between 5-10<sup>th</sup> weeks, the same high-intensity exercise was applied. At the end of the 10<sup>th</sup> week, the individuals who have adapted to this intensity reported subsequent increases in muscle pain in response to acute exercise. Increases in VAS are compatible with blood lactate concentrations. The Borg ratio perceived scale is a widely used method for assessment of fatigue in response to exercise [40]. As predicted, our subjects expressed increases in their fatigue following the 1<sup>st</sup>, 5<sup>th</sup> and 10<sup>th</sup> week exercise sessions. On the other hand, while the pre-exercise Borg ratio reported in the 5<sup>th</sup> week was higher than basal value, the fatigue expressed on the 10<sup>th</sup> week, before exercise was not different from 1<sup>st</sup> and 5<sup>th</sup> week pre-exercise values. Adaptation again, may be the explanation of this issue.

The dramatic elevation in oxygen use during high-intensity exercise is often associated with the formation and accumulation of free radicals. If these free radicals are not neutralized, oxidative damage occurs [41]. The effect of eccentric exercises on the initiation and progression of muscle fiber damage by stimulating the formation of reactive oxygen species was reported [42]. Excessive elevation in oxidative stress together with alteration in redox state is thought to play a role in the pathophysiology and impaired adaptation to exercise. For this reason, it is important to investigate the alterations in oxidative stress in response to exercise [43]. In the literature, when examining the oxidative stress response to exercise, usually specific oxidant-antioxidant enzymes involved were studied [44, 45]. In order to evaluate the overall oxidant-antioxidant status in the body, we determined TOC, TAC and calculated OSI. No significant alteration in TOC, TAC and OSI in response to progressive NHE was observed in the current study. We applied a low-intensity acute exercise protocol with familiarization. These factors may have affected the lack of change in these parameters in the acute period. Similarly, Unver et al. [46] didn't find any change in TOC, TAC and OSI when they applied acute NHE with 1 set and 7 repetitions to active healthy men at the ages of

19-21. Stagos et al. [47] reported that oxidative stress and TAC wasn't altered after 24, 48, and 72 hours following a single session of isokinetic eccentric exercise in young individuals. Up to our knowledge, there is no study in the literature examining the oxidative stress response to prolonged NHE. Kilic Toprak et al. [48] have found no statistically significant difference in TOC levels in healthy young male in response to chronic 12-week progressive resistance exercise. Unaltered oxidative stress in response to prolonged progressive NHE determined here in, may be attributed to adaptation process.

Alterations in oxidative stress in response to detraining of 5 and 10 weeks was also investigated. 2 and 4 weeks of detraining period was demonstrated to increase oxidative stress markers such as  $O_2^-$  and thiobarbutyric acid levels following swimming exercise (60 minutes/day, 5 days/week, for 8 weeks) in rats. The  $O_2^-$  radical level after detraining of 2 weeks was higher than of 4-week detraining period. The authors also reported high TAC levels during detraining [49]. Our results, in line with the data of Bradic et al's. [49] demonstrate that, 5 weeks of detraining causes increment in TOC and OSI, while detraining of 10 weeks results in decrement (return to basal level) in these parameters. Since Bradic et al's. [49] detraining periods were considerably shorter than ours, these authors could not observe effects of late detraining [49].

The effects of 10-week progressive NHE, which aims to increase the hamstring strength and to eliminate the imbalance in the hamstring/quadriceps muscle strength ratio [50], was demonstrated in young male, sedentary active individuals in the current study. Our results demonstrate that, NHE is useful to increase anaerobic performance, biceps femoris long head area, pennation and knee angle without affecting oxidative stress and CK activity. Some of the gains of NHE were reversed by detraining. These results may guide the selection of the exercise type to increase performance and muscle strength. In order to reveal the effects of NHE on various physiological parameters more clearly, it is necessary to conduct larger studies on women, athletes from various sports branches and different training levels. It is also important to determine how long the gains

obtained through exercise can be preserved by detraining. Further studies with longer detraining periods may also be recommended.

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**Ethics committee approval:** Pamukkale University Non-Interventional Clinical Research approved by the Ethics Committee (date: 08.08.2018 and number: 60116787-020/53191)

#### Authors' contributions to the article

H.C.O., O.K.E., H.E.A., E.K.T. and M.B.K. constructed the main idea and hypothesis of the study. H.C.O., O.K.E., H.E.A., E.K.T. and M.B.K. developed the theory and organized the material and method section. H.C.O. and M.B.K. wrote the discussion part of the article and O.K.E., H.E.A., E.K.T. reviewed and gave the necessary approval. In addition, all authors discussed the entire study and approved the final version.