Urinary steroid profile after the completion of concentric and concentric/eccentric trials with the same total workload

R. Timon¹, G. Olcina¹, P. Tomas-Carus², D. Muñoz¹, F. Toribio³, A. Raimundo² and M. Maynar⁴

¹Department of Physical Education and Sport. Sport Science Faculty, University of Extremadura, Cáceres, Spain, ²Department of Sport and Health, University of Evora, Evora, Portugal, ³Department of Analytical Chemistry and Electrochemistry, School of Sciences, University of Extremadura, Badajoz, Spain, ⁴Department of Physiology, Sport Science Faculty, University of Extremadura, Cáceres, Spain

(Received on December, 2008)

R. TIMON, G. OLCINA, P. TOMAS-CARUS, D. MUÑOZ, F. TORIBIO, A. RAIMUNDO and M. MAYNAR. Urinary steroid profile after the completion of concentric and concentric/eccentric trials with the same total workload. J Physiol Biochem, 65 (2), 105-112, 2009.

High intensity strength training causes changes in steroid hormone concentrations. This could be altered by the muscular contraction type: eccentric or concentric. The aim of this study was to compare the effect of the completion of a short concentric (CON) and concentric/eccentric (CON/ECC) trial on the urinary steroid profile, both with the same total work. 18 males performed the trials on an isokinetic dynamometer (BIODEX III) exercising quadriceps muscles, right and left, on different days. Trial 1(CON): 4x10 Concentric knee extension + relax knee flexion, speed 60°/second; rest 90 seconds between each series and 4 minutes between each leg exercise. Trial 2(CON/ECC): 4x5 concentric knee extension + Eccentric knee flexion under similar conditions. Urine samples were taken before the exercise and one hour after finishing it. Androsterone, Etiocholanolone, DHEA, Androstenedione, Testosterone, Epitestosterone, Dehydrotestosterone, Estrone, B-Estradiol, Tetrahydrocortisone, Tetrahydrocortisol, Cortisone and Cortisol (free, glucoconjugated and sulfoconjugated) urinary values were determined using gas chromatography/mass spectrometry techniques. No significant differences were noted in Total Work and Average Peak Torque, although Maximum Peak Torque in the CON/ECC trial was higher than in the CON trial. These results demonstrate no changes in the steroid profile before and after trials, or when comparing CON to CON/ECC trials. The data suggest that eccentric contractions do not cause hormonal changes different to the ones produced by concentric contractions, when they are performed in strength short trials with the same total workload.

Key words: Androgens, Corticosteroids, Workload, Concentric, Eccentric, Gas chromatography/mass spectrometry.

Correspondence to R. Timón (Fax. +34 927 257 461; e-mail: rtimon@unex.es).

Endurance strength exercise has been shown to produce important changes in the synthesis, metabolism, and urinary excretion of steroids (13, 18, 26). These hormones have been used to evaluate the stress of exercise, thus cortisone and cortisol urinary metabolites are used to evaluate organic catabolic activity and testosterone and its metabolites to evaluate anabolic activity (14, 22). However, these hormonal changes also depend on intensity, workload, number of repetitions, recovery time and contraction types (8, 21, 24, 29). Thus, significant changes are only produced when the load intensity exceeds 70% of one repetition maximum (1 RM) (23, 27).

The modifications produced in the hormonal metabolism as a result of strength exercises with different contraction types have been studied (8, 11, 15). The eccentric contraction has been proved to be more effective in increasing the protein synthetic rate (19) and muscular strength (12), and is able to reach a higher torque peak than the concentric contraction (7). However, it has been documented in several papers that concentric contraction, under the same absolute load, generates higher metabolic stress, higher activation of the sympathetic nervous system and motor unity recruitment (4, 9). On the other hand, the cortisol level was higher in a concentric protocol than in an eccentric one, under the same absolute load (11). However, this higher metabolic stress seems not to affect the normal androgen hormonal response, since no significant changes were observed in total testosterone levels (8). Accordingly, in another study, where the same relative load for the concentric and eccentric trials was used, no significant differences were registered (15).

Studies described above have tried to compare the steroid hormone response by using free weights and 1RM to quantify the load. Nevertheless, the most reliable method to evaluate muscle strength is an isokinetic dynamometer, which standardizes the exercise performance and quantifies the workload in each protocol, thus avoiding any possible errors (20).

Therefore, the main purpose of this research is to observe and to compare the changes in steroid urinary excretion after the completion of concentric and concentric/eccentric short trials with the same total workload, using an isokinetic dynamometer.

Material and Methods

Subjects.- Eighteen university students, who did not usually do strength training, were recruited to participate in this study. The mean $(\pm SD)$ physical characteristics of the subjects were: age, 24.11 ± 2.61 years; height, 1.75 ± 0.06 m; weight, 72.05± 8.33 kg; lean mass, 55.59 ± 4.59 kg; BMC (body mineral content) 2.80 ± 0.41 kg; fat mass 12.91 ± 4.16 kg. Participants underwent a medical examination to discard the possibility of any pathology and were screened for dietary habits and use of anabolic steroids. Taking steroids or adherence to a reduced calorie or low fat diet were exclusion criteria. Subjects were informed about the procedure of the study and they all provided consent by filling in a form. The experiment was developed in agreement with the policy statement of the Declaration of Helsinki.

Experimental design.– The day before the trial, subjects were evaluated by DXA. Lean mass, BMC and fat mass were analyzed. The study employed a counterbalanced design which compared the effect of URINARY STEROIDS IN CON AND CON/ECC TRIALS

the completion of a short concentric and concentric/eccentric trial on the urinary steroid profile. The subjects of the study carried out two trials, with an interval of five days between them. Each subject did the trial at the same time of the day to avoid the influence of circadian rhythms. The temperature and humidity conditions were 29±1.6 °C and 35-40 %, respectively.

The concentric as well as the concentric/eccentric trials were performed on an isokinetic dynamometer (BIODEX System 3), at a speed of 60 degrees per second. The concentric trial consisted of the execution of 4 sets of 10 repetitions for both, left and right quadriceps. A maximal voluntary concentric contraction of the quadriceps was performed during the extension. Then, leg was relaxed and returned to its original position due to gravity action during the flexion. The recovery time was 90 seconds between each set and 4 minutes between each leg exercise. The concentric/eccentric trial consisted of the execution of 4 sets of 5 repetitions, on both quadriceps. A maximal voluntary concentric contraction of the quadriceps was performed in each extension and a maximal voluntary eccentric contraction of the quadriceps was performed in flexion, therefore, muscular contractions performed by each quadriceps were the same in both trials (amounting to 40). The recovery times were similar to the ones set for the concentric trial.

Based on the isokinetic report, information for each leg was collected on the total workload performed (measured in Joules), the maximum and the average torque peak (measured in Newtons per meter) in order to make sure that the total workload in both trials was equal.

Urine analysis.- Urine samples were collected before and one hour after the

completion of both trials. All urine samples were frozen at -20 °C until analysis. Interpretation of the analytical data of the urine assays requires the sample density to be in the range of 1.005-1.025 g/ml, and the pH to be in the normal range of 4.7-7.8. All the study samples fulfilled these conditions.

The creatinine levels of all the samples were determined in order to report the steroid concentrations relative to this fundamental urine parameter. The method of analysis is based on the work of Heinegard & Tiderstrom (10). These assays were performed within 24 h of the urine collection, using Sigma's Creatinine 555-A kit and a UNICAM 5625 spectrophotometer.

By using gas chromatography-mass spectrometry technique (GC/ MS). The following hormones and metabolites -in their free, gluconjugated and sulfoconjugated fractions- were measured: Androsterone, Etiocholanolone, DHEA, Androstenedione, Testosterone, Epitestosterone, Dehydrotestosterone, Estrone, β-Estradiol, Tetrahydrocortisone, Tetrahydrocortisol, Cortisone and Cortisol. Before their analysis, the urine samples were prepared for their injection in the chromatograph through hydrolysis, extraction and derivation process, as described (27). The resulting mixture was transferred to a vial and was immediately encapsulated in order to avoid a possible reaction of these components to the humidity, until the analysis of the solution by gaseous chromatography took place, by injecting 1 μ L in the chromatograph. To carry out this analysis, an Agilent gas Chromatographer 6890N with mass detector 5973 Network and injector 7683 series, with a Factor Four Capillary Column VF-1ms, 25 m x 0.25mm internal diameter, was used. The analyses were

carried out under the following conditions: carrier gas He, flow rate 0.6 mL/min splitless injection system, solvent delay 16,90 min, injector temperature of 250 °C. The programmed temperatured was: 100 °C during 1.5 min; from 100 °C to 280 °C, 10 °C/min; from 280 °C to 300 °C, 10 °C/min; 300 °C for 2 min.

Statistical analysis.- The statistical software program SPSS 15.0 was used to analyze and process the data. Significant differences of the hormonal changes that took place before and after each trial were determined by the application of a oneway repeated measures general linear model. On the other hand, one-factor Anova was performed to compare the hormone changes occurring in the concentric and the concentric/eccentric trials. One-factor Anova was also used to compare the total workload, the torque peak average and the maximum torque reached in each trial. A significant level of 95% was required in all cases.

Results

There were no significant differences in the creatinine urinary values between samples before and after the trials in both the concentric trial (from 1.06 ± 0.24 to 1.12 ± 0.32 mg/ml) and the concentric/ eccentric trial (from 1.11 ± 0.18 to $1.23 \pm$ 0.21 mg/ml).

Table I shows the data referring to the total workload and the maximum and the average torque peaks for each leg in the two trials. It can be observed that the maximum torque peak, for the right and left quadriceps, was significantly higher (p<0.01) in the concentric/eccentric trial than in the concentric one.

Finally, Table II shows steroid levels measured after the completion of the con-

Table I. Comparison of isokinetic values (mean \pm SD) for each leg in the two trials.

Data	Concentric	Concentric/ Eccentric	
Total Wo	orkload (J)		
Right	6256.56±1152.1	6299.58±1172.9	
Left	6374.92±112	6387.47±1103.8	
Maximu	n Peak Torque (N•	m)	
Right	230.61±28.4	312.28±64.5**	
Left	226.48±28.4	295.13±56.3**	
Average	Peak Torque (N•m	า)	
Right	199.65±24.6	230.51±57.3	
Left	193.18±25.3	211.42±55.1	

**p < 0.01 vs. concentric trial. N=18.

centric and concentric/eccentric trials. No significant differences have been observed in any of them. Moreover, data of the concentric trial have been compared with those of the concentric/eccentric trial, but no significant changes have been observed.

Discussion

The creatinine excretion remained constant during the trial. Thus, it is known that creatinine excretion rates are fairly constant in absence of renal pathology, therefore this parameter may be used to determine the real urinary excretion of steroid metabolites (2).

After analyzing the isokinetic report, the maximum torque at 60°/s during the concentric/eccentric trial is higher than the one reached during the concentric trial. The eccentric strength can be 20-50% higher than the concentric strength (3) and therefore, its torque peak would be higher in the first one (7). However, both protocols registered similar values when the torque peak average was analyzed, which could show that the torque

Steroid	Concentric trial		Concentric/Eccentric	
	Before	After	Before	After
Androsterone	7163.65 (1865.8)	6602.70 (1639.72)	6710.55 (1324.8)	5904.28 (1734.74)
Etiocholanolone	5754.56 (1521.88)	4041.48 (1746.54)	3981.02 (1603.38)	2884.69 (1246.75)
DHEA	122.02 (125.73)	154.06 (205.81)	97.79 (117.13)	118.26 (143.18)
Androstenedione	75.40 (65.26)	74.55 (53.70)	40.03 (33.29)	21.26 (16.90)
Testosterone	259.12 (140.59)	224.35 (166.40)	225.41 (177.25)	214.74 (183.12)
Epitestosterone	213.43 (110.28)	190.09 (134.10)	175.68 (79.03)	163.08 (83.44)
Dehydrotestosterone	199.95 (103.30)	192.45 (130.72)	199.44 (78)	180.73 (165.05)
Estrone	24.17 (6.43)	26.09 (10.18)	22.07 (12.30)	20.03 (6.50)
β -estradiol	11.73 (1.81)	14.09 (10.61)	16.50 (13.38)	13.50 (5.39)
Tetrahydrocortisone	8754.51 (1387.80)	7469.83 (1206.98)	9382.07 (1632.82)	8356.40 (1968.59)
Tetrahydrocortisol	4107.28(1344.88)	4064.03 (1533.22)	4617.98 (1139.91)	4297.50 (1809.39)
Cortisone	413.39 (29.45)	344.15 (81.37)	478.61 (97.82)	410.25 (70.82)
Cortisol	480.21 (36.63)	409.00 (44.43)	700.63 (21.64)	430.80 (63.90)

Table II. *Urinary excretion of steroid hormones and their metabolites after the two trials.* Values are expressed as ng steroid/mg creatinine [Mean (SD)] n=18.

peak reached can not be maintained throughout the protocol due to the fatigue (5, 28).

Focusing on the hormonal changes, the urinary steroid profile has not changed significantly following the completion of both protocols, although maximum effort was required. Related studies indicate that the hormonal response appears with high strength training loads (16, 23). Different arguments may explain why the urinary steroid profile remained unchanged. The intensity of the workload, recovery periods and the technique are not as important as the volume of work in order to produce hormonal changes (6). In this study, the volume of the workload of the protocols were small. On the other hand, the hormonal response will depend on the size and the quantity of the activated muscular mass (17, 25). In this study, the protocols act on only one muscular chain (quadriceps). Finally, trained athletes show a higher androgenic response during the exercise in comparison to the untrained ones (1). The subjects under

study were university students who did not usually train, so it could have had some influence on the results.

When comparing the concentric and the concentric/eccentric trial, this data have shown that the contraction type has not been a deciding factor in order to alter the urinary steroid profile (androgens and corticosteroids) in strength short trials. The androgen response in this research is consistent with the results reflected in other studies that suggest that the concentric and eccentric contractions produce similar total and free testosterone responses to the same relative workload (15) as well as the same absolute workload (8). On the other hand, a research concluded that the concentric contraction produced higher stress and higher cortisol levels than the eccentric one (11). In that study, 4 sets of 12 repetitions at 80% of 1 RM were performed for four different exercises: bench press, leg extension, military press and leg curl; while in this study, only one quadriceps exercise for each leg was carried out.

For all these reasons, we conclude that significant differences do not exist in the urinary steroid profile after the completion of a short concentric and a concentric/eccentric trial with the same total workload. Factors such as the total workload and the number of muscular groups involved could be more decisive than the contraction type to produce hormonal changes in short trials. Moreover, one could conclude that an eccentric short training is a good method to increase the maximum strength peak without incurring high metabolic fatigue.

R. TIMON, G. OLCINA, P. TOMAS-CARUS, D. MUÑOZ, F. TORIBIO, A. RAI-MUNDO y M. MAYNAR. Perfil esteroideo urinario tras la realización de una sesión de ejercicio concéntrico y otra concéntrico/excéntrico con similar carga de trabajo total. J Physiol Biochem, 65 (2), 105-112, 2009.

El entrenamiento de fuerza de alta intensidad provoca variaciones en la concentración de esteroides. El tipo de contracción muscular, excéntrica o concéntrica, podría ser un factor que la alterase. El objetivo de este estudio fue comparar el efecto de la realización de una sesión corta de ejercicio concéntrico (CON) y otra concéntrica/excéntrica (CON/EXC), con la misma carga de trabajo total, sobre el perfil esteroideo urinario. 18 hombres realizaron dos sesiones de ejercicio de corta duración utilizando una máquina isocinética (BIODEX III) en días diferentes y trabajando los músculos cuádriceps de ambas piernas. La sesión de ejercicio 1 (CON) fue un 4 x 10 rep de extensión concéntrica de rodilla más relajación en el movimiento de flexión, a una velocidad de 60°/segundo y con una recuperación de 90 seg. entre cada serie y 4 minutos entre cada una de las piernas. La sesión de ejercicio 2 (CON/ EXC) fue un 4x 5 rep. de extensión concéntrica de rodilla más flexión excéntrica de rodilla, con las mismas condiciones de velocidad y recuperación. Muestras de orina fueron se tomaron antes del ejercicio y una hora después de finalizarlo. Los niveles urinarios (fracción

libre, glucoconjugada y sulfoconjugada) de Androsterona, Etiocolanolona, DHEA, Androstenodiona, Testosterona, Epitestosterona, Dehidrotestosterona, Estrona, β-estradiol, Tetrahidrocortisona, Tetrahidrocortisol, Cortisona v Cortisol, se determinaron usando técnicas de cromatografía de gases-espectrometría de masas. No se encontraron diferencias significativas en los valores de Trabajo Total y de Pico Torque Medio, aunque los valores de Pico Torque Máximo fueron más alto en el CON/EXC ejercicio que en el CON. Tampoco se observó ningún cambio en el perfil esteroideo urinario entre antes y después de las sesiones de ejercicio, o comparando las sesiones CON/EXC con la CON. Por tanto, los datos sugieren que las contracciones excéntricas no producen alteraciones hormonales diferentes a las producidas por las contracciones concéntricas, cuando se trata de sesiones de ejercicio de fuerza de corta duración con similar carga de trabajo total.

Palabras clave: Andrógenos, Corticosteroides, Carga de trabajo, Concéntrico, Excéntrico, Cromatografía de gases-espectrometría de masas.

References

- Ahtiainen, J.P., Pakarinen, A., Kraemer, W.J., Hakkinen, K. (2004): Acute hormonal responses to heavy resistance exercise in strength athletes versus nonathletes. *Can J Appl Physiol*, 29, 527-543.
- Bakonska-Pacon, E. (2006): Creatinine clearance and 24-hour creatinine excretion profile in yhe urine of people after physical exercises. *Biol Sport*, 23, 157-170.
- Bamman, M.M., Shipp, J.R., Jiang, J., Gower, B.A., Hunter, G.R., Goodman, A., Mclafferty, C.L., Urban, R.J. (2001): Mechanical load increases muscle IGF-1 and androgen receptor mRNA concentrations in humans. *Am J Physiol Endocrinol Metab*, 280, 383-390.
- Carrasco, D.I., Delp, M.D., Ray, C. A. (1999): Effect of concentric and eccentric muscle actions on muscle sympathetic nerve activity. *J Appl Physiol*, 86, 558-563.
- Crenshaw, A.G., Karlsson, S., Styf, J., Backlund, T., Friden, J. (1995): Knee extension torque and intramuscular pressure of the vastus lateralis

muscle during eccentric and concentric activities. *Eur J Appl Physiol*, **70**, 13-19.

- Crewther, B., Cronin, J., Keogh, J., Cook, C. (2008): The salivary testosterone and cortisol response to three loading schemes. *J Strength Cond Res*, 22, 250-255.
- Dudley, G.A., Tesch, P.A., Miller, B.J., Buchanan, P. (1991): Importance of eccentric actions in performance adaptations to resistance training. *Aviat Space Environ Med*, 62, 543-550.
- Durand, R.J., Castracane, V.J., Hollander, D.B., Tryniecki, J.L., Bamman, M.M., O'neal, S., Hebert, E.P., Kraemer, R.R. (2003): Hormonal responses from concentric and eccentric muscle contractions. *Med Sci Sports Exerc*, **35**, 937-943.
- Enoka, R.M. (1996): Eccentric contractions require unique activation strategies by the nervous system. J Appl Physiol, 81, 2339-2346.
- Heinegard, D., Tiderstrom, G. (1973): Determination of serum creatinine by a direct colorimetric method. *Clin Chem Acta*, 43, 305.
- Hollander, D.B., Durand, R.J., Trynicki, J.L., Larock, D., Castracane, V. D., Hebert, E. P., Kraemer, R. R. (2003): RPE, pain, and physiological adjustment to concentric and eccentric contractions. *Med Sci Sports Exerc*, 35,1017-1025.
- Hortobabagyi, T., Devita, P., Money, J., Barrier, J. (2000): Effects of standard and eccentric overload strength training in young women. *Med Sci Sports Exerc*, 33, 1206-1212.
- Izquierdo, M., Ibañez, J., González-Badillo, J. J., Hakkinen, K., Ratamess, N.A., Kraemer, W.J., French, D.N., Eslava, J., Altadill, A., Asiain, X., Gorostiaga, E.M. (2006): Differential effects of strength training leading to failure versus not to failure on hormonal responses, strength, and muscle power gains. *J Appl Physiol*, 100, 1647-1656.
- Jerjes, W.K., Peters, T.J., Taylor, N.E., Wood, P.J., Wessely, S., Cleare, A.J. (2006): Diurnal excretion of urinary cortisol, cortisone, and cortisol metabolites in chronic fatigue syndrome. *J Phychosom Res*, 60, 145-153.
- Kraemer, R.R., Hollander, D.B., Reeves, G.V., Francois, M., Ramadan, Z.G., Meeker, B., Tryniecki, J.L., Hebert, E.P., Castracane, V.D. (2006): Similar hormonal responses to concentric and eccentric muscle actions using relative loading. *Eur J Appl Physiol*, **96**, 551-557.
- Kraemer, R.R., Kilgore, J.L., Kraemer, G.R., Castracane, V.D. (1992): Growth hormone, IGF-1 and testosterone responses to resistive exercise. *Med Sci Sports Exerc*, 24, 1346-1352.

- Kraemer, W.J., Dudley, G.A., Tesch, P.A., Gordon, S.E., Hather, B.M., Volek, J.S., Ratamess, N.A. (2001): The influence of muscle action on the acute growth hormone response to resistance exercise and short-term detraining. *Growth Horm IGF Res*, 11, 75-83.
- Kraemer, W.J., Gordon, S.E., Fleck, S.J., Marchitelli, L.J., Mello, R., Dziados, J.E., Friedl, K., Harman, E., Maresh, C., Fry, A.C. (1991): Endogenous anabolic hormonal and growth factor responses to heavy resistance exercise in males and females. *Int J Sports Med*, 12, 228-235.
- Lowe, D.A., Warren, G.L., Ingalss, C.P., Boorstein, D.B., Armstrong, R.B. (1995): Muscle function and protein metabolism after iniation of eccentric contraction induced injury. *J Appl Physiol*, 79, 1260-1270.
- Ly, L.P., Handelsman, D.J. (2002) Muscle strength and ageing: methodological aspects of isokinetic dynamometry and androgen administration. *Clin Exp Pharmacol Physiol*, **29**, 37-47.
- Marx, J.O., Ratamess, N.A., Nindl, B.C., Gotshalk, L.A., Volek, J.S., Dohi, K., Bus, J.A., Gómez, A.L., Mazzetti, S.A., Fleck, S.J., Hakkinen, K., Newton, R.U., Kraemer, W.J. (2001): Low volume circuit versus high-volume periodized resistance training in women. Med Sci Sports Exerc, 33, 635-643.
- Nishikaze, O., Furuya, E. (1998): Stress and anticortisols-17-ketosteroid sulfate conjugate as a biomarker in tissue repair and recovery. J UOEH, 20, 273-295.
- Schwab, R., Johnson, G.O., Housh, T.J., Kinder, J.E., Weir, J.P. (1993): Acute effects of different intensities of weighlifting on serum testosterone. *Med Sci Sports Exer*, 25, 1381-1385.
- Smilios, I., Pilianidis, T., Karamouzis, M., Tokmakidis, S. P. (2003): Hormonal responses alter various resistance exercise protocols. *Med Sci Sports Exerc*, 35, 644-654.
- Stokes, K.A., Nevill, M.E., Hall, G.M., Lakomy, H.K. (2002): Growth hormone responses to repeated maximal cycle ergometer exercise at different pedalling rates. *J Appl Physiol*, 92, 602-608.
- 26. Timon-Andrada, R., Maynar-Marino, M., Munoz-Marín, D., Olcina-Camacho, G.J., Caballero, M.J., Maynar-Marino, J.I. (2007): Variations in urine excretion of steroid hormones after an acute session and after a 4-week programme of strength training. *Eur J Appl Physiol*, **99**, 65-71.
- Timon, R., Olcina, G., Muñoz, D., Maynar, J. I., Caballero, M. J., Maynar, M. (2008): Determination of urine steroid profile in untrained men to

evaluate recovery after a strength training session. J Strength Cond Res, 22,1087-1093.

- Yeung, S.S., Yeung, E.W. (2008): Shift of peal torque angle after eccentric exercise. Int J Sports Med, 29, 251-256.
- 29. Zhou, Z.H., Liu, J.H., Jin, Y.L., He, P., Zhao, P.L., Wang, X.M. (2000): Effects of different loading exercises on endogenous sex hormones and their metabolites. *Hunan Yi Ke Da Xue Bao*, 25, 23-26.