

UNIVERSIDAD PABLO DE OLAVIDE

PROGRAMA DE DOCTORADO EN CIENCIAS DE LA ACTIVIDAD FÍSICA Y DEL DEPORTE

LA TENSIOMIOGRAFÍA COMO HERRAMIENTA PARA EVALUAR LAS PROPIEDADES MECÁNICAS MUSCULARES ANTE DIFERENTES ENTORNOS DE RENDIMIENTO FÍSICO Y DEPORTIVO

> D. FRANCISCO JOSÉ PIQUERAS SANCHIZ 2022



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Tesis para optar al grado de doctor internacional presentada por: D. Francisco José Piqueras Sanchiz

> Directores: Dr. Óscar García García Dr. Fernando Pareja Blanco Sevilla, 2022



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DOCTORANDO

Francisco José Piqueras Sanchiz

Óscar García García Fernando H Pareja Blanco

DIRECTORES

A mi mujer, Por acompañarme en todo este viaje

DOCUMENTO ESTANCIA INTERNACIONAL



UNIVERSIDADE DA MAIA - ISMAI 25/11/2021

TO WHOM IT MAY CONCERN

Francisco Piqueras Sanchiz successfully completed 3 months research work in our Research Center in Sports Sciences, Health Sciences and Human Development, at the University of Maia, Maia, Portugal, where, under the supervision of Dr Fábio Nakamura, he has participated in:

- Research projects in a professional soccer club related to neuromuscular training and GPS.

- Assist in the day-to-day running of the study.

In addition, we have discussed and shared views about other potential projects of collaboration we have in common, which we will seek to develop in the future.

Furthermore, Francisco Piqueras Sanchiz has demonstrated a high level of scientific knowledge combined with an extensive practical experience with important applied aspects related to the physiology and biomechanics of sports and exercise.

Sincerely,

Josepus V.ma

Dr João Viana, PhD CIDESD Vice-Director CIDESD-ISMAI Director PhD in Sports Sciences Course Coordinator

Research Center in Sports Sciences, Health Sciences and Human Development - CIDESD University of Maia - ISMAI

Av. Carlos Oliveira Campos - Castelo da Maia, 4475-690 Maia Portugal

Email: jviana@ismai.pt Office: +351 22 986 60 70

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ÍNDICE

1.	PUBLICACIONES Y DIVULGACIÓN DE LOS RESULTADOS	14
2.	RESUMEN	16
3.	GLOSARIO DE ACRÓNIMOS EMPLEADOS	23
4.	ÍNDICE DE TABLAS	27
5.	ÍNDICE DE FIGURAS	31
6.	INTRODUCCIÓN	37
7.	ESTADO ACTUAL DEL CONOCIMIENTO	
7	.1 Parámetros tensiomiográficos	41
	7.1.1 Deformación radial máxima (Dm)	
	7.1.2 Tiempo de contracción (Tc)	
7	'.2 Fiabilidad	50
7	.3 Protocolo de medición de tensiomiografía	53
	7.3.1.1 Localización del músculo	53
	7.3.1.2 Distancia entre electrodos	55
	7.4.1.1 Tipo de parche y duración del estímulo	56
	7.4.1.2 Experiencia del evaluador	56
7	7.5 Protocolo de medición	57
	7.5.1 Fatiga muscular	58
	7.5.1.1 Fatiga inducida por entrenamiento de resistencia	58
	7.5.1.2 Fatiga inducida por entrenamiento de fuerza	60
	7.5.2 Daño muscular	63
	7.5.3 Relación entre parámetros tensiomiográficos y rendimiento	65
	7.5.4 Valores de referencia de los distintos parámetros tensiomiográficos e modalidades deportivas, así como adaptaciones de estos ante diferentes interv	n diferentes zenciones de
	entrenamiento	67
	7.5.5 Recuperación	68
8. F	PLANTEAMIENTO DEL PROBLEMA	74
9. H	HPÓTESIS y OBJETIVOS	79
10.	ESTUDIO I	83
11.	ESTUDIO II	98
12.	ESTUDIO III	117
13.	PERSPECTIVAS DERIVADAS DE LOS TRABAJOS DE LA PRESENTE TES	IS
DO	CTORAL	
14.	CONCLUSIONES	139
15.	APLICACIONES PRÁCTICAS	144

16. LIMITACIONES DE LOS ESTUDIOS	146
17. FUTURAS LÍNEAS DE INVESTIGACIÓN	149
18. REFERENCIAS	153
ANEXO I CONSENTIMIENTO INFORMADO	166
ANEXO II PUBLICACIÓN CIENTÍFICA Y DIVULGACIÓN DE RESULTADOS	168

1. PUBLICACIONES Y DIVULGACIÓN DE LOS RESULTADOS

La Tesis Doctoral que aquí se presenta es un compendio de tres publicaciones.

- Artículo 1 Piqueras-Sanchiz, F., Martín-Rodríguez, S., Pareja-Blanco, F., Baraja-Vegas, L., Blázquez-Fernández, J., Bautista, I. J., & García-García, Ó. Mechanomyographic Measures of Muscle Contractile Properties are Influenced by Electrode Size and Stimulation Pulse Duration. (2020). *Sci Rep.* 18;10(1):8192. doi: 10.1038/s41598-020-65111-z.
- Artículo 2 Piqueras-Sanchiz, F., Cornejo-Daza, P. J., Sánchez-Valdepeñas, J., Bachero-Mena, B., Sánchez-Moreno, M., Martín-Rodríguez, S., García-García, Ó., & Pareja-Blanco, F. Acute Mechanical, Neuromuscular, and Metabolic Responses to Different Set Configurations in Resistance Training. (2021). *J Strength Cond Res*. Epub ahead of print. 2021 Jun 15. doi: 10.1519/JSC.000000000004068.
- Artículo 3 Piqueras-Sanchiz, F., Martínez-Aranda, L. M., Pareja-Blanco, F., Rodríguez-Ruiz, D., & García-García, Ó. Evolution of contractile properties of the lower limb muscles throughout a season in elite futsal players. (2020). J Sports Med Phys Fitness. 60(7):965-973. doi: 10.23736/S0022-4707.20.10345-1.

Los artículos están incluidos en los anexos y ya han sido publicados en revistas incluidas en la lista de Factor de Impacto, *Journal Citation Reports* (JCR).

Comunicaciones orales en Congresos Internacionales:

- Francisco Piqueras-Sanchiz, Fernando Pareja-Blanco, Saúl Martín-Rodríguez, Óscar García-García. (2019). Lower stimulus lengths increase measurement error between 10-18%: methodological implications for tensiomyographic measurements. (1st international sport forum on strength, training and nutrition, Madrid). Oral communication.
- **Francisco Piqueras-Sanchiz**, Fernando Pareja-Blanco, Óscar García-García. (2019). Acute metabolic, neuromuscular and mechanical response of squat exercise protocols that differ in set configuration. (1st International Sport Forum on Strength, Training and Nutrition, Madrid). Oral communication.
- Francisco Piqueras-Sanchiz, Óscar García-García, Juan Sánchez-Valdepeñas Pedro Cornejo-Daza, Miguel Sánchez-Moreno, Beatriz Bachero-Mena, Fernando Pareja-Blanco. (2021). Relationships between changes in muscle contractile properties and muscle hypertrophy after a strength training program. (2nd International Conference on Technology in Physical Activity and Sport, Sevilla). Oral communication.
- Francisco Piqueras-Sanchiz, Fernando Pareja-Blanco, Saúl Martín Rodríguez, Óscar García-García (2022). Is radial displacement velocity at 90% sensitive enough to discriminate between fatigue and non-fatigue state after strenuous exercise? (VI Congreso Internacional Optimización del Entrenamiento y Readaptación Físico– Deportiva, Sevilla). Oral communication.

2. RESUMEN

ESTUDIO I: Influencia del tamaño del electrodo y la duración del pulso de estimulación sobre las propiedades contráctiles del músculo medidas con tensiomiografía.

Resumen

Este estudio tuvo como objetivo determinar los efectos de la duración del pulso y el tamaño del electrodo en las propiedades contráctiles del músculo. En el estudio participaron 36 hombres jóvenes y sanos (edad 24.8 \pm 5.8 años; altura 1.78 \pm 0.06 m; masa corporal 71.8 \pm 7.3 kg; realizando ejercicio físico 3.5 ± 1.2 h·semana⁻¹). Se utilizó la tensiomografía (TMG) para evaluar las propiedades mecánicas de los músculos recto femoral (RF) y vasto medial (VM) de la pierna dominante. Además, se tuvo en cuenta el tamaño de los electrodos (3.2-5 cm) y la longitud del estímulo (0.2, 0.5 y 1 ms) en las mediciones. Los parámetros incluidos fueron: desplazamiento radial máximo (Dm), tiempo de contracción (Tc), tiempo de retardo (Td), tiempo sostenido (Ts) y tiempo de relajación media (Tr). Se cuantificó la fiabilidad relativa y absoluta. Para analizar los efectos del electrodo y la longitud del estímulo, se utilizó un análisis de varianza de medidas repetidas. Los parámetros Dm y Tc mostraron para ambos músculos una excelente fiabilidad relativa (0.95-0.99) y absoluta (1.6-4.2%). Sin embargo, Ts y Tr mostraron valores bajos de fiabilidad absoluta (4.4-40.9%). En relación al Td, la fiabilidad relativa es (0.85-0.88) y la absoluta (1.6-2.7%). La longitud del estímulo aplicado y el tamaño de los electrodos influyeron significativamente en las propiedades contráctiles del músculo (p < 0.05; $\eta^2_{p} = 0.09$ -0.60). La Dm aumentó significativamente tanto con el incremento de la duración del estímulo como con el tamaño del electrodo en ambos músculos. Sin embargo, la Tc y la Td se vieron menos afectadas por ambas condiciones. En conclusión, tanto la duración del estímulo como la dimensión del electrodo influyen en la respuesta muscular obtenida. Además, los parámetros Dm y Tc mostraron una alta fiabilidad, mientras que lo contrario se observó para Ts y Tr. Como aplicación práctica, se sugiere que es necesaria una duración del pulso del estímulo de 1 ms junto con un electrodo de 5×5 cm para alcanzar una evaluación fiable y reproducible de las propiedades contráctiles de los músculos RF y VM.

STUDY I: Mechanomyographic measures of muscle contractile properties are influenced by electrode size and stimulation pulse duration.

Abstract

The aim of this study was to determine the effects of changing pulse duration and electrode size on muscle contractile properties. Thirty-six healthy young males participated in the study (age 24.8 \pm 5.8 years; height 178.2 \pm 0.6 cm; body mass 71.8 \pm 7.3 kg; self-reported weekly moderate intensity activity 3.5 ± 1.2 h·week⁻¹). Tensiomyography was used to assess rectus femoris (RF) and vastus medialis (VM) muscles' neuromuscular properties according to the electrode size (3.2-5 cm) and the stimulus length (0.2, 0.5, and 1 ms). Maximal radial displacement (Dm); contraction time (Tc); delay time (Td); sustained time (Ts) and half relaxation time (Tr) were measured. Only the dominant leg was examined. Relative and absolute reliability was quantified. To analyze the effects of the electrode and the stimulus length, a repeated-measures analysis of variance was used. Dm and Tc parameters showed for both muscles an excellent relative (0.95-0.99) and absolute (1.6-4.2%) reliability. However, Ts and Tr showed low values of absolute reliability (4.4-40.9%). With regards to Td, the relative reliability is (0.85-0.88) and the absolute reliability (1.6-2.7%). The duration of the stimulus length applied to the RF and VM and electrode size significantly influenced muscles' contractile properties (p < 0.05; $\eta^2_p = 0.09$ -0.60). The Dm increased substantially as the duration of the stimulus increased and with the use of larger electrodes in both muscles. However, Tc and Td were less affected by both conditions and their behaviour was not entirely clear. Practically, our study suggests that a stimulus pulse duration of 1 ms together with a 5 imes5 cm electrode is necessary to reach a reliable assessment of both RF and VM muscles contractile properties.

ESTUDIO II: Respuestas mecánicas, neuromusculares y metabólicas agudas ante diferentes configuraciones de la serie en el entrenamiento de fuerza.

Resumen

El objetivo de este estudio fue investigar el efecto de la configuración de la serie durante el entrenamiento de fuerza sobre la actividad neuromuscular, la respuesta metabólica y las propiedades contráctiles del músculo. Dieciséis hombres entrenados en fuerza realizaron 2 sesiones de entrenamiento en el ejercicio de sentadilla que consistieron en: (a) 3 series de 8 repeticiones con 5 minutos de descanso entre series (3×8) y (b) 6 series de 4 repeticiones con 2 minutos de descanso entre series (6×4). La intensidad del entrenamiento (75% de una repetición máxima [1RM]), el volumen total (24 repeticiones), el descanso total (10 minutos) y la densidad de entrenamiento se igualaron entre los protocolos. Se realizó una batería de pruebas antes y después de cada protocolo: (a) TMG, (b) concentración de lactato y amonio en sangre, (c) salto con contramovimiento (CMJ), y (d) contracción isométrica voluntaria máxima (MVIC) en el ejercicio de sentadilla. Se registraron los valores de fuerza, velocidad y potencia, junto con los datos electromiográficos, para cada repetición a lo largo de cada protocolo. El protocolo 6×4 resultó en mayor rendimiento mecánico (es decir, mayor fuerza, velocidad y potencia) y en marcadores neuromusculares de fatiga más bajos (es decir, menor actividad electromiográfica y mayor frecuencia mediana) durante el ejercicio en comparación con el protocolo 3×8 , especialmente en las últimas repeticiones de cada serie. El protocolo 3×8 indujo mayores concentraciones de lactato y amonio, mayores reducciones en la altura del salto, así como mayores deterioros en la velocidad de deformación derivada de la TMG después del ejercicio que el 6×4 . Por lo tanto, la implementación de series de menos repeticiones con intervalos de descanso más cortos y frecuentes entre series atenúa los deterioros en el rendimiento mecánico, especialmente en las últimas repeticiones de cada serie. Estos efectos pueden estar mediados por menores alteraciones neuromusculares, menor estrés metabólico y mejor mantenimiento de las propiedades contráctiles del músculo.

STUDY II: Acute mechanical, neuromuscular and metabolic responses to different set configurations in resistance training.

Abstract

The aim of this study was to investigate the effect of set configuration on mechanical performance, neuromuscular activity, metabolic response, and muscle contractile properties. Sixteen strength-trained men performed 2 training sessions in the squat exercise consisting of (a) 3 sets of 8 repetitions with 5 minutes rest between sets (3×8) and (b) 6 sets of 4 repetitions with 2 minutes rest between sets (6×4) . Training intensity (75% one repetition maximum), total volume (24 repetitions), total rest (10 minutes), and training density were equalized between protocols. A battery of tests was performed before and after each protocol: (a) TMG, (b) blood lactate and ammonia concentration, (c) countermovement jump, and (d) maximal voluntary isometric contraction in the squat exercise. Force, velocity, and power output values, along with electromyography data, were recorded for every repetition throughout each protocol. The 6×4 protocol resulted in greater mechanical performance (i.e., higher force, velocity, and power) and lower neuromuscular markers of fatigue (i.e., lower root mean square and higher median frequency) during the exercise compared with 3×8 , particularly for the last repetitions of each set. The 3×8 protocol induced greater lactate and ammonia concentrations, greater reductions in jump height, and greater impairments in TMG-derived velocity of deformation after exercise than 6×4 . Therefore, implementing lower-repetition sets with shorter and more frequent interset rest intervals attenuates impairments in mechanical performance, especially in the final repetitions of each set. These effects may be mediated by lower neuromuscular alterations, reduced metabolic stress, and better maintained muscle contractile properties.

ESTUDIO III: Evolución de las propiedades contráctiles de los músculos de las extremidades inferiores a lo largo de una temporada en jugadores de fútbol sala de élite.

Resumen

Este estudio tuvo como objetivo analizar los cambios producidos durante la temporada, así como determinar el potencial efecto acumulativo de un programa de entrenamiento de fuerza en jugadores de fútbol sala sobre las propiedades contráctiles de la musculatura de las piernas. Catorce jugadores de fútbol sala de élite (2 porteros, 4 defensas, 4 extremos y 3 pivotes) fueron evaluados mediante TMG en las semanas 11 (Pre-), 18 (Mid-) y 28 (Post-) de la temporada. Se evaluó el Dm, Tc, Td y la velocidad de desplazamiento radial (90%) Dm (VrD₉₀). Después de la segunda medición, se incluyó un programa de entrenamiento de fuerza basado en la musculatura del tren inferior. Se llevó a cabo una vez por semana durante 9 semanas. Para el estudio, se utilizó un análisis de varianza de medidas repetidas para detectar los cambios a lo largo de la temporada. Se incluyeron dos factores: el tiempo, que se utilizó como factor intra-sujeto y la posición específica, que se utilizó como factor entre-sujetos. De esta manera, se observó un incremento respecto a los niveles basales para Tc, Td y Dm durante la temporada. Sin embargo, las adaptaciones de las propiedades contráctiles fueron específicas de cada músculo. Además, un programa de fuerza durante la temporada de 9 semanas de duración con una sesión por semana no tuvo efectos significativos sobre las propiedades contráctiles de los jugadores de fútbol sala. Por último, no se observaron diferencias al comparar las distintas posiciones. En vista de los resultados, durante la temporada, se produce un deterioro de las propiedades mecánicas musculares de las piernas en jugadores profesionales de fútbol sala. Además, un entrenamiento de fuerza, realizado una vez por semana en la fase final de la temporada, no fue suficiente para revertir estos deterioros, pero al menos permitió que no se deterioraran más dichas propiedades musculares.

STUDY III: Evolution of contractile properties of the lower limb muscles throughout a season in elite futsal players.

Abstract

This study aimed to analyze the changes in the contractile properties produced during the season, as well as to determine the potential cumulative effect of a resistance training (RT) program in futsal players. Fourteen elite futsal players (2 goalkeepers, 4 defenders, 4 wingers and 3 pivots) were assessed by TMG at 11th, 18th, and 28th week of the season. The Dm; Tc; Td, and radial displacement velocity (90%) Dm (VrD₉₀) were assessed. After the second measurement, a RT program was included in the regular training sessions and focused on the lower body musculature. It was performed once per week for 9 weeks. Repeated measures analysis of variance was used to detect in-season changes. Two factors were included: Time was used as the within-subject factor and the specific position was used as the between-subject factor. It was observed an increment in Tc for several muscles throughout the season: biceps femoris (BF; p = 0.02), semitendinosus (ST; p = 0.04), adductor longus (AL; p = 0.008) and gastrocnemius medialis (GM; p=0.009). Similarly, there were significant increments in Dm for GM (p = 0.02) and AL (p = 0.05), as well as increases in Td for BF (p = 0.002). Moreover, no significant changes in VrD₉₀ between time points 2-3 (analysis of RT effect) were observed. Additionally, the players' positions reported no significant changes for any of the variables analyzed. Taken together, an increase respect to baseline levels was observed for Tc, Td and Dm during the season. However, the adaptations to contractile properties were muscle specific. In addition, an in-season 9-week RT program (1-weekly) had no significant effects on the contractile properties of futsal players. Lastly, there were no differences when comparing different positions.

GLOSARIO DE ACRÓNIMOS EMPLEADOS

3. GLOSARIO DE ACRÓNIMOS EMPLEADOS

ANOVA: análisis de la varianza **BB**: bíceps braquial BF: bíceps femoral **BR**: músculo braquial CK: creatina kinasa CMJ: salto con contramovimiento **CWI**: cold water inmersión CV: coeficiente de variación Dm: Desplazamiento máximo radial **DJ**: *drop jump* DOMS: dolor muscular de aparición retardada **DEP**: distancia entre parches **DSI**: índice de Dimitrov **ED**: extensor digital EIMD: daños musculares inducidos por el ejercicio EMG: electromiografía **ER**: erector espinal ES: tamaño del efecto FDS: flexor superficial del dedo GC: grupo control GEE: grupo ejercicio excéntrico GICAF: grupo de inmersión continua en agua fría GIIAF: grupo de inmersión intermitente en agua fría **GM**: gastronemio medial GPI + FE: grupo acondicionamiento isquémico más fase excéntrica GPS: grupo peso muerto GS: grupo sentadilla **GHT**: grupo *hip thrust* IC: intervalo de confianza ICC: índice de correlación intraclase

MDC: mínimo cambio detectable

MDF: frecuencia mediana MHC-I: cadena pesada de miosina de tipo I MMG: mecanomiografía MNF: frecuencia media MSE: es el término medio de error cuadrado de las medidas repetidas ANOVA MVIC: máxima contracción isométrica voluntaria MUAP: potencial de acción de la unidad motora **RF**: recto femoral **RMS**: root mean square **ROM**: rango de movimiento SD: desviación estándar SEM: error estándar de la medida sEMG: EMG de superficie SNG: Sonomiografía **SJ**: squat jump SOL: sóleo TA: tibial anterior TB: triceps braquial Tc: tiempo de contracción Td: tiempo de retardo **TENS**: estimulación nerviosa eléctrica transcutánea TMG: tensiomiografía Tr: tiempo de relajación Ts: tiempo de contracción sostenida **UM**: unidad motora V10: Velocidad del 10% del Dm V90: Velocidad del 90% del Dm Vc: Velocidad de contracción Vd: velocidad de deformación **VIFT**: velocidad final del test intermitente de 30-15 VL: vasto lateral VM: vasto medial VMG: vibromiografía VML: vasto medial largo

VMO: vasto medial oblicuo
Vrn: Velocidad de respuesta normalizada
VO₂max: consumo máximo de oxígeno
WBV: whole body vibration

ÍNDICE TABLAS

4. ÍNDICE DE TABLAS

Tabla 1. Desplazamiento radial máximo (Dm) antes (Pre) y después (Post) de 35 días dedescanso en cama. Tomado de Pisot et al. (2008)

Tabla 2. Cambios en el grosor muscular y propiedades contráctiles de los músculos del cuádriceps en respuesta a 6 semanas de entrenamiento de fuerza de la parte inferior del cuerpo.

 Adaptado de Wilson et al. (2019)

Tabla 3. Índice de correlación intraclase (ICC) de los principales parámetros tensiomiográficos. Desplazamiento radial máximo (Dm); tiempo de contracción (Tc); tiempo de retardo (Td). Tabla elaborada a partir de Rodríguez et al. (2017a) y Macgregor et al. (2018)

Tabla 4. Superficie (en cm²) para el 3%, 5% y 10% del error relativo según el punto de medición de referencia. El error relativo se presenta para el tiempo de contracción y el desplazamiento radial máximo, por separado. Adaptado de Simunic et al. (2019)

Tabla 5. Medidas obtenidas en el bíceps femoral en 3 ángulos diferentes. Tomado de Ditroillo et al. (2011)

Tabla 6. Intervalo de confianza al 95% y cálculo bayesiano e interpretación de los resultados del autor con un 80% de probabilidad previa. Adaptado de Martín-Rodríguez, et al. 2017)

Tabla 7. Propiedades contráctiles del músculo: valores descriptivos antes (Pre) y después (Post) del ejercicio de fatiga para ambos tipos de contracciones musculares. Adaptado de Muñoz-López et al. (2020)

Tabla 8. Respuesta de los parámetros de tensiomografía tras pruebas de resistencia o fuerza

 para medir la fatiga inducida por dichos protocolos.

Tabla 9. Diferencias entre la primera y la segunda medición Dm (desplazamiento radial máximo); Tc (tiempo de contracción). Pre: antes de empezar el estudio; Post: tras cumplir el periodo de 10 semanas. Tomado de García-García et al. (2016)

Tabla 10. Comparación del comportamiento de las variables de tensiomiografía evaluadas en el recto femoral entre los grupos según el momento de medición. Adaptado de Sánchez-Ureña et al., (2018)

Tabla 11. Comparación del comportamiento de las variables de tensiomiografía evaluadas en el bíceps femoral entre los grupos según el momento de medición. Adaptado de Sánchez-Ureña et al., (2018)

Tabla 12. Sujeto bloqueado. A: 0.2 milisegundos (ms); B: 0.5 ms; C: 1 ms.

Tabla 13. Fiabilidad de los parámetros tensiomiográficos del músculo Vasto Medial (n = 36).

Tabla 14. Fiabilidad de los parámetros tensiomiográficos del músculo Recto Femoral (n = 36).

Tabla 15. Efectos de la longitud del estímulo y del tamaño de los electrodos sobre diferentes parámetros tensimiográficos.

Tabla 16. Valores medios del rendimiento mecánico y de los parámetros neuromusculares alcanzados durante la totalidad de los protocolos (24 repeticiones).

Tabla 17. Efectos de diferentes protocolos de entrenamiento de fuerza sobre las propiedades contráctiles musculares evaluadas con tensiomiografía.

Tabla 18. Respuesta mecánica, neuromuscular y metabólica ante diferentes protocolos de ejercicios de fuerza.

Tabla 19. Distribución de las cargas de entrenamiento y competición en un microciclo tipo.

Tabla 20. Evolución de las propiedades contráctiles a lo largo de una temporada y tras un programa de entrenamiento de fuerza en jugadores de fútbol sala de élite.

ÍNDICE DE FIGURAS

5. ÍNDICE DE FIGURAS

Figura 1. Ilustración del equipo de tensiomografía (TMG) y la clásica onda de respuesta de tic con todos sus parámetros. 1 = Sensor mecánico y electrodos. 2 = Trípode con mano manipuladora. 3 = Unidad de adquisición de datos. 4 = Onda típica extraída de la respuesta de la TMG. Tomado de Martín-Rodríguez et al. (2017a)

Figura 2. Relación entre los cambios de grosor muscular y los cambios en el desplazamiento radial máximo (Dm) para el músculo gastronemio medial. * p<0.01. Tomada de Pisot et al. (2008)

Figura 3. Correlación entre los cambios observados en el desplazamiento radial máximo (Dm) en el bíceps femoral (BF) y los cambios en el grosor muscular de este músculo (C); correlación entre los cambios en el Dm y los cambios en el ángulo de penneación en el vasto medial oblicuo (VMO) (D). Tomado de Simunic et al. (2019)

Figura 4. Cronograma de intensidad y ejercicios. Tomado de Wilson et al. (2019)

Figura 5. Valor medio e intervalos de confianza (95%) de la relación entre el porcentaje de fibras musculares de tipo 1 y tiempos de contracción de la respuesta del vientre muscular a la estimulación de las contracciones en biceps braquial (BB), triceps braquial (TB), flexor superficial del dedo (FDS), extensor digital (ED), biceps femoral (BF), tibial anterior (TA) gastronemio medial (GCM), sóleo (SOL), y músculo braquial (BR), de los sitios a) superficiales y b) profundos. Adaptado de Dahmane et al. (2001)

Figura 6. Modelos de regresión univariante entre el tiempo de contracción medido por TMG y el porcentaje de fibras tipo I, evaluadas por el método de cadena pesada de miosina (% MHC-I). Tomado de Simunic et al. (2011)

Figura 7. Fiabilidad relativa dentro de la prueba y del test con intervalos de confianza (CI) del 95% para el desplazamiento radial máximo (Dm). Tomado de Lohr et al. (2019)

Figura 8. Fiabilidad relativa dentro de la prueba y del test con intervalos de confianza (CI) del 95% para el tiempo de contracción (Tc). Tomado de Lohr et al. (2019)

Figura 9. Fiabilidad relativa dentro de la prueba y del test con intervalos de confianza (CI) del 95% para el tiempo de retardo (Td). Tomado de Lohr et al. (2019)

Figura 10. Tres sensores de tensiomografía que se colocaron paralelos para la realización del estudio (B); los veintisiete (3 x 9) puntos de medición en cada músculo (A). La distancia lateral y longitudinal entre los puntos de medición vecinos era de 1.5 cm. Tomado de Simunic et al. (2019)

Figura 11. Puntos de medición en biceps braquial (A), vasto lateral (B), vasto medial (C), recto femoral (D), y erector espinal (E). Tomado de Simunic et al. (2019)

Figura 12. Desplazamiento radial máximo (Dm) medido mediante tensiomiografía a cuatro distancias entre electrodos. Se presentan los datos medios e individuales de cada uno de los 10 participantes. Diferencia significativa entre los grupos, p<0.05. Tomado de Wilson et al. (2019)

Figura 13. Típica progresión de la curva de desplazamiento. Tomado de Macgregor et al. (2018)

Figura 14. A: Almohadilla triangular TMG, 139°; B: Almohadilla triangular utilizada estudio Martín-San Agustín et al. (2020)

Figura 15. Desplazamiento radial máximo (Dm) y tiempo de contracción (Tc) en el bíceps braquial a lo largo del estudio. Tomado de Hunter et al. (2012). Se observaron efectos significativos de (p<0.001) los efectos de grupo, tiempo e interacción se observaron para todos los intervalos de tiempo. /**p<0.001. ^tp<0.001 significativamente menor que la medida previa al daño; **p<0.001 [¥]p<0.001 significativamente mayor que la medida previa al daño.

Figura 16. Parámetros TMG extraídos de la curva desplazamiento-tiempo en el Vasto Medial.

Figura 17. Curva desplazamiento-tiempo del Vasto Medial de la pierna dominante, de acuerdo al tamaño de los electrodos (3.2–5 cm) y a la duración del estímulo (0.2, 0.5, y 1 ms).

Figura 18. Curva desplazamiento-tiempo del Recto Femoral de la pierna dominante, de acuerdo al tamaño de los electrodos (3.2–5 cm) y a la duración del estímulo (0.2, 0.5, y 1 ms).

Figura 19. Representación sistemática del diseño de estudio incluyendo los 2 protocolos de fuerza analizados y los tests empleados antes y después de los protocolos

Figura 20. Evolución del rendimiento mecánico, a través de los valores propulsivos de A) fuerza (MPF), B) velocidad (MPV), y C) potencia (MPP) a lo largo de las 24 repeticiones en ambos protocolos. Los datos hacen referencia a la media \pm SD, n = 16. 3x8: protocolo que consistía en 3 series de 8 repeticiones al 75% de 1RM; 6x4: protocolo que consistía en 6 series

de 4 repeticiones al 75% de 1RM. Diferencias significativas intra-protocolo con respecto a las primeras repeticiones: *p < 0.05. Diferencias significativas inter-protocolo en cada repetición: *p < 0.05.

Figura 21. Evolución de los parámetros neuromusculares a lo largo de las 24 repeticiones. A) Media del "*root mean square*" de los músculos Vasto Medial y Vasto Lateral; and B) Frecuencia mediana de los músculos Vasto Medial y Vasto Lateral. Los datos se expresan como media \pm SD, n = 16. 3x8: protocolo que consistía en 3 series de 8 repeticiones 75% de 1RM; 6x4: protocolo que consistía en 6 series de 4 repeticiones 75% de 1RM. Los datos se normalizaron con respecto a la primera repetición de cada protocolo de fuerza. Diferencias significativas dentro del protocolo con respecto a la primera repetición: *p < 0.05. Diferencias significativas entre los protocolos en la repetición correspondiente: #p < 0.05.

Figura 22. Diseño experimental y tests realizados

Figura 23. Ejercicios realizados durante el programa y cargas de trabajo. A) Elástico unipodal para el ejercicio de extension de cadera (~48 kg); B) Elástico unipodal para la aducción de cadera; C) Gemelo bipodal en Kbox; D) Prevención tendinopatía unipodal (kettlebell) (6-8 kg); E) Sentadilla bipodal (22-26 kg); F) aceleración-desaceleración con elástico (~48 kg); G) Saltos en condiciones de estabilidad e inestabilidad

INTRODUCCIÓN
6. INTRODUCCIÓN

La exigencia que supone desarrollar un programa de entrenamiento, con independencia de que se utilice para mejorar la calidad de vida o para alcanzar el éxito en el máximo nivel deportivo, obliga a técnicos y deportistas a llevar un control riguroso de los cambios que tienen lugar en el organismo como respuesta adaptativa (crónica o aguda) a las cargas de trabajo.

Los adelantos que han proporcionado en los últimos años el desarrollo tecnológico y la investigación científica, unidos al conocimiento cada vez más profundo que hoy día se tiene de cada modalidad deportiva, permiten que el proceso de preparación de los deportistas se realice de forma más precisa, tratando de optimizar el logro de objetivos específicos (salud, condición física o rendimiento deportivo).

En la actualidad, los técnicos deportivos disponen de numerosos métodos y herramientas para valorar el entrenamiento y los cambios que se provocan en el funcionamiento, composición o morfología de las diferentes estructuras, órganos y sistemas del cuerpo humano. Lo verdaderamente trascendente es que, con el paso del tiempo, estos procedimientos y herramientas de evaluación son cada vez más baratos, sencillos y fiables, a la vez que menos cruentos e invasivos.

Uno de los principales objetivos de la investigación aplicada al entrenamiento deportivo ha sido desarrollar medios y procedimientos que permitan evaluar con precisión los cambios en la musculatura de un sujeto por efecto de la práctica regular o puntual de ejercicio físico. Los músculos, estructuras encargadas de generar la tensión (fuerza) necesaria para ejecutar un movimiento, pueden ser examinados desde diferentes perspectivas según el interés del evaluador (forma, composición, funcionalidad, etc.).

Así, en la década de los 90, el profesor Vojko Valencic, de la *Faculty of Electrical Engineering* en Liubliana (Eslovenia), desarrolló una herramienta conocida como tensiomiografía, a partir de ahora TMG. Su principal objetivo era evaluar el estado funcional muscular en los pacientes con patologías neuromusculares (Valencic, 1990). Su aplicación rápidamente se trasladó al campo del deporte debido a los trabajos realizados en la mencionada universidad (*Laboratory of Biomedical Visualization and Muscle Biomechanics* y el *Laboratory for Computacional Electromagnetics de la Universidad de Ljubljana*). (Valencic, & Knez, 1997). Sus estudios con deportistas se intensificaron cuando este grupo de trabajo comenzó a colaborar con el equipo olímpico esloveno durante su preparación para los Juegos Olímpicos de Sydney (2000) y los Juegos Olímpicos de Invierno de Salt Lake City (2002)

(Valencic, Djordjevic, Knez, Dahmane, Coh, Jurcic-Zlobec, Praprotnik, Simunic, Kersevan, Bednarik, & Godina, 2000; Valencic, Knez & Simunic, 2001).

Por lo tanto, y en base al interés que despierta la TMG en mi persona, la presente Tesis Doctoral se ha diseñado en base a tres estudios: uno de carácter metodológico, en el que se analizan qué características debe tener la evaluación de TMG para proporcionar valores fiables (Estudio I), otro de aplicación de la herramienta TMG a un protocolo de entrenamiento que permite un control preciso del estímulo aplicado al deportista (entrenamiento de fuerza cuantificado por la velocidad de ejecución) (Estudio II), y otro con una perspectiva más holística, sobre cómo utilizar la TMG para examinar la evolución de las propiedades mecánicas de los músculos más relevantes en las piernas en jugadores de futsal de la máxima categoría española a lo largo de una temporada (Estudio III). Por tanto, los objetivos principales de cada uno de los estudios que componen la presente tesis doctoral son: 1) analizar los efectos de la duración del pulso y el tamaño del electrodo en las propiedades contráctiles del músculo; 2) investigar la respuesta aguda en las diferentes propiedades mecánicas musculares, estrés metabólico y fatiga neuromuscular tras la realización de dos protocolos de entrenamiento de fuerza que diferían en el número de repeticiones dentro de la serie; y 3) analizar los cambios en las propiedades contráctiles producidas durante la temporada en jugadores profesionales de fútbol sala, así como determinar el posible efecto acumulativo de un programa de entrenamiento de fuerza realizado una vez por semana sobre las propiedades contráctiles en estos jugadores.

7. ESTADO ACTUAL DEL CONOCIMIENTO

El sistema neuromuscular se puede definir como la red de neuronas y músculos que participan en el control del movimiento y la postura (Abbas, 2014). De esta manera, un sistema neuromuscular adecuado, es importante para poder evitar lesiones o dolencias innecesarias, así como para alcanzar el perfeccionamiento técnico de una acción motriz. Por tanto, siguiendo a Romero y Tous (2010), el sistema neuromuscular se basa en dos vías: la vía aferente (procesamiento de la información) y la vía eferente (ejecución). La actividad neuromuscular se puede evaluar mediante diferentes herramientas que se detallarán a continuación.

Una herramienta que se utiliza para determinar la respuesta neuromuscular es la electromiografia (EMG). Dentro de las distintas técnicas utilizadas, la EMG de superficie (sEMG) registra la actividad eléctrica mediante sensores de superficie. Según Jaramillo-Yánez, Benalcázar & Mena-Maldonado (2020), esta actividad eléctrica se produce a partir de dos estados: el primero de ellos es cuando un músculo esquelético está en reposo, donde cada una de las células musculares (es decir, las fibras musculares) tiene un potencial eléctrico de aproximadamente ~80 mV (Weiss, Weiss & Silver, 2015), mientras que el segundo estado es cuando cambia el potencial eléctrico de la unidad motoro (UM), la cual está compuesta por fibras musculares y una neurona motora, se produce una liberación de calcio al retículo sarcoplasmático y se da la contracción muscular. Estas diferencias de potencial eléctrico tienen lugar cuando una motoneurona activa una unión neuromuscular enviando dos potenciales de acción intracelulares en direcciones opuestas. Luego, se propagan despolarizando y repolarizando cada una de las fibras musculares (Rodriguez-Falces, Navallas, Gila, Malanda, & Dimitrova, 2012). La suma de los potenciales de acción intracelulares de todas las fibras musculares de una UM se denomina potencial de acción de la UM (MUAP). Por lo tanto, cuando un músculo esquelético se contrae, el EMG es una suma lineal entre varios MUAPs (Weiss et al., 2015). Hasta la fecha, los índices que se han propuesto para caracterizar las señales de EMG para evaluar la fatiga muscular pueden agruparse en dos clases: índices lineales y no lineales (Kahl & Hofmann, 2016). Los índices lineales más utilizados son la raiz cuadrada media de la señal (RMS, en ingles root mean square), la frecuencia media (MNF), la frecuencia mediana (MDF) y el índice de Dimitrov (DSI) (González-Izal, Malanda, Gorostiaga, & Izquierdo, 2012). A su vez, se ha demostrado que una disminución de la MNF o de la MDF y un aumento del DSI indican un desplazamiento del sEMG de las frecuencias altas a las bajas, lo que se asocia con una disminución de la velocidad de conducción de los potenciales de

acción en los músculos fatigados (Dimitrov, Arabadzhiev, Mileva, Bowtell, Crichton, & Dimitrova, 2006; Rampichini, Taian, Castiglioni, & Merati, 2020).

Por otra parte, una técnica que sirve para la evaluación del control de las propiedades mecánicas musculares es la mecanomiografía, en adelante MMG. La MMG se puede definir como el conjunto de técnicas que tratan de medir las propiedades mecánicas del músculo en respuesta a una contracción muscular voluntaria o estimulada eléctricamente (Ibitoye, Hamzaid, Zuniga, & Abdul Wahab, 2014). Entre las diferentes técnicas de MMG, se encuentra la vibromiografía (VMG), la cual es una técnica que permite registrar sonidos de baja frecuencia creados durante la actividad muscular. Por su parte, la sonomiografía (SNG) es un procedimiento médico de diagnóstico que utiliza ondas sonoras de alta frecuencia (ultrasonidos) para producir imágenes visuales dinámicas de los órganos, los tejidos o el flujo sanguíneo dentro del cuerpo.

Otra herramienta que ha tenido gran acogida en el ámbito científico es el miotono, "Myoton" de aquí en adelante. Dicha herramienta está incluida dentro de la familia de la MMG. El Myoton es una técnica no invasiva e indolora que permite evaluar las propiedades mecánicas y viscoelásticas del músculo como son: la rigidez muscular, el tono muscular, la elasticidad del músculo, el tiempo de relajación y la profundidad muscular (Garcia-Bernal, Heredia-Rizo, Gonzalez-Garcia, Cortés-Vega, & Casuso-Holgado, 2021). La rigidez muscular se refiere a la resistencia de los tejidos blandos a una fuerza externa y se calcula utilizando la respuesta de oscilación natural amortiguada, registrada por un acelerómetro incorporado (Pruyn, Watsford, & Murph, 2016). El tono muscular se cuantifica mediante la frecuencia natural de la señal de aceleración, mientras que la elasticidad del músculo, que es inversamente proporcional a la rigidez, se determina mediante las oscilaciones secuenciales cuando el músculo recupera su forma a partir de la deformación (Ilahi, Masi, White, Devos, Hederson, & Nair, 2020). Por su parte, el tiempo de relajación refleja el tiempo que tarda el músculo en volver a su posición natural (Ilahi et al., 2020), y la profundidad muscular se define como la elongación gradual del músculo bajo una constante tensión (Kawczyński, Mroczek, Andersen, Stefaniak, Arendt-Nielsen, & Madeleine, 2018). Así, el *Myoton* se ha utilizado para evaluar las propiedades del músculo (Gervasi, Sisti, Matori, Andreazza, Benelli, Sestili, Rocchi, & Calavalle, 2017; Park, Yang, Kim, Heo, Uhm, & Yoon, 2017) y la rigidez del tendón (Cristi-Sánchez, Danes-Daetz, Neira, Ferrada, Yáñez Díaz, & Silvestre Aguirre, 2019; Pozarowszczyk, Pawlaczyk, Smoter, Zarzycki, Mroczek, Kumorek, Witkowski, & Adam, 2017).

Asimismo, otra de las herramientas de evaluación y control de las propiedades mecánicas musculares incluidas dentro de la MMG es la TMG. Sobre este método girará la

presente tesis doctoral y sobre la cual se profundizará a continuación. La TMG ha recibido gran aceptación en la última década por parte de la comunidad científica y clínica (Macgregor, Hunter, Orizio, Fairweather, & Ditroilo 2018). El dispositivo incorpora un sensor de desplazamiento de alta precisión (4 µm) que se coloca perpendicularmente con una ligera tensión previa (0.2 N·cm⁻²) en la superficie del vientre del músculo para registrar la deformación radial del músculo tras una estimulación externa evocada eléctricamente (Valencic, & Knez, 1997). Esta pre-tensión se considera una diferencia fundamental entre la TMG y otras técnicas de MMG (Simunic, Degens, Rittweger, Narici, Mekjavić, & Pisot, 2011). De hecho, Križaj, Simunic, & Zagar (2008) confirmaron que esta pre-tensión es fundamental para poder realizar mediciones con el sensor de desplazamiento en TMG en comparación con otras herramientas de MMG (Macgregor et al., 2018). Por ejemplo, las principales diferencias entre la TMG y el Myoton son: 1) la TMG utiliza encóder óptico de alta precisión mientras que el myoton utiliza un acelerómetro; 2) en TMG se estimula el musculo de forma eléctrica para obtener una contracción involuntaria, mientras el Myoton es totalmente pasivo, es decir, no hay estimulación alguna y por tanto no mide una respuesta, solamente mide la deformación del tejido y cuándo lo comprime. 3) Los parámetros obtenidos son distintos, el Myoton mide principalmente la oscilación en la aceleración (tono, stiffness, elasticidad, relajación y ration de deformación) mientras que la TMG evalúa parámetros relacionados con la deformación que produce el estímulo eléctrico y el tiempo que se tarda en responder al mismo; 4) el Myoton permite realizar mediciones sobre tendones, mientras que la TMG sólo músculos; 5) la interpretación de los parámetros por tanto no es extrapolable entre dispositivos, ya que el sensor utilizado para medir no es el mismo y el protocolo de medición es completamente diferente. A continuación, se describirán las principales variables obtenidas durante la medición de las propiedades mecánicas musculares con la herramienta TMG.

7.1 Parámetros tensiomiográficos

La TMG proporciona cinco parámetros: desplazamiento radial máximo (Dm), tiempo de contracción (Tc), tiempo de retardo (Td), tiempo de relajación (Tr) y el tiempo de contracción sostenida (Ts) (Figura 1). El Dm se define como la deformación transversal del músculo y viene expresado en milímetros (Pisot et al., 2008). La reducción del Dm a largo plazo se interpreta como un aumento de la rigidez muscular y viceversa (Pisot et al., 2008), sin embargo, el Dm puede bajar también de manera aguda debido a la fatiga inducida por el ejercicio (Piqueras-Sanchiz, Cornejo-Daza, Sánchez-Valdepeñas, Bachero-Mena, Sánchez-

Moreno, Martín-Rodríguez, García-García, & Pareja-Blanco, 2021). El Td representa el tiempo que tarda la estructura muscular analizada en alcanzar el 10% del desplazamiento total observado tras una estimulación (10% de la Dm), y se cuantifica en milisegundos, al igual que el resto de las variables de TMG relacionadas con el tiempo. Como es lógico, su valor dependerá del tipo de fibra dominante en esa estructura, de su estado de fatiga y de su nivel de potenciación y activación (Dahmane, Raja, Djordjevic, Simunic, & Valencic, 2005). Por su parte, el Tc es el tiempo que transcurre desde el Td (10% de Dm) hasta que se alcanza el 90% de la Dm. El Tr, es el tiempo que se toma entre el 90% y el 50% de la Dm en la curva descendente (Figura 1). Por último, el Ts es el tiempo en el cual la contracción se mantiene, y se calcula midiendo el tiempo transcurrido entre el momento en que la Dm alcanza el 50% hasta el momento en el que la Dm vuelve al 50% de Dm. El hecho de que la TMG analice la función muscular de forma no invasiva y selectiva es especialmente apreciado por los entrenadores, preparadores físicos, fisioterapeutas y científicos deportivos, que buscan preferentemente métodos de evaluación precisos y prácticos que no perturben sus rutinas profesionales (Martín-Rodríguez, Loturco, Hunter, Rodríguez-Ruiz, & Munguia-Izquierdo, 2017a; Alentorn-Geli, Alvarez-Diaz, Ramon, Marin, Steinbacher, Rius, Seijas, Ares, & Cugat, 2015; Gil, Loturco, Tricoli, Ugrinowitsch, Kobal, Abad, & Roschel, 2015).



Figura 1. Ilustración del equipo de tensiomografía (TMG) y la clásica onda de respuesta de tic con todos sus parámetros. 1 = Sensor mecánico y electrodos. 2 = Trípode con mano manipuladora. 3 = Unidad de adquisición de datos. 4 = Onda típica extraída de la respuesta de la TMG. Tomado de Martín-Rodríguez et al. (2017a)

Al poner en relación estas medidas directas, se utiliza con frecuencia un parámetro derivado del ratio entre Dm y Tc. Valencic & Knez (1997) y Macgregor, Ditroilo, Smith, Fairweather, & Hunter (2016), definen este parámetro como la velocidad de contracción (Vc) con la siguiente ecuación (Ec. 1):

$$V_c = \frac{Dm}{Tc} [mm/ms] \tag{1}$$

Por su parte, Loturco, Pereira, Kobal, Kitamura, Ramírez-Campillo, Zanetti, Abad, & Nakamura (2016) utilizan otra ecuación para obtener la velocidad de contracción (Ec. 2):

$$V_c = \frac{Dm}{Td + Tc} [m/ms]$$
⁽²⁾

Donde la Vc es el total del Dm dividido entre el Td y Tc. Por su parte Valenzuela, Montalvo, Sánchez-Martínez, Torrontegi, De La Calle-Herrero, Domínguez-Castells, Maffiuletti, & De La Villa (2018), prefieren llamar a Vc, velocidad de deformación en lugar de velocidad de contracción, ya que depende principalmente de la rigidez del músculo, para evitar de esta forma, confusión con la velocidad a la que los sarcómeros se acortan.

A su vez, otra de las medidas derivadas que relaciona Tc y Dm es la velocidad de respuesta normalizada (Vrn):

$$V_{rn} = \frac{\Delta d_r}{\Delta t_c} [mm/ms]$$
⁽¹⁾

$$V_{rn} = \frac{V_r}{Dm} = \frac{\Delta d_r / \Delta t_c}{Dm} \left[\frac{mm/ms}{mm} \right]$$
(2)

$$V_{rn} = \frac{0.8}{t_c} [mm/ms] \tag{3}$$

La ecuación de Vrn (Ec. 3) se ha utilizado en varios estudios (Piqueras-Sanchiz, Martin-Rodriguez, Gonzalez-Hernandez, & Garcia Garcia, 2017; Rodríguez Ruiz, Quiroga Escudero, Rodríguez Matoso, Sarmiento Montesdeoca, Losa Reyna, Saá Guerra, Perdomo Bautista, García Manso, 2012) donde Δdr es el Dm mientras que Δtc representa el Tc (Ec. 1). Además, Vrn es la respuesta de velocidad normalizada y el 0.8 es un valor constante (Ec. 2).

Por último, destacar dos parámetros adicionales que pueden aportar información distinta a los anteriores: velocidad del 10% del Dm (V₁₀) y velocidad del 90% del Dm (V₉₀). Estas ecuaciones indican que V₁₀ (Ec. 6) y V₉₀ (Ec. 7) pueden ser entendidas como el ratio de deformación del Dm hasta un 10% (10% Dm/Td) y hasta un 90% (Dm/(Td + Tc)), en V₁₀ y V₉₀, respectivamente. Estas dos derivaciones fueron obtenidas en base a que Dm es equivalente al desplazamiento máximo del vientre muscular y Tc es la deformación entre 10% y 90% del Dm a la que se une el 10% inicial del Td (de Paula Simola et al., 2015; 2016).

$$V_{10} = \frac{10\%Dm}{Td} \left[\frac{mm}{ms}\right] \tag{6}$$

$$V_{90} = \frac{Dm}{Tc + Td} \left[\frac{mm}{ms}\right] \tag{7}$$

Tras lo comentado, conveniene destacar que la velocidad de deformación, como puntualiza Valenzuela et al. (2018), y velocidad de contracción como definen Loturco et al. (2016), Macgregor et al. (2016), y de Paula Simola et al. (2015), son el mismo concepto pero llamado de diferente manera. De este modo, velocidad de deformación (Vd) es (Ec. 8):

$$Vd = \frac{Dm}{Tc + Td} \left[\frac{mm}{ms}\right]$$
(8)

7.1.1 Deformación radial máxima (Dm)

Una de las cuestiones más relevantes en TMG ha sido determinar la relación que tiene el Dm con la estructura del músculo y la relación que se establece con el grosor muscular. El primero que examinó dicha relación fue Pisot, Narici, Simunic, De Boer, Seynnes, Jurdana, Biolo, & Mekjavic (2008), en un estudio en el que participaron 10 sujetos sanos, los cuales estuvieron durante 35 días en cama, realizándose mediciones de TMG pre y post estos 35 días sobre 4 músculos: bíceps braquial (BB), vasto medial (VM), bíceps femoral (BF) y gastrocnemio medial (GM) (Tabla 1). Se observó una correlación significativa negativa entre los cambios en el grosor muscular medido con ecógrafo del GM y los cambios en la Dm (r = -0.70; p<0.01) (Figura 2). Así, aquellos sujetos que experimentaron una mayor pérdida de masa muscular también fueron aquellos que presentaron mayores incrementos de Dm, los cuales están asociados a una menor rigidez y tono muscular.

Tabla 1. Desplazamiento radial máximo (Dm) antes (Pre) y después (Post) de 35 días de descanso en cama. Tomado de Pisot et al. (2008).

Músculo	PRE	POST	Cambio relativo (%)
BB (mm)	15.8 ± 2.0	15.0 ± 1.8	-5
VM (mm)	8.7 ± 1.6	10.8 ± 1.5	24*
BF (mm)	5.3 ± 1.0	$\textbf{6.6} \pm 1.0$	26*
GM (mm)	4.2 ± 0.8	5.5 ± 1.1	30*

Datos son expresados como media \pm desviación estándar. *p < 0.01 indica diferencias estadísticamente significativas con respecto al Pre. BB: bíceps braquial, Vm: vasto medial; BF: bíceps femoral; GM: Gastronemio medial.



Figura 2. Relación entre los cambios de grosor muscular y los cambios en el desplazamiento radial máximo (Dm) para el músculo gastronemio medial. * *p*<0.01. Tomada de Pisot et al. (2008)

En la misma línea se encuentra el estudio de Simunic et al. (2019), en el que se realizaron 5 mediciones con TMG a los 1-10-16-28-35 días de estar encamados sobre 3 músculos: VM oblicuo (VMO), VM largo (VML) y BF. Tras 35 días tumbados en una cama con una inclinación de 6°, el grosor muscular disminuyó entre un 16-23%, siendo similar a la cantidad de atrofia observada en otros estudios (Alkner, & Tesch, 2004). Por su parte, en el

estudio de Simunic et al. (2019) se refleja un descenso del grosor muscular entre un 13–17% en todos los músculos (p<0.001) y un aumento del Dm (entre un 42% y un 84%) tras 35 días de reposo. Por último, Simunic et al. (2019), también observaron correlaciones significativas negativas en el BF entre los cambios en el Dm y los cambios en el espesor muscular ($r^2 = 0.74$; p<0.001), así como una relación positiva entre los cambios en el Dm del VMO y los cambios en el ángulo de penneación de dicho músculo ($r^2 = 0.61$, p<0.05) (Figura 3).



Figura 3. Correlación entre los cambios observados en el desplazamiento radial máximo (Dm) en el bíceps femoral (BF) y los cambios en el grosor muscular de este músculo (C); correlación entre los cambios en el Dm y los cambios en el ángulo de penneación en el vasto medial oblicuo (VMO) (D). Tomado de Simunic et al. (2019)

Los hallazgos observados en los estudios de Pisot et al. (2008), y Simunic et al. (2019), sugieren que el Dm podría tenerse en cuenta como un parámetro que puede ser usado para detectar tempranamente la disfunción muscular inducida en sujetos encamados. Especialmente, los incrementos de Dm parecen estar asociados a pérdida de masa muscular, al menos en sujetos que vivencien características similares a las expuestas en estos estudios.

Por otro lado, Wilson et al. (2019a), analizaron las posibles relaciones existentes entre el espesor múscular medido con ultrasonido y el parámetro Dm medido con TMG en 2 músculos: recto femoral (RF) y vasto lateral (VL). Los 33 sujetos no entrenados fueron divididos en 3 grupos: grupo peso muerto (GPS); grupo sentadilla (GS) y grupo *hip thrust* (GHT) (Figura 4). Durante 6 semanas, los participantes realizaron el ejercicio correspondiente a su grupo de entrenamiento (GPS, GS y GHT) dos veces por semanas con una intensidad entre el 75% y el 85% de 1RM, además de realizar otros ejercicios complementarios. Los 3 protocolos de entrenamiento de fuerza indujeron descensos significativos (p<0.05*) del Dm del RF, mientras que no se observaron cambios en el Dm del VL (Tabla 2). Además, se observó una correlación negativa y significativa entre el Dm del RF y el espesor muscular de dicho músculo (p<0.001, r = -0.50). Los autores concluyeron que la TMG podría proporcionar información sobre la arquitectura muscular.

Day One	Day Two
Main Exercise (Back squat, deadlift, or hip	Main Exercise (Back squat, deadlift, or hip
thrust)	thrust)
Weeks 1 & 2: 3 x 8* - 75% 1RM	Weeks 1-6: 3 x 10 – 70% 1RM
Weeks 3 & 4: 4 x 6* - 80% 1RM	
Weeks 5 & 6: 5 x 4* - 85% 1RM	
* Last set is AMRAP	
Bench Press	Dumbbell Chest supported Row
Weeks 1 & 2: 3 x 10 – 70% 1RM (RPE 7)	Weeks 1 & 2: 3 x 10 – 70% 1RM (RPE 7)
Weeks 3 & 4: 3 x 8 – 75% 1RM (RPE 7-8)	Weeks 3 & 4: 3 x 8 – 75% 1RM (RPE 7-8)
Weeks 5 & 6: 3 x 6 – 80% (RPE 8)	Weeks 5 & 6: 3 x 6 – 80% (RPE 8)
Underhand-grip pulldown	Incline Press
Weeks 1 & 2: 3 x 10 – 70% 1RM (RPE 7)	Weeks 1 & 2: 3 x 10 – 70% 1RM (RPE 7)
Weeks 3 & 4: 3 x 8 – 75% 1RM (RPE 7-8)	Weeks 3 & 4: 3 x 8 – 75% 1RM (RPE 7-8)
Weeks 5 & 6: 3 x 6 – 80% (RPE 8)	Weeks 5 & 6: 3 x 6 – 80% (RPE 8)
Military Press	Seated Row
Weeks 1 & 2: 3 x 10 – 70% 1RM (RPE 7)	Weeks 1 & 2: 3 x 10 – 70% 1RM (RPE 7)
Weeks 3 & 4: 3 x 8 – 75% 1RM (RPE 7-8)	Weeks 3 & 4: 3 x 8 – 75% 1RM (RPE 7-8)
Weeks 5 & 6: 3 x 6 – 80% (RPE 8)	Weeks 5 & 6: 3 x 6 – 80% (RPE 8)

Figura 4. Cronograma de intensidad y ejercicios. Tomado de Wilson et al. (2019a)

Tabla 2. Cambios en el grosor muscular y propiedades contráctiles de los músculos del cuádri	ceps en respuesta
a 6 semanas de entrenamiento de fuerza de la parte inferior del cuerpo. Adaptado de Wilson et	al. (2019a).

Músculo	Parámetros	Sentadilla		Peso 1	nuerto	Hip thrust		
		Pre	Post	Pre	Post	Pre	Post	
Recto	Dm (mm)	6.1 ± 1.5	$3.9^* \pm 1.6$	6.6 ± 1.6	$5.8^* \pm 1.9$	7.0 ± 2.4	$5.9^* \pm 1.9$	
femoral	Espesor	2.3 ± 0.3	$2.5^*\pm0.3$	2.1 ± 0.3	$2.2^*\pm0.3$	2.1 ± 0.3	$2.3^*\pm0.3$	
	muscular (cm)							
Vasto	Dm (mm)	4.1 ± 2.8	4.2 ± 1.5	4.2 ± 2.3	3.9 ± 2.0	4.0 ± 1.8	3.7 ± 1.0	
lateral	Espesor	1.9 ± 0.5	$2.1^{\ast}\pm0.5$	2.0 ± 0.5	$2.2^{\ast}\pm0.5$	2.0 ± 0.5	$2.1^{\ast}\pm0.5$	
	muscular (cm)							

Media \pm desviación standard. Dm: desplazamiento radial máximo. Cambios significativos $p < 0.05^*$

Un segundo estudio que también tuvo como objetivo analizar la relación entre los cambios producidos por el entrenamiento de fuerza sobre el espesor muscular y los parámetros contráctiles musculares fue el realizado por Kojić, Mandić & Ilić (2021). Durante 7 semanas, 18 sujetos, 9 hombres y 9 mujeres que no habían realizado ejercicios de fuerza 8 meses antes, se sometieron a un programa de entrenamiento de fuerza basado en realizar sentadillas dos

veces por semana. De la semana 1 a la 3, el programa consistía en realizar 3 series con una intensidad del 60% de 1RM y con 2 minutos de recuperación. De la semana 4 a la 7, la intensidad era del 70%, realizando 4 series y 2 minutos de recuperación. Todas las series de entrenamiento se realizaron hasta el fallo muscular. Los resultados obtenidos en TMG en el grupo de hombres por músculos fueron los siguientes: para el RF, el Dm disminuyó un 19.1% (pre: 10.53 ± 3.93 mm; post 8.62 ± 3.18 mm, p<0.01) y el el Tc no cambió de manera significativa (pre: 26.21 ± 10.51 ms; post 27.81 ± 4.89 ms). Para el VL, el Dm descendió un 17% (pre: 6.69 ± 2.86 mm; post 5.72 ± 4.33 mm) mientras que el Tc permaneció estable (pre: 21.37 ± 10.83 ms; post: 21.97 ± 6.85 ms). Con respecto al grupo de mujeres, el Dm en el RF también descendió (pre: 9.25 ± 4.27 mm; post 7.98 ± 3.48 mm, p<0.05) y el Tc permaneció sin cambios significativos (pre: 27.48 ± 8.16 ms; post 28.31 ± 8.33 ms). El VL en el grupo de mujeres, también mostró un descenso, en este caso del 6% (pre: 6.50 \pm 3.96 mm; post 6.10 \pm 3.72 mm) y el Tc también permaneció sin cambios para este músculo (pre: 22.11 ± 5.99 ms; post: 23.98 ± 10.70 ms). Los autores concluyen diciendoque tras 7 semanas de entrenamiento, se produce un aumento de la rigidez muscular por igual para ambos sexos y músculos, no habiendo diferencias significativas entre sexos.

7.1.2 Tiempo de contracción (Tc)

Otra de las cuestiones relevantes en TMG es determinar si los parámetros temporales obtenidos mediante esta herramienta podrían estar relacionados con la composición de las fibras musculares. En concreto los estudios de Dahmane, Valencic, Knez, & Er 2001; Dahmane, Raja, Djordjevič, Šimunič, & Valenčič, 2005), y Simunic et al. (2011), establecieron que hay una relación del Tc con el porcentaje de fibras tipo I.

Dahmane et al. (2001), observaron relaciones entre el Tc y el porcentaje de fibras I (r = 0.93; p<0.001) en un estudio que contó con 30 sujetos: 15 eran cadáveres y 15 sujetos sanos. En esta línea, Dahmane et al. (2005) corroboraron que los parámetros temporales se correlacionaban con la composición muscular, pero esta vez tuvieron en cuenta la sección transversal superficial y profunda. Los autores hallaron una correlación entre el Tc y los músculos que tienen más fibras tipo I (parte superficial: r = 0.76, p<0.01; parte profunda del músculo: r = 0.90, p<0.001; Figura 5), de manera que los músculos que presentaban un mayor porcentaje de fibras tipo I eran los músculos que presentaban valores de Tc más lentos.



Figura 5. Valor medio e intervalos de confianza (95%) de la relación entre el porcentaje de fibras musculares de tipo 1 y tiempos de contracción de la respuesta del vientre muscular a la estimulación de las contracciones en biceps braquial (BB), triceps braquial (TB), flexor superficial del dedo (FDS), extensor digital (ED), biceps femoral (BF), tibial anterior (TA) gastronemio medial (GCM), sóleo (SOL), y músculo braquial (BR), de los sitios a) superficiales y b) profundos. Adaptado de Dahmane et al. (2001).

La principal limitación que tenían dichos estudios es que se analizaron correlaciones entre biopsias de sujetos muertos con mediciones de TMG en sujetos vivos. Con el fin de resolver dicha limitación, Simunic et al. (2011), realizaron sobre el mismo músculo (VM) biopsias musculares y mediciones con TMG. Los autores encontraron una correlación positiva significativa entre la proporción de la cadena pesada de la miosina tipo I y el Tc (r = 0.88; p<0.001) (Figura 6). Estos hallazgos parecen indicar que el Tc puede informar de forma indirecta del porcentaje de fibras tipo I en el músculo evaluado. No obstante, aún es necesario determinar cuál es la causa de que solo se establezca relación con este tipo de fibras y si este parámetro es sensible a los cambios en la composición muscular.



Figura 6. Modelos de regresión univariante entre el tiempo de contracción medido por TMG y el porcentaje de fibras tipo I, evaluadas por el método de cadena pesada de miosina (% MHC-I). Tomado de Simunic et al. (2011).

7.2 Fiabilidad

En ciencia, la fiabilidad se puede definir como la confianza que se tiene en el buen comportamiento o funcionamiento del material (Liaw, Hsieh, Lo, Chen, Lee, & Lin, 2008). La fiabilidad relativa es el grado en que los individuos mantienen su posición en una muestra con mediciones repetidas, mientras que la fiabilidad absoluta es el grado en que las mediciones repetidas varían para los individuos (Klavora, Gaskovski, & Forsyth, 1995).

Para determinar la fiabilidad relativa y absoluta se han usado habitualmente los siguientes indicadores:

Fiabilidad relativa:

- Índice de correlación intraclase (ICC) mide la concordancia entre dos o más valoraciones cuantitativas, en el que valores inferiores a 0.50 se califican como de baja fiabilidad, entre 0.50 y 0.75 indican una fiabilidad moderada, entre 0.75 y 0.90 expresan una buena fiabilidad y los valores superiores a 0.90 indican una fiabilidad excelente (McGraw & Wong, 1996).

Fiabilidad absoluta:

- El error estándar de la medida (SEM) se puede definir como un índice para la precisión de una medida y refleja la consistencia de las puntuaciones dentro de los sujetos individuales (Weir, 2005). Se calcula de la siguiente forma: SEM = $\sqrt{MS_E}$, donde MS_E es el término medio de error cuadrado de las medidas repetidas ANOVA.

- El coeficiente de variación (CV) junto con el respectivo intervalo de confianza (IC) del 95%. Se calcula de la siguiente forma: $CV = \sqrt{(SEM/promedio de los elementos) \cdot 100}$. Un CV mayor al 10% se interpreta como una fiabilidad absoluta insuficiente (Ditroilo et al., 2013; Tous-Fajardo et al., 2010).

- Mínimo cambio detectable (MDC) se define como el tamaño de la diferencia entre dos mediciones repetidas para así distinguir diferencias reales (o cambio relevante) de la medición error. Este parámetro se calcula a través de la siguiente fórmula: MDC = SEM x 1.96 x $\sqrt{2}$ (Weir, 2005).

En relación con la TMG, existen varios estudios en los que se han analizado la fiabilidad de los distintos parámetros obtenidos a través de esta herramienta en diferentes poblaciones (Tabla 3).

Tabla 3. Índice de correlación intraclase (ICC) de los principales parámetros tensiomiográficos. De	esplazamiento
máximo (Dm); tiempo de contracción (Tc); tiempo de retardo (Td). Tabla elaborada a partir de Ro	odríguez et al.
(2017a) y Macgregor et al. (2018).	

Estudio		Parámetros		DS	Estudio		Parámetros		
Rodríguez et al. (2017a)		Dm	Тс	Td	Macgregor et al. (2018)		Dm	Тс	Td
Carrasco et al. (2011)		0.92	0.83	0.89	Rey et al. (2012)		0.95	0.86	0.82
Rodriguez-Matoso et al. (2010)	ICC	0.92	0.97	0.90	Simunic 2012	ICC	0.99	0.98	0.89
Tous-Fajardo et al. (2010)		0.97	0.92	0.86	Ditroilo et al. (2013)		0.86-0.95	0.62-0.92	0.56-0.62
Krizaj et al. (2008)		0.98	0.97	0.94	García-García et al. (2018)		0.99	0.97	0.91

En este sentido, se ha determinado que existe una fiabilidad relativa inter-día (48 horas) entre moderada, buena y excelente para los parámetros Dm, Tc y Td (Tabla 3) en sujetos sanos, sobre distintos ángulos (0° - 45° - 90°) y diferentes músculos (VM, VL y BF) (Ditroilo Hunter, Haslam, & De Vito, 2011). Por otro lado, entre evaluadores con poca experiencia también se ha observado una buena-excelente fiabilidad relativa y absoluta en la medición de TMG en el músculo VM en personas sanas (Tous-Fajardo et al., 2010).

A su vez, la fiabilidad relativa también ha sido evaluada sobre el BB dentro de la sesión por Krizaj et al. (2008) y por Wilson, Jones, Johnson, & Francis (2019b), sobre el RF. Asimismo, a tenor de los resultados obtenidos por Lohr et al. (2019) en una revisión sistemática y meta-análisis sobre la fiabilidad de los parámetros de TMG, las variables más fiables en TMG parecen ser el Dm (ICC: 0.98), el Tc (ICC: 0.96) y el Td (ICC: 0.91) (Figuras 7, 8 y 9). Para conocer la fiabilidad del Dm, estos autores (Lohr, et al., 2018) incluyeron 11 estudios sobre un total de 209 sujetos, siendo el ICC (95% IC) = 0.98 (0.96-0.99) (Figura 7). Respecto al Tc, incluyeron a 9 artículos sobre una población de 181 sujetos, observando un ICC (95% IC) = 0.96 (0.93-0.98) (Figura 8). Por último, para el análisis de la fiabilidad del parámetro Td se examinaron 7 estudios con un total de 147 sujetos, siendo el ICC (95% IC) = 0.91 (0.88-0.96) (Figura 9).

Study	Total	Ĩ	Relativ	ve reliat	oility - D	m	Rel.	95%-CI	Weight	
Garia-García et al., 2018	50					4	0.99	[0.98; 1.00]	33.8%	
Simunic, 2012	10						0.99	[0.97; 1.00]	20.7%	
Krizaj et al., 2008	13					÷	0.98	[0.96; 1.00]	15.0%	
Tous-Fajardo et al., 2010	18					÷	0.97	[0.94; 1.00]	11.2%	
Lohr et al., 2018a	24						0.95	[0.92; 0.99]	7.3%	
Paula Simola et al., 2016aexp1	20						0.94	[0.88; 0.99]	3.9%	
Paravlic et al., 2017	18						0.93	[0.86; 0.99]	2.6%	
Rey et al., 2012	15					-+-	0.92	[0.84; 1.00]	1.9%	
Wilson et al., 2018	10					-+-	0.92	[0.82; 1.02]	1.2%	
Ditroilo et al., 2013	21					-+-	0.92	[0.84; 0.99]	2.4%	
Ditroilo et al., 2011	<mark>10 ←</mark>	+	-				0.37	[-0.20; 0.93]	0.0%	
Random effects model	209					•	0.98	[0.96; 0.99]	100.0%	
Prediction interval								[0.95; 1.00]		
Heterogeneity: $\tau^2 < 0.0001$		-	1	1	1			-		
		0	0.2	0.4	0.6	0.8 1				

Figura 7. Fiabilidad relativa dentro de la prueba y del test con intervalos de confianza (CI) del 95% para el desplazamiento radial máximo (Dm). Tomado de Lohr et al. (2019).



Figura 8. Fiabilidad relativa dentro de la prueba y del test con intervalos de confianza (CI) del 95% para el tiempo de contracción (Tc). Tomado de Lohr et al. (2019).

Study	Total	I	Relativ	e relia	bility - 1	۲d	Rel.	95%-CI	Weight
Simunic, 2012 Krizaj et al., 2008 Garía-García et al., 2018 Paula Simola et al., 2016aexp1 Tous-Fajardo et al., 2010 Rey et al., 2012 Ditroilo et al., 2013	10 13 50 20 18 15 21						- 0.95 - 0.94 0.91 0.90 0.86 0.82 0.60	[0.88; 1.01] [0.87; 1.01] [0.86; 0.96] [0.81; 0.99] [0.73; 0.98] [0.65; 0.99] [0.32; 0.88]	21.2% 20.9% 32.2% 13.5% 6.8% 3.9% 1.5%
Random effects model Prediction interval Heterogeneity: $\tau^2 = 0.0004$	147	0	0.2	0.4	0.6	0.8	0.91	[0.88; 0.95] [0.84; 0.98]	100.0%

Figura 9. Fiabilidad relativa dentro de la prueba y del test con intervalos de confianza (CI) del 95% para el tiempo de retardo (Td). Tomado de Lohr et al. (2019).

Estos datos están en la línea con los expuestos por Martín-Rodríguez et al. (2017a) y Macgregor et al. (2018) (Tabla 2). Por lo que se puede extraer que los parámetros Dm, Tc y Td son altamente fiables y son los que deberían tenerse en cuenta a la hora de realizar valoraciones de TMG.

7.3 Protocolo de medición de tensiomiografía

En este apartado se presentan los criterios para la realización de una correcta medición con TMG. Debido a que los valores de los parámetros evaluados con TMG pueden ser influenciados por el punto de colocación de sensor y electrodos, parece adecuado utilizar una guía electromiográfica para la localización de los músculos a medir, como la propuesta por Hermens, Freriks, Disselhorst-Klug, & Rau (2000).

7.3.1.1 Localización del músculo

Un aspecto que se debe tener en cuenta es la localización del vientre muscular. Con el fin de determinar las diferencias de medición entre la zona del vientre muscular y zonas adyacentes, Simunic et al. (2019), evaluaron 27 localizaciones distintas en los siguientes músculos: BB, VL, VM, RF, y erector espinal (ES) (Figuras 10 y 11).



Figura 10. Tres sensores de tensiomografía que se colocaron paralelos para la realización del estudio (B); los veintisiete (3 x 9) puntos de medición en cada músculo (A). La distancia lateral y longitudinal entre los puntos de medición vecinos era de 1.5 cm. Tomado de Simunic et al. (2019).



Figura 11. Puntos de medición en biceps braquial (A), vasto lateral (B), vasto medial (C), recto femoral (D), y erector espinal (E). Tomado de Simunic et al. (2019).

Dichos autores (Simunic et al., 2019) utilizaron para el posterior análisis el parámetro "Error Relativo", que definieron como la diferencia relativa entre cada punto de medición en comparación con el punto de medición de referencia. De los datos recogidos en la Tabla 4 se desprende que la utilización de una guía es clave para una correcta medición pues tanto el Dm como el Tc se vieron modificados por el punto de evaluación, ya que a mayor distancia del vientre muscular mayor fue el error relativo.

Tabla 4. Superficie (en cm^2) para el 3%, 5% y 10% del error relativo según el punto de medición de referencia. El error relativo se presenta para el tiempo de contracción y el desplazamiento radial máximo, por separado. Adaptado de Simunic et al. (2019).

Error relativo	Tien	npo de contr	acción	Desp	Desplazamiento máximo			
	3%	5%	10%	3%	5%	10%		
BB	6.9	10.4	23.1	4.0	5.6	9.5		
VL	3.8	7.1	16.6	3.8	6.0	11.4		
VM	4.6	8.8	25.2	8.2	9.4	12.3		
RF	9.5	15.8	26.7	6.2	9.0	16.2		
ES	3.7	5.9	15.2	2.4	3.9	7.6		

BB: Biceps braquial; VL: Vasto lateral: VM: Vasto medial; RF: Recto femoral; ER: Erector espinal

7.3.1.1 Posición del ángulo articular durante la medición

Los ángulos en los que se halla fijada la articulación durante la medición también parece ser otro factor que modula los valores obtenidos con TMG. Ditroillo et al. (2011), determinaron que al evaluar el BF en sujetos sanos, el ángulo de la rodilla (0°-45°-90°) modificaba significativamente el Dm y el Tc (Tabla 5). Una forma de estandarizar los valores obtenidos con TMG sería por lo tanto utilizar los foam originales de TMG puesto que siempre se estaría midiendo con la misma angulación.

	BF	
Ángulo (°)	Tc (ms)	Dm (mm)
0	26.20 ± 15.03 ^{ab}	$2.38 \pm 1.31^{\rm a}$
45	42.36 ± 16.05	4.11 ± 1.99^{b}
90	43.92 ± 20.76	2.28 ± 1.84

Tabla 5. Medidas obtenidas en el bíceps femoral en 3 ángulos diferentes. Tomado de Ditroillo et al. (2011).

 $Media \pm desviación \ standard$

^aSignificativamente diferente de 45° (*p*<0.01).

^bSignificativamente diferente de 90° (p<0.01).

Tc: tiempo de contracción; Dm: desplazamiento radial máximo

7.3.1.2 Distancia entre electrodos

Otro apartado dentro de los protocolos de medición que influye en la fiabilidad de la medida es la distancia entre parches (DEP). Wilson et al. (2019b), comprobaron que se debe tener presente la DEP en cada medición. Para ello, realizaron mediciones en el RF con una separación entre parches de 5-7-9-11 cm (Figura 12). Así, los resultados mostraron que la distancia adecuada para medir en TMG es una DEP entre 5 y 7 cm. La literatura científica sugiere que a mayor DEP, mayores valores de Dm (Tous et al., 2010; Wilson et al., 2019b), lo cual podría deberse a un aumento del reclutamiento de fibras musculares (García-García et al., 2015). Además, se sabe que las neuronas superficiales y de mayor diámetro son las primeras en ser reclutadas mediante estimulación eléctrica (Mortimer & Bhadra, 2004). Por lo tanto, es lógico sugerir que a medida que la DEP aumenta, el número de células nerviosas motoras que se estimulan incrementará. Los resultados de Wilson et al. (2019b), están en sintonía con los obtenidos por Tous et al. (2010), en el VL, pero en este caso con una DEP entre 3 y 5 cm.



Figura 12. Desplazamiento radial máximo (Dm) medido mediante tensiomiografía a cuatro distancias entre electrodos. Se presentan los datos medios e individuales de cada uno de los 10 participantes. Diferencia significativa entre los grupos, *p*<0.05. Tomado de Wilson et al. (2019b).

7.4.1.1 Tipo de parche y duración del estímulo

Un factor que podría influir en las evaluaciones realizadas con TMG podría ser la duración del impulso y el tamaño del parche utilizado, puesto que la energía eléctrica necesaria para estimular un músculo está influenciada por la duración del impulso eléctrico, como se muestra en la siguiente ecuación de Alon, Kantor & Ho (1994): $E = V \times I \times \Delta t$, donde E es la energía eléctrica, V son los voltios, I es igual a corriente y Δt es la duración del impulso. Por lo tanto, parece que duraciones del impulso menores a 1 ms podrían provocar menor actividad eléctrica y no estimular suficientes unidades motoras (Bickel, Gregory, & Dean, 2011). A su vez, Milner-Brown, Stein, & Yemm (1973), analizaron en un aparato de estimulación nerviosa eléctrica transcutánea (TENS) los posibles efectos del tamaño del electrodo a nivel neural y motor, llegando a la conclusión de que cuanto más grande es el electrodo, mayor es la participación de las unidades motoras.

7.4.1.2 Experiencia del evaluador

Otro factor a tener en cuenta a la hora de establecer la fiabilidad de la medida es la experiencia del evaluador. La fiabilidad entre observadores ha sido abordada cuando los dos evaluadores carecen de experiencia, ya que solo se habían familiarizado con la TMG en dos sesiones previas (Tous et al., 2010). No obstante, desde nuestro conocimiento, no se ha encontrado ningún estudio que examine la influencia de la experiencia del evaluador sobre la fiabilidad de la medida. Sin embargo, es habitual en muchos de los trabajos publicados que en

la parte metodológica se reflejen los años de experiencia de los evaluadores. Esta circunstancia podría ser debida a que los evaluadores más expertos tienen más experiencia en localizar el vientre muscular y colocar el sensor de desplazamiento de forma perpendicular. Este punto de vista deberá ser comprobado en el futuro.

7.5 Protocolo de medición

En TMG existen principalmente 3 tipos de protocolos que se utilizan habitualmente para medir:

<u>Medición 1</u>: Conocido como "protocolo incremental". Se empieza por 20-30 mA y se va aumentando progresivamente de 10 ó 20 mA hasta llegar a 100 mA, que es el máximo estímulo que permite el estimulador eléctrico (Figura 13). Esta es la forma más usada para medir (García-García, Cancela-Carral, Martínez-Trigo, & Serrano-Gómez, 2013; Piqueras-Sanchiz et al., 2019). Tiene como ventaja que sabemos con exactitud el Dm y la intensidad a la cual cada músculo alcanza su pico máximo.



Figura 13. Típica progresión de la curva de desplazamiento. Tomado de Macgregor et al. (2018).

<u>Medición 2</u>: Un único estímulo a la máxima intensidad (Rodríguez-Ruiz et al., 2014). Tiene como principal ventaja el ahorro de tiempo, ya que, se realiza una sola medición por deportista/paciente. Como principal inconveniente se podría destacar que no se sabe con exactitud si el Dm es el máximo que se puede alcanzar.

<u>Medición 3:</u> 50-75-100 mA (Rey, Lago-Peñas, & Lago-Ballesteros, 2012). Tiene como inconvenientes que solo se realizan 3 mediciones y por ende quizás no se sepa con exactitud si

el Dm pico obtenido es el máximo que se puede alcanzar, o si se alcanzaría el Dm máximo con alguna de las intensidades intermedias. 7.5 Aplicaciones prácticas del uso de la TMG

7.5.1 Fatiga muscular

La fatiga muscular se define típicamente como la reducción temporal de la capacidad de un músculo para producir fuerza o potencia (Enoka & Duchateau, 2008). La disminución de la fuerza o de la potencia puede atribuirse a factores centrales (cerebro y/o médula espinal) o factores periféricos (músculo o sistema nervioso periférico). Los factores centrales incluyen la alteración de las neuronas motoras al disminuir la actividad de los neurotransmisores alterando la excitabilidad en la corteza y la inhibición de la excitabilidad espinal por vía aferente (Gandevia, Allen, Butler, & Taylor, 1996). La fatiga periférica puede surgir por el deterioro del acoplamiento excitación-contracción a través de la liberación de calcio o por la inhibición de la actividad de las enzimas a través de la acidosis local (Walker, Blazevich, Haff, Tufano, Newton, & Häkkinen, 2016).

En la literatura científica existen numerosos estudios en los cuales se ha evaluado la fatiga muscular con TMG (Wiewelhove, Raeder, Meyer, Kellmann, Pfeiffer, & Ferrauti, 2015; García-Manso, Rodríguez-Ruiz,Rodríguez-Matoso, de Saa, Sarmiento, & Quiroga, 2011b). Conviene destacar que la fatiga ha sido inducida por ejercicios de resistencia, tanto aeróbica (García-Manso et al., 2011b) como anaeróbica (Wiewelhove et al., 2015), por protocolos de fuerza usando como medio una maquina isocinética (Hunter, Galloway, Smith, Tallent, Ditroilo, Fairweather, & Howatson, 2012), o por una combinación de ambas capacidades (de Paula Simola et al., 2015).

7.5.1.1 Fatiga inducida por entrenamiento de resistencia

Uno de los primeros estudios en el que se examinaron los efectos de la fatiga muscular localizada tras realizar un esfuerzo de larga duración fue el de García-Manso et al. (2011a). En dicho estudio, 19 deportistas experimentados (edad 37.9 ± 7.1 años; altura 177.5 ± 4.6 cm; peso: 73.6 ± 6.5 kg) realizaron un triatlón de ultra resistencia (3.8 km de natación, 180 km en bicicleta, y seguidamente una maratón, es decir 42.2 km de carrera) y se realizó la TMG sobre

el RF y BF antes y después de completar dicha prueba. Los resultados mostraron un aumento significativo en el BF del 16% en el Tc (sumando pierna izquierda más pierna derecha, pre: 65.1 ± 22.1 ms; post: 77.4 ± 28.5 ms, p<0.05) y del 19% en el Dm (pre: 10.8 ± 3.5 mm; post: 13.4 ± 4.6 mm, p<0.05) pero no en el RF. Giovanelli, Taboga, Rejc, Simunic, Antonutto, & Lazzer (2016) también evaluaron el efecto de la fatiga inducida por la carrera de larga duración (una maratón cuesta arriba) en las propiedades musculares por medio de la TMG y examinaron si los cambios en la mecánica de la carrera y en los parámetros de la TMG eran diferentes entre los corredores más rápidos y los más lentos. En este caso, el Dm en el VL aumentó un 19% entre los deportistas más rápidos (pre: 6.6 ± 1.7 mm; post: 8.1 ± 3.0 mm, p<0.05) y un 12% entre los deportistas más lentos (pre: 6.2 ± 2.3 mm; post: 7.9 ± 2.8 mm, p<0.05). Además, también se observó un descenso del Tc para los dos tipos de corredores (rápidos: -15%, pre: 25.8 ± 5.4 ms; post: 22.0 ± 3.5 ms, p<0.001; lentos: -11%, pre: 25.3 ± 4.1 ms; post: 22.6 ± 2.4 ms, p<0.001). Los autores concluyeron que el Dm y el Tc se vieron alterados debido a la fatiga producida por la carrera, especialmente en los corredores más rápidos.

Wiewelhove et al. (2015) también analizaron el efecto de la fatiga producida por un protocolo de resistencia, en este caso un esfuerzo intermitente de alta intensidad, sobre los parámetros evaluados en la TMG. El protocolo se basó en 11 entrenamientos distribuidos en 6 días sobre 22 sujetos (11 hombres y 11 mujeres) bien entrenados, donde se realizaban ejercicios intermitentes de alta intensidad a lo largo de 35 minutos. Los ejercicios de la parte principal consistían en correr series de entre 2 y 4 minutos al 80-85% de la velocidad final del test (VIFT) y sprints de 40 metros. Tras el programa de entrenamiento (11 sesiones), se observó un aumento del 10% del Tc en el RF (pre: 29.0 \pm 3.8 ms, post: 31.7 \pm 4.8 ms, p<0.05) y del 28% en el BF (pre: 37.7 ± 0.7 ms, post: 44.9 ± 12.9 ms, p < 0.05). Wiewelhove et al. (2017) realization otro estudio sobre fatiga y TMG esta vez en tenistas, en el que se realizaron 7 sesiones en 4 días. En cada sesión, los jugadores realizaron tres series de ocho repeticiones de carrera, con 20 s de recuperación pasiva entre repeticiones y 6 minutos de recuperación pasiva entre cada serie. Cada repetición tenía una duración de 15 s y consistía en series de velocidad lanzada de 20 m al 90% de la VIFT. En este caso, no se observaron cambios significativos en las variables de TMG evaluadas, ya que la Dm pasó de 8.8 ± 1.9 mm a 8.1 ± 2.0 mm (cambio $-8.7 \pm 0.9\%$) y el Tc de 31.3 \pm 3.9 ms a 31.5 \pm 4.6 ms, (cambio: 0.2 \pm 2.0%). Por lo tanto, estos autores concluyeron que los parámetros de TMG no eran lo suficientemente sensibles para detectar cambios en el rendimiento y, en consecuencia, no es capaz de detectar la fatiga. No obstante, dicho estudio recibió una letter de Martín-Rodríguez, Zubac, Piqueras-Sanchiz, Bautista, & Simunic (2017b), ya que basándose en el G^{*}*Power* software y en los cálculos Bayesanos, los

resultados en relación a la Dm decían todo lo contrario. Los IC al 95% y los cálculos bayesianos de Dm, indican que la probabilidad de asociación era mayor al 75% si el tamaño de la muestra hubiese sido mayor (Tabla 6).

Tabla 6. Intervalo de confianza al 95% y cálculo bayesiano e interpretación de los resultados del autor con un 80% de probabilidad previa. Adaptado de Martín-Rodríguez, et al. 2017). M: media; SD: desviación estándar; CI: intervalo de confianza; Dm: Desplazamiento máximo.

TMG parámetro	$M \pm SD (95\% \text{ CI}) \text{ pre-post}$	<i>p</i> -value	Cálculo bayesano		
			% de no asociación	% de asociación	
Dm	$-0.7 \pm 0.1 \ (-0.76; \ 0.64)$	0.178	24.31	75.69	

7.5.1.2 Fatiga inducida por entrenamiento de fuerza

En relación a la evaluación de la fatiga inducida por distintos protocolos de ejercicio de fuerza, Raeder, Wiewelhove, Simola, Kellmann, Meyer, Pfeiffer, & Ferrauti (2016), sometieron a 23 deportistas bien entrenados durante 6 días a 11 sesiones de fuerza (sentadilla con *flywheel*; ejercicios de sobrecarga excéntrica; ejercicios pliométricos; series descendientes; series múltiples) con el objetivo de generar un alto estrés a nivel muscular. Se observó un descenso del Dm en el VM de un 14.5% (tamaño del efecto [ES] = -0.60) y del V₉₀ en el mismo músculo de un 15.5% (ES = -0.62), los cuales, tras 3 días de recuperación no habían vuelto a sus valores basales. Por su parte, Hunter et al. (2012) examinaron la fatiga producida sobre el músculo BB tras un protocolo de 5x10 contracciones excéntricas de extensión de codo en una máquina isocinética. Tras la realización de este ejercicio, el Dm y el Tc disminuyeron de forma estadísticamente significativa (p < 0.01). Los autores sugierieron que la TMG es útil para detectar el deterioro de la función muscular producido por el entrenamiento de fuerza. Por su parte, Macgregor et al. (2016) emplearon un programa de estimulación eléctrica durante 5 minutos para provocar fatiga en el gemelo medial (Gm). Como resultado de dicho protocolo, el Dm del Gm descendió un 17% (p<0.05). Asimismo, de Paula Simola et al. (2016) realizaron un estudio con deportistas entrenados en el que éstos fueron divididos en dos grupos. Un grupo realizó ejercicios de fuerza combinados con ejercicios de CORE. Los ejercicios de fuerza son los que se han descrito al inicio de este párrafo en el estudio llevado a cabo por Raeder, et al. (2016). Un segundo grupo realizó ejercicios de alta intensidad basados en sprints de Wingate. Los resultados mostraron una disminución significativa del Dm en el VL de un 20% en el grupo

de fuerza y de un 11% en el grupo de resistencia. Además, también se observó una disminución significativa del Tc de un 4% en el de grupo de fuerza y de un 5% en el grupo de resistencia.

A su vez, la TMG se ha utilizado en distintos métodos que analizan fatiga producida por entrenamientos de fuerza:

• Contracciones isométricas

Agustín, Medina-Mirapeix, Casaña-Granell, García-Vidal, Lillo-Navarro, & Benítez-Martínez (2020), realizaron un estudio con 39 sujetos donde midieron los parámetros de TMG en 3 músculos: RF-VL-VM. Tras la realización de un protocolo de fatiga basado en una contracción isométrica al 70% de la máxima contracción isométrica voluntaria (MVIC) durante 60 segundos, se observó una disminución significativa del Dm en todos los músculos evaluados: RF: 25% (p<0.05); VL: 18% (p<0.05) y VM: 17% (p<0.001). En relación con el parámetro Tc, sólo se observó una disminución significativa en el VM: 8% (p<0.05). Sin embargo, se debería mencionar como posible limitación del citado trabajo, el uso de una almohadilla diferente a las recomendadas por el fabricante (Figura 14). Como se pudo ver en el artículo de Ditroilo et al. (2011), diferentes ángulos en las articulaciones pueden dar lugar a respuestas distintas.



Figura 14. A: Almohadilla triangular TMG, 139°; B: Almohadilla triangular utilizada estudio Martín-San Agustín et al. (2020).

Contracciones concéntricas-excéntricas

Por su parte, Muñoz-López, De Hoyo, Nuñez, & Sañudo (2022), evaluaron las propiedades contráctiles musculares pasivas después de un ejercicio isocinético de fatiga que incluía contracciones concéntricas o excéntricas. En primer lugar, se realizó la primera medición con TMG. Inmediatamente después de esta medición, los sujetos realizaron un

calentamiento estandar tras lo cual se realizó una segunda medición con TMG. A continuación, los sujetos ejecutaron una MVIC y un protocolo de fatiga consistente en realizar 32 repeticiones máximas en una máquina isocinética de extensión y flexión de rodilla a 180%. Se observó un descenso en todos los parámetros medidos con TMG (Dm y Tc) (Tabla 7).

Tabla 7. Propiedades contráctiles del músculo: valores descriptivos antes (Pre) y después (Post) del ejercicio de fatiga para ambos tipos de contracciones musculares (concéntrica-excéntrica. Adaptado de Muñoz-López et al. (2020). Los datos se expresan como media \pm SD. SD: desviación estándar; Tiempo de contracción: Tc; Desplazamiento máximo: Dm.

		VASTO LA	ATERAL		BÍCEPS FEMORAL					
	CONCÉN	CONCÉNTRICO EXCÉNTR		TRICO	CONCÉ	NTRICO	EXCÉN	INTRICO		
	Pre	Post	Pre	Post	Pre	Post	Pre	Post		
Тс	22.82 ± 3.68	22.03 ± 3.80	22.63 ± 3.52	20.47 ± 2.90	39.00 ± 17.83	35.42 ± 19.48	36.23 ± 16.29	30.46 ± 15.19		
(ms)										
Dm	5.28 ± 1.61	4.08 ± 1.47	5.39 ± 1.70	5.11 ± 1.85	6.21 ± 2.91	4.05 ± 2.93	6.19 ± 3.19	4.52 ± 3.08		
(mm)										

Tras lo expuesto en este apartado, se puede decir que la TMG es una herramienta que parece detectar la fatiga como bien viene reflejado en los artículos de Cè, Longo, Limonta, Coratella, Rampichini, & Esposito (2020) y Lohr et al. (2019).

A continuación, se muestra una tabla de elaboración propia en la que se detallan los protocolos de fatiga, músculos evaluados y efectos producido en los distintos parámetros de TMG evaluados (Tabla 8).

Estudio fatiga	Tipo de prueba	Músculo	PARÁMETROS TMG	
			Dm	Тс
García-Manso et al. (2011)	Resistencia: Ironman	BF	+	+
Hunter et al. (2012)	5 x 10 contracciones excéntricas en máquina isocinética	BB	-	-
Macgregor et al. (2012)	Programa de estimulación eléctrica	Gm	-	-
Wiewelhove et al. (2015)	Resistencia: ejercicios intermitentes de alta intensidad	RF	+	=

Tabla 8. Respuesta de los parámetros de tensiomografía tras pruebas de resistencia o fuerza para medir la fatiga inducida por dichos protocolos.

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		BF	-	-
Giovanelli et al. (2016)	Resistencia: Maratón cuesta arriba	VL	+	-
	Grupo resistencia: HIIT sobre bicicleta		-	-
Paula Simola et al. (2016)	Grupo fuerza: 1: sentadilla con <i>flywheel</i> ; 2: ejercicios de sobrecarga excéntrica; 3: ejercicios pliométricos; 4: series descendientes; 5: series múltiples	VL	-	-
Raeder et al. (2016)	Fuerza: 1: sentadilla con <i>flywheel</i> ; 2: ejercicios de sobrecarga excéntrica; 3: ejercicios pliométricos; 4: series descendientes; 5: series múltiples	VM	-	=
Franz et al. (2018)	Dos grupos: a) 3x10 al 80% de 1RM		-	=
	b) 3x10 al 80% de 1RM + pre acondicionamiento isquémico	BB	-	-
Harsem et al. (2019)	6x10 repeticiones al 110% de un 1 RM realizando el ejercicio curl de bíceps	BB	-	=
Martín-San Agustín et al. (2020)	Contracciones isométricas al 70% del MVIC	RF	-	=
	32 repeticiones máximas en una máquina	VL	-	-
Muñoz-López et al. (2020)	isocinética de extensión y flexión de rodilla a 180° por segundo.	VM	-	=

BF: bíceps femoral; RF: recto femoral; VL: vasto lateral; BB: bíceps braquial; Gm: Gemelo medial; RM: repetición máxima; MVIC: contracción máxima isométrica voluntaria; +: aumento; -: disminución; = ni baja ni sube; Dm: desplazamiento radial máximo; Tc: tiempo de contracción. Elaboración propia.

7.5.2 Daño muscular

La actividad física, en particular las contracciones musculares excéntricas de alta intensidad, puede producir daños musculares inducidos por el ejercicio (EIMD) (Tanabe, Maeda, Akazawa, Zempo-Miyaki, Choi, Ra, Imaizumi, Otsuka, & Nosaka, 2015). El EIMD produce una respuesta inflamatoria que se asocia con una disminución de la capacidad de generar fuerza muscular, disminución del rango de movimiento (ROM), hinchazón localizada, dolor muscular de aparición retardada (DOMS) y aumento de las proteínas musculares en la sangre (creatina quinasa (CK), lactato deshidrogenasa y mioglobina) (Fatouros & Jamurtas, 2016).

Existen varios estudios que se han centrado en detectar los cambios provocados en las propiedades contráctiles, evaluadas a través de TMG, tras la realización de protocolos excéntricos de alta intensidad. El primer estudio en analizar esta cuestión fue el realizado por Hunter et al. (2012), en el que los 19 participantes en el estudio no podían realizar ningún tipo de ejercicio previo 14 días antes de la ejecución del protocolo. Se tomaron muestras de CK, circunferencia de las extremidades y se realizaron evaluaciones con TMG sobre el BB antes, justo al terminar y 1-2-3-4-5-6 días después de la realización de 5 x 10 extensiones de codo unilaterales excéntricas en una máquina isocinética con 60 segundos de recuperación entre serie. El brazo dominante se tomó como grupo control. Se observó que 24 horas después del protocolo de fatiga, el Dm había bajado un 31% con respecto a los valores basales (p < 0.01). Al séptimo día los valores de Dm se habían recuperado un 17% con respecto a los niveles post, pero aún permanecían muy por debajo de los valores mostrados previos al ejercicio (Figura 15). A su vez, hubo una correlación moderada y positiva entre los cambios de Dm y de MVIC a las 24 horas (r = 0.55, p<0.05). En relación al Tc, éste aumentó un 15% el primer día con respecto a la toma inicial y un 21% el segundo día en relación a la toma inicial. Del mismo modo que la Dm, el TC no había vuelto a sus valores iniciales incluso 6 días después de haber realizado el ejercicio de fuerza.



Figura 15. Desplazamiento radial máximo (Dm) y tiempo de contracción (Tc) en el bíceps braquial a lo largo del estudio. Tomado de Hunter et al. (2012). Se observaron efectos significativos de (p<0.001) los efectos de grupo, tiempo e interacción se observaron para todos los intervalos de tiempo. /**p<0.001. ^tp<0.001 significativamente menor que la medida previa al daño; **p<0.001 [¥]p<0.001 significativamente mayor que la medida previa al daño.</p>

En la misma línea, Franz, Behringer, Harmsen, Mayer, Krauspe, Zilkens, & Schumann (2018) también realizaron un estudio con 19 sujetos divididos en dos grupos: grupo ejercicio excéntrico (GEE, n = 9) y grupo acondicionamiento isquémico más fase excéntrica (GPI + FE, n = 10). Los protocolos consistieron en: GEE 3x10 al 80% de 1RM de la fase concéntrica en el ejercicio curl de bíceps; GPI + FE: tres series de 5 minutos de isquemia, separados por 5

minutos de reperfusión después de cada ciclo más 3x10 al 80% de 1RM de la fase concéntrica en el ejercicio curl de bíceps. Algunos parámetros evaluados fueron: Dm y Tc del BB medidos con TMG y CK. Se realizaron 7 mediciones en el siguiente orden: pre-, post-ejercicio, 20 minutos, 2, 24, 48 y 72 horas post-ejercicio. Se observó un descenso significativo del Dm en ambos grupos (GEE: 11.6 ± 3.9 mm y GPI + FE: 13.8 ± 2.7 mm, *p*<0.05). Por su parte, el Tc bajó significativamente un 16% en el grupo GPI + FE (pre: 24.3 ± 3.2 ms; post 20.9 ± 3.1 ms; *p*<0.05) y un 4% en el grupo GEE (pre: 23.9 ± 4.2 : post 22.9 ± 4 ms). Respecto a la CK, se halló un aumento del 60% en el grupo GPI + FE y de un 221% en el grupo GEE a las 48 horas, siendo las diferencias significativas entre ambos protocolos (*p*<0.005). Los valores no consiguen volver a su estado basal tras 72h en ninguna de las variables evaluadas.

Harmsen, Franz, Mayer, Zilkens, Buhren, Schrumpf, Krauspe, & Behringer (2019), realizaron un estudio sobre el músculo BB en 10 sujetos no entrenados, en concreto se realizaron 6x10 repeticiones excéntricas en el ejercicio de curl de bíceps con el 110% 1RM de la fase concéntrica en dicho ejercicio con 1 minuto de recuperación entre series. Las pruebas que se realizaron fueron: CK y valoraciones con TMG sobre el BB en 5 momentos: preejercicio, post-ejercicio, 20 minutos, 2 horas y tras 72 horas. Tras la realización del ejercicio se observó un descenso en Dm de un 32% (p<0.05) así como un aumento de la CK a las 24 horas de un 6978%, (p<0.05). Además, se encontró una correlación en los cambios entre el Dm y la CK (r = 0.74, p<0.05). Con toda esta información, se podría indicar que la TMG es una herramienta válida como marcador de daño muscular después de u n ejercicio excéntrico. Por lo tanto, la TMG podría representar una alternativa no invasiva y rentable para cuantificar el grado de daño muscular después de intervenciones de este tipo.

7.5.3 Relación entre parámetros tensiomiográficos y rendimiento

En este apartado se van a abordar las relaciones entre los parámetros tensiomiográficos con variables que tienen cierta capacidad explicativa sobre el rendimiento deportivo. Así, en ciclistas profesionales se han encontrado correlaciones entre el Dm del RF y del BF con el consumo máximo de oxígeno (VO₂max) (r = 0.64 y r = 0.68, p<0.05) y del Dm del BF con la potencia aeróbica máxima (r = 0.62, p<0.05) (García-García et al., 2013). Asimismo, Valenzuela et al. (2018), también observaron correlaciones entre las propiedades contráctiles y la potencia medida en jugadoras olímpicas de rugby 7 a través de la prueba Wingate. En

dicho estudio, se observaron correlaciones negativas entre la potencia y los valores de Dm y Td en el VL (r = -0.75 y r = -0.61 respectivamente). Por lo que se puede decir que el Dm podría explicar parte del rendimiento en estos parámetros de importancia en el rendimiento de deportistas profesionales como el ciclismo o el rugby 7.

En la literatura científica existen otros estudios que también se ha centrado en analizar las posibles relaciones de los parámetros evaluados a través de TMG con pruebas de rendimiento indicadoras del rendimiento físico como el "*squat jump*" (SJ), el salto con contramovimiento (CMJ), el *drop jump* (DJ) y el sprint en diferentes distancias: 10-20-30 metros. En este sentido, Loturco et al. (2019) realizaron un estudio con velocistas (n = 15) y saltadores de longitud (n=4) profesionales en el que encontraron correlaciones moderadas entre el DJ (altura del cajón: 45 cm) y la Vc del BF (r = 0.53, p<0.05) y entre la Vc del BF y el índice de fuerza reactiva (tiempo de vuelo/tiempo de contacto) medido durante el DJ (r = 0.68, p<0.05). Además, también se observó una correlación moderada entre la Vc del RF y la potencia media propulsiva en el ejercicio de sentadilla (r = 0.61, p<0.05).

En la misma línea se encuentra el estudio realizado por Gil, Loturco, Tricoli, Ugrinowitsch, Kobal, Abad, & Roschel (2015), esta vez con jugadores profesionales de fútbol y pruebas de velocidad. Los resultados mostraron correlaciones negativas entre el Dm del BF y del RF y el tiempo de contacto durante el DJ (r = -0.50 y r = -0.51; p<0.05, respectivamente).

Un aspecto a destacar en TMG es la variable "sumatorio", es decir, la suma de los músculos evaluados para cada parámetro. Este enfoque permite ver los parámetros de TMG en su conjunto en lugar de forma individual para cada músculo. García-García, Cuba-Dorado, Fernández-Redondo, & López-Chicharro (2018), fueron pioneros en comprobar la relación entre el sumatorio de los parámetros de TMG y pruebas rendimiento. En concreto, cincuenta ciclistas profesionales, participantes en la Copa de España de ciclismo de ruta, realizaron una prueba incremental máxima en un cicloergómetro. En dicho estudio, el protocolo comenzó con 2 minutos de descanso en el cicloergómetro, durante los cuales se obtuvieron muestras de sangre para determinar la concentración de lactato y la frecuencia cardíaca en reposo. Posteriormente, iba aumentando la potencia en 20 W · min⁻¹ mientras se mantenía una frecuencia de pedaleo constante de 80-90 rpm. La prueba continuó hasta que el ciclista era incapaz de mantener el ritmo de pedaleo establecido. Así, en base a una regresión lineal múltiple, se estableció una r² = 0.68 teniendo en cuenta el Tc del RF, el Dm del VM y del VL, la Vrn del BF y el sumatorio del Tc y del Dm del VL, VM, RF, BF.

7.5.4 Valores de referencia de los distintos parámetros tensiomiográficos en diferentes modalidades deportivas, así como adaptaciones de estos ante diferentes intervenciones de entrenamiento

La TMG se ha utilizado para conocer los estados iniciales de los deportistas antes del inicio del periodo competitivo. Así, García-García et al. (2013), fue el primer grupo que trató de establecer valores de referencia en el ciclismo, para a partir de esos valores hacer un seguimiento a lo largo de la temporada. Las pruebas se realizaron en el período preparatorio y en el periodo competitivo. Dichos autores sugirieron que la información obtenida a través de la herramienta no invasiva TMG podría ayudar a proporcionar un asesoramiento adecuado para el diseño y control de las cargas de entrenamiento individualizadas y obtener información del efecto del entrenamiento sobre la fatiga neuromuscular.

Por su parte, García-García, Serrano-Gómez, Hernández-Mendo, & Tapia-Flores (2016), también establecieron valores de referencia con TMG para un equipo profesional de primera división del fútbol español al comienzo de la temporada. Una vez realizada dicha evaluación, durante 10 semanas se llevó a cabo un programa de entrenamiento que consistía en realizar dos veces por semana los siguientes ejercicios: media sentadilla, *leg-press*, multisaltos y propiocepción. Los músculos evaluados fueron: VM, VL, RF y BF y los 5 parámetros de la TMG (Dm, Tc, Td, Ts y Tr). Se observaron descensos significativos en el parámetro Tc del VM (pre: 35.2 ± 5.4 ms, post: 28.7 ± 6.7 ms, *p*<0.05) y del RF (pre: 38.3 ± 3.3 ms, post: 31.5 ± 5.8 ms, *p*<0.05) y también del Dm en el VM (pre: 8.4 ± 1.4 mm, post: 7.2 ± 1.1 mm, *p*<0.05) (Tabla 9).

Tabla 9. Diferencias entre la primera y la segunda medición Dm (desplazamiento muscular); Tc (tiempo de contracción). Pre: antes de empezar el estudio; Post: tras cumplir el periodo de 10 semanas. Tomado de García-García et al. (2016).

Músculo	TMG	PRE	POST	TMG	PRE	POST
VM	Tc (ms)	35.2 ± 5.4	$28.7\pm6.7*$	Dm (mm)	8.4 ± 1.4	$7.2 \pm 1.1*$
VL	Tc (ms)	36.9 ± 4.4	$28.5. \pm 7.2$	Dm (mm)	5.9 ± 1.5	5.5 ± 1.8
RF	Tc (ms)	38.3 ± 3.3	$31.5\pm5.8*$	Dm (mm)	9.8 ± 2.4	8.6 ± 2.4
BF	Tc (ms)	28.9 ± 5.9	29.8 ± 4.6	Dm (mm)	5.3 ± 1.3	6.6 ± 1.9

* Diferencias significativas entre la primera y segunda medición (*p*<0.05)*. VM: Vasto medial; VL: Vasto lateral; RF: Recto femoral; BF: Bíceps femoral.

Por último, Zubac & Simunic (2017), realizaron un programa de entrenamiento basado en multisaltos durante 8 semanas. Dicho estudio contó con un grupo control (n=10) y un grupo experimental (n=10). Se realizaron 3 entrenamientos semanales, comenzando por 5 series de 10 repeticiones y se fue progresivamente incrementando el número de series hasta llegar a 8 series de 10 repeticiones. Se hizo hincapié en saltar lo máximo posible y estar el menor tiempo posible en contacto con el suelo en cada salto. Los saltos eran siempre con ambas piernas y había 2 minutos de descanso entre cada serie. Los músculos evaluados fueron: BF, TA, VL, GL. Los resultados en relación al Tc fueron una disminución significativa en el VL (8.7%; p<0.001), BF (26.7%; p<0.05), TA (32.9%; p<0.05) y GL (25.8%; p<0.05), pero no en el GM (8.1%; p<0.005). En relación al Dm, también se observó una disminución significativa en el BF (26.5%; p = 0.032), GM (14.9%; p<0.05), GL (31.5%; p<0.05), pero no en el TA (16.8%; p<0.005) ni en el VL (6.0%; p<0.005).

7.5.5 Recuperación

El proceso de recuperación guarda particular importancia tras sesiones de entrenamiento y competición (Rattray, Argus, Martin, Northey, & Driller, 2015). La inmersión en agua fría (*"cold water inmersión"* [CWI]) es el método de recuperación más utilizado por los especialistas en las ciencias del deporte, tanto entre los atletas de alto rendimiento como entre los aficionados, que buscan minimizar fatiga y acelerar los procesos de recuperación Calleja-Gonzalez, Mielgo-Ayuso, Sanchez-Ureña, Ostojic, & Terrados, 2019; Calleja-González, Terrados, Mielgo-Ayuso, Delextrat, Jukic, Vaquera, Torres, Schelling, Stojanovic, & Ostojic, 2016).

Por otro lado, en los últimos años, el uso del rodillo de espuma o "foam roller" se ha convertido en una práctica común en todo tipo de entornos deportivos y es muy apreciada en el ámbito de la fuerza y el acondicionamiento físico para aumentar la eficacia del entrenamiento o la preparación para la competición así como para acelerar la recuperación después del ejercicio (Monteiro, Costa, Corrêa Neto, Hoogenboom, Steele, & Silva Novaes, 2019). Asimismo, también se ha expandido rápidamente en la última década la vibración de cuerpo entero ("whole body vibration" [WBV]) en programas de rendimiento y la recuperación de lesiones (Cloak et al., 2010; Gojanovic & Henchoz, 2012). Los investigadores han demostrado que el WBV puede mejorar aspectos del rendimiento físico, como la fuerza (Gual, Fort-

Vanmeerhaeghe, Romero-Rodríguez, & Tesch, 2016) y la flexibilidad (Marshall & Wyon, 2012). Además, también se ha utilizado como un tratamiento de recuperación para reducir el dolor muscular y la pérdida de fuerza después del ejercicio Koh, Cho, Kim, Cho, Kim, & Bo 2013; Kosar, Candow, & Putland, 2012). En este ámbito, la TMG podría ser una herramienta útil para evaluar la efectividad de las diferentes metodologías utilizadas en los procesos de recuperación (Frank et al., 2019; Muñoz-López, et al., 2020; Harmsen et al., 2019; Hunter et al., 2012).

7.5.5.1 Crioterapia

García-Manso, Rodríguez-Matoso, Rodríguez-Ruiz, Sarmiento, de Saa, & Calderón, (2011b) fueron pioneros en el análisis de la influencia del tratamiento con frío sobre los parámetros de TMG. Estos autores (García-Manso et al., 2011b) observaron que repetidas inmersiones en agua fría (4 x 4 minutos a 4°C) causaron considerables alteraciones en las propiedades mecánicas de los músculos. Estas alteraciones afectaron especialmente al Dm y a la Vrd. Los autores postularon que las alteraciones en el Dm podrían deberse a una reducción en la capacidad de transporte de Ca2⁺ y a los cambios producidos en las propiedades viscoelásticas del músculo.

Un segundo estudio que también utilizó la TMG para examinar el efecto de la inmersión sobre las propiedades mecánicas musculares es el de (Sánchez-Ureña, Rojas-Valverde, & Gutiérrez-Vargas, 2018). En este estudio, 39 deportistas fueron divididos en 3 grupos: un grupo de inmersión intermitente en agua fría (GIIAF), un grupo de inmersión continua en agua fría (GICAF) y un grupo de control (GC). Los sujetos realizaron un calentamiento estándar, después un protocolo de fatiga consistente en 8 x 8 CMJ con 90 segundos de recuperación entre series, y posteriormente se sometieron a inmersiones en agua fría. El grupo GIIAF estuvo 12 minutos de forma intermitente, 2 minutos dentro $(12 \pm 0.4^{\circ}C)$ y 1 minuto fuera $(23 \pm 0.5^{\circ}C)$ del agua fría, el grupo GICAF estuvo 12 minutos de inmersión continua a $12 \pm 0.4^{\circ}C$ y los sujetos pertenecientes al grupo control estuvieron 12 minutos sentados en una sala a $23 \pm 0.5^{\circ}C$. Se realizaron 4 mediciones de TMG sobre el BF y el RF: pre, post, 24 h-post y 48 h-post. Se observaron alteraciones de los parámetros medidos con TMG en todos los protocolos sin diferencias entre ellos (Tablas 10 y 11). Así se llega a la conclusión de que tanto protocolos continuos como intermitentes fueron ineficaces para acelerar la recuperación en los parámetros evaluados con TMG tras un protocolo de fatiga con CMJ.

	Grupo control			Immersión continua			Immersión intermitente			р
Parámetro	Pre	24h	48h	Pre	24h	48h	Pre	24h	48h	value
Tc (ms)	28.3 ±4.3	28.9 ± 5.8	29.2 ± 6.3	29.1 ± 4.3	30.6 ± 3.6	30.6 ± 3.1	28.2 ± 3.6	29.1 ± 3.5	29.24 ± 4.53	0.89
Dm (mm)	6.9 ± 1.8	6 ± 1.8	5.9 ± 1.7	7.8 ± 2.6	7.4 ± 2.1	7.9 ± 2.3	7.2 ± 2.9	6.6 ± 2.7	6.71 ± 2.54	0.35

Tabla 10. Comparación del comportamiento de las variables de tensiomiografía evaluadas en el **recto femoral** entre los grupos según el momento de medición. Adaptado de Sánchez-Ureña et al., (2018).

Tc: tiempo de contracción; Dm: desplazamiento muscular; Pre: valores antes del protocolo de saltos; 24h: valores correspondientes a las mediciones realizadas 24 horas post-esfuerzo; 48h: valores correspondientes a las mediciones realizadas 48 horas post-esfuerzo.

Tabla 11. Comparación del comportamiento de las variables de tensiomiografía evaluadas en el **bíceps femoral** entre los grupos según el momento de medición. Adaptado de Sánchez-Ureña et al., (2018).

	Grupo control			Immersión continua			Immersión intermitente			р
Parámetro	Pre	24h	48h	Pre	24h	48h	Pre	24h	48h	value
Tc (ms)	27.8 ± 4.3	29.6 ± 6.8	27.8 ± 7.8	27.4 ± 6.3	27.8 ± 6.1	26.3 ± 5.2	29.5 ± 6.6	28.3 ± 6.8	29.8 ± 8.1	0.39
Dm (mm)	3.01 ± 1.2	2.9 ± 1.3	2.4 ± 1.4	4.1 ± 1.6	4.2 ± 1.6	3.9 ± 1.6	4.1 ± 2	4.2 ± 2.5	4.5 ± 2.4	0.34

Tc: tiempo de contracción; Dm: desplazamiento muscular; Pre: valores antes del protocolo de saltos; 24h: valores correspondientes a las mediciones realizadas 24 horas post-esfuerzo; 48h: valores correspondientes a las mediciones realizadas 48 horas post-esfuerzo.

7.5.5.2 Métodos de recuperación usando vibración de cuerpo entero o masaje con rodillo de espuma

La recuperación del deportista es muy importante, especialmente en deportes donde hay poco tiempo para recuperar. Así en ejercicios intermitentes cortos, realizados en condiciones anaeróbicas, se ha recomendado introducir recuperación activa entre ejercicios en lugar de una recuperación pasiva para disminuir la concentración de lactato en sangre (Billat, Slawinksi, Bocquet, Chassaing, Demarle, & Koralsztein 2001). Sin embargo, y aunque todo parece indicar que la recuperación activa da lugar a una eliminación más rápida de los diferentes metabolitos que la recuperación pasiva, existen dudas sobre cómo estos tipos de recuperación influyen en el rendimiento del ejercicio posterior, probablemente debido a las diferencias metodológicas entre los estudios, especialmente en relación con la tarea utilizada como criterio de rendimiento (Franchini, Moraes Bertuzzi, Takito, & Kiss, 2009). De este modo, el propósito del estudio realizado por Carrasco, Sañudo, De Hoyo, Pradas, & Da Silva (2011), fue evaluar la eficacia de un programa de recuperación usando una plataforma vibratoria como medio de recuperación (5 series de 2 minutos con un minuto de recuperación a una frecuencia de 20 hz) versus un programa de recuperación pasiva (15 minutos sentados en un banco) tras la realización de un esfuerzo de 2 minutos en un cicloergómetro al 100% de su VO_{2 max}. No se encontraron diferencias significativas en el Dm del RF entre ambos medios de recuperación (Vibratoria, pre: 7.57 ± 0.9 mm; post: 4.37 ± 0.67 mm; Recuperación pasiva, pre: 7.86 ± 0.43 mm; post: 4.57 ± 0.58 mm). Por su parte, Murray, Jones, Horobeanu, Turner, & Sproule (2016), trataron de analizar la influencia del *foam roller* en el ROM de los flexores de cadera sobre 12 jugadores de squash entrenados. Estos autores no encontraron diferencias significativas sobre los diferentes parámetros de TMG evaluados (Dm y Tc) en el RF tras la realización de un único ejercicio de 60 segundos con un foam roller (Tc, pre: 27.60 ± 4.14 ms; post: 26.35 ± 5.23 ms; Dm, pre: 8.90 ± 2.50 mm; post: 8.69 ± 2.58 mm). En consecuencia, Murray et al. (2016) concluyeron que el uso del *foam roller* durante 60 segundos no provoca cambios significativos en las propiedades contráctiles del RF.

PLANTEAMIENTO DEL PROBLEMA
8. PLANTEAMIENTO DEL PROBLEMA

Considerando la información descrita en el apartado de "Estado Actual del Conocimiento", parece existir una falta de conocimiento en torno al uso de la TMG a nivel metodológico y de su aplicación en diferentes protocolos de entrenamiento, dándose las condiciones necesarias para justificar la formulación de uno o varios problemas de investigación.

Así, en la literatura científica se ha estudiado cómo afecta el dato recogido con la TMG en relación a la separación entre parches (Wilson et al., 2019a; Tous-Fajardo et al., 2010), ángulos en los que se halla fijada la articulación durante la medición (Ditroilo, Hunter, et al., 2011) y la fiabilidad de la medición (Martín-Rodríguez, et al., 2017a), pero se desconoce cómo afecta el tamaño del parche utilizado o la duración del impulso a las mediciones en TMG.

Asimismo, aunque la TMG es una herramienta capaz de detectar fatiga periférica (Wiewelhove et al., 2021; 2017; Hunter et al., 2012), existen muy pocos estudios en los que se realicen protocolos de fatiga incluyendo tanto medidas de TMG como otros indicadores de rendimiento físico, como el CMJ o la MVIC (Wiewelhove et al., 2021; 2017), especialmente en los que se comparen diferentes protocolos de entrenamiento de fuerza. De tal manera que se hace necesario realizar un estudio donde se incluyan pruebas de rendimiento que son *gold standard* como el CMJ o el MVIC con protocolos de fuerza para ver si la TMG es capaz de detectar las diferencias en los estímulos aplicados en cada entrenamiento.

Un aumento excesivo de la fatiga durante las sesiones de entrenamiento de fuerza puede ser perjudicial para los atletas que se centran en maximizar adaptaciones neuromusculares (Pareja-Blanco et al., 2020; Pareja-Blanco et al., 2017). Por lo tanto, introducir periodos de descanso entre series con menos repeticiones podría ser una estrategia eficaz para atenuar la fatiga y mantener el rendimiento mecánico (es decir, la producción de fuerza, la velocidad de movimiento y, como consecuencia, la potencia mecánica) durante las sesiones de fuerza (Tufano et al., 2017). No obstante, para nuestro conociemiento, solo un estudio ha analizado el efecto de emplear diferentes configuraciones de la serie durante el entrenamiento de fuerza sobre diferentes marcadores de rendimiento físico y las propiedades mecánicas musculares evaluadas con TMG (Tufano et al., 2020). Tufano et al. (2020), comparó un protocolo tradicional que consistió en 4 series de 10 repeticiones con 95 segundos de descanso entre serie en un ejercicio isocinético de extensión de rodilla, con un protocolo usando el mismo ejercicio, pero introduciendo descansos más cortos y más frecuentes: 20 x 2 con 15 segundos de

descanso. No se encontraron diferencias significativas entre ambos protocolos, pero sí se observó una tendencia hacia una ligera subida del Dm con el protocolo de menos repeticiones y una ligera caída del Dm en el protocolo tradicional. Como se puede observar, la información sobre los efectos agudos en protocolos de entrenamiento de fuerza utilizados en la práctica donde se evalúen las propiedades contráctiles es escasa en la literatura. A la luz de estas consideraciones, un conocimiento más detallado a nivel mecánico, neuromuscular y metabólico ante diferentes configuraciones de la serie durante el entrenamiento de fuerza proporcionaría a entrenadores y científicos una mejor comprensión de los efectos en los programas de entrenamiento y podría conducir a nuevos avances en la prescripción del ejercicio.

Por último, existen algunos estudios en los cuales se han medido las propiedades contráctiles con deportistas profesionales (García-García et al., 2018, 2017, 2016; Rodríguez-Matoso et al., 2015), pero hasta el momento sólo un estudio ha realizado mediciones durante la temporada en un equipo profesional de fútbol incluyendo un programa de entrenamiento de fuerza para ver el comportamiento de las propiedades contráctiles (García-García et al., 2016). Los autores concluyen que los parámetros Tc y Dm parecen ser sensibles a los cambios producidos durante un programa de entrenamiento de fuerza. Esto podría ser útil para monotorizar las cargas de entrenamiento. No obstante, se desconoce si estos hallazgos podrían ser extrapolables a otros deportistas de similar nivel pero en diferente deporte como el fútbol-sala.

La problemática general planteada y los objetivos se abordaron a través de 3 problemas de investigación:

Estudio I: ¿Cuál es la influencia del tamaño del electrodo y de la duración del pulso sobre las propiedades contráctiles del músculo medidas con TMG?

Estudio II: ¿Cuáles son las respuestas mecánicas musculares, así como las neuromusculares y metabólicas ante diferentes configuraciones de la serie durante el entrenamiento de fuerza?

Estudio III: ¿Cuál es la evolución de las propiedades contráctiles de los músculos de las extremidades inferiores a lo largo de una temporada en jugadores de fútbol sala de élite? ¿Tendría algún efecto sobre las propiedades mecánicas musculares en estos deportistas la inclusión de un entrenamiento de fuerza?

HIPÓTESIS y OBJETIVOS

9. HIPÓTESIS y OBJETIVOS

Existen numerosos estudios que han utilizado parches de 5 x 5 cm (García-García et al:, 2018, 2017, 2016, 2013) y no tantos que hayan usado parches de 3.2 x 3.2 cm (Loturco et al., 2016). Hasta la fecha, todos los estudios han utilizado parches de 5 x 5 cm para conocer la fiabilidad de los parámetros de TMG (Wilson et al., 2019b; Ditroilo et al., 2013; Tous et al., 2010), pero ninguno se ha centrado en conocer si existen diferencias según el tamaño del parche o la duración del impulso. Es posible que una mayor duración del pulso y un mayor tamaño de parche resulte en un mayor reclutamiento de UMs, lo que puede resultar en una menor variabilidad absoluta y relativa de la medida. Por lo tanto, nuestra hipótesis es la siguiente:

Hipótesis 1: El uso de electrodos de 5 x 5 cm combinado con una duración del impulso de 1 ms sobre el VL y RF permitirá una mayor fiabilidad de los datos que cuando se utiliza un electrodo de 3.2 x 3.2 cm y un impulso de 0.2, 0.5 ó 1 ms.

Objetivo 1: Analizar la influencia de la duración del impulso y del tamaño de los electrodos en TMG sobre la evaluación de las propiedades contráctiles a través de TMG en los músculos VM y RF.

Como se ha comentado en el planteamiento del problema, la información sobre los efectos agudos ante protocolos de entrenamiento de fuerza utilizados en la práctica donde se tenga en cuenta las propiedades contráctiles es escasa en la literatura. Actualmente solo existe un estudio, Tuffano et al. (2020) se centraron en protocolos isocinéticos de extensión unilateral de rodilla. Estos autores observaron una mayor pérdida de fuerza y una tendencia a producir mayores alteraciones en las propiedades mecánicas musculares tras la realización de configuraciones de la serie más largas. Por lo tanto, nuestra hipótesis es la siguiente:

Hipótesis 2: Un protocolo de fuerza de 3 x 8 repeticiones al 75% de1RM con igual número de repeticiones y tiempo total de descanso, provocará una mayor pérdida de fuerza y estrés metabólico, junto con una mayor alteración de las propiedades mecánicas musculares evaluadas con TMG que un protocolo de fuerza de 6 x 4 repeticiones.

Objetivo 2: Investigar la respuesta aguda en las diferentes propiedades mecánicas musculares, estrés metabólico, y fatiga neuromuscular tras la realización de dos entrenamientos de fuerza que diferían en la configuración del grado de esfuerzo dentro de la serie.

Se han observado cambios en las propiedades mecánicas musculares a lo largo de la temporada en diferentes tipos de deportistas (García-García et al., 2016). Además, solo existe un estudio en la literatura científica que haya evaluado en deportistas de élite los efectos que provoca un entrenamiento de fuerza sobre futbolistas en las distintas variables evaluadas con TMG (García-García et al., 2016). Estos autores observaron descensos del Tc y del Dm en el RF, VL y VM en futbolistas tras la realización de un programa de entrenamiento de fuerza. Por lo tanto, nuestra hipótesis es la siguiente:

Hipótesis 3: Jugadores profesionales de fútbol sala experiementarán un deterioro de las propiedades mecánicas de los principales músculos de las piernas durante una temporada. Además, un entrenamiento de fuerza basado en 7 ejercicios, con una duración de 9 semanas y con una periodicidad de 1 vez por semana, producirá atenuaciones en los deterioros observados en las propiedades mecánicas de los principales músculos de las extremidades inferiores durante la temporada en jugadores profesionales de fútbol sala.

Objetivo 3: Observar los cambios en las propiedades contráctiles musculares producidas durante la temporada en jugadores profesionales de fútbol sala y determinar el posible efecto acumulativo de un programa de entrenamiento de fuerza, realizado una vez por semana, sobre dichas propiedades.

ESTUDIO I

10. ESTUDIO I

Artículo I

TITLE: MECHANOMYOGRAPHIC MEASURES OF MUSCLE CONTRACTILE PROPERTIES ARE INFLUENCED BY ELECTRODE SIZE AND STIMULATION PULSE DURATION

Scientific Report

Abstract

The aim was to determine the effects of changing pulse duration and electrode size on muscle contractile properties. Thirty-six healthy young male participated in the study (age 24.8 \pm 5.8 years; height 178.2 \pm 0.6 cm; body mass 71.8 \pm 7.3 kg; self-reported weekly moderate intensity activity 3.5 ± 1.2 h·week⁻¹). Tensiomyography was used to assess rectus femoris (RF) and vastus medialis (VM) muscles neuromuscular properties of the dominant leg according to the electrode size (3.2-5 cm) and the stimulus length (0.2, 0.5, and 1 ms). Maximal radial displacement (Dm); Contraction time (Tc); Delay time (Td); Sustained time (Ts) and Half relaxation time (Tr) were measured. Relative and absolute reliability was quantified. To analyse the effects of the electrode and the stimulus length, a repeated-measures analysis of variance was used. Dm and Tc parameters showed for both muscles an excellent relative (0.95-0.99) and absolute reliability (1.6-4.2%). However, Ts and Tr showed low values of absolute reliability (4.4-40.9%). The duration of the stimulus length applied to the RF and VM and electrode size significantly influences muscle's contractile properties (p < 0.05; $\eta^2_p = 0.09$ -0.60). The Dm increases substantially as the duration of the stimulus increases and with the use of the larger electrode in both muscles. However, Tc and Td are less affected by both conditions and not entirely clear. Practically, our study suggests that a stimulus pulse duration of 1 ms together with a 5×5 cm electrode is necessary to reach a reliable and reproducible assessment of both RF and VM muscles contractile properties.

Keywords: muscle contractile properties, stimulus length, reliability, electrode size

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MECHANOMYOGRAPHIC MEASURES OF MUSCLE CONTRACTILE PROPERTIES ARE INFUENCED BY ELECTRODE SIZE AND STIMULATION PULSE DURATION.

¹Francisco Piqueras-Sanchiz, ²Saúl Martín-Rodríguez, ¹Fernando Pareja-Blanco, ³Luis Baraja-Vegas, ³Jorge Blázquez-Fernández, ⁴Iker J. Bautista & ⁵ÓscarGarcía-García*

¹Physical Performance & Sports Research Center, Universidad Pablo de Olavide, Seville, Spain. ²Department of Physical Education, University of Las Palmas de Gran Canaria, Las Palmas de Gran Canaria, Spain. ³Department of Physiotherapy, Catholic University of Valencia, Valencia, Spain. ⁴Faculty of Physiotherapy and Podology, Catholic University of Valencia, Valencia, Spain. ⁵Laboratory of Sports Performance, Physical Condition and Wellness, Faculty of Education and Sport Sciences, Universidade de Vigo, Pontevedra, Spain

Introduction

Mechanomyography (MMG) is a set of different methods to record mechanical properties, such as muscle's belly displacement and contraction time (Tc), in response to either voluntary or electrically stimulated muscle contraction (Ibitoye et al., 2014) Among the different MMG methods as vibromyography or sonomyography, tensiomyography (TMG) has received great attention in the last decade by the scientific and clinical community (Macgregor et al., 2018). This method has been utilized in a variety of applications, including estimating myosin heavy chain composition (Simunic et al., 2011), determining muscle fibre type populations (Dahmane et al., 2005; Zubac & Simunic, 2017), detecting stiffness or atrophy (Simunic et al., 2019; Pisot et al., 2008), assessing athlete's performance (García-García-García et al., 2019), evaluating muscle fatigue in laboratory or field conditions (Macgregor et al., 2018) or identifying muscle dysfunctions and treatment-effects (Baraja-Vegas et al., 2019; Sánchez-Ureña et al., 2018) among other functions. TMG is one of the MMG techniques that records muscle contractile properties in response to electrically stimulated muscle contractions, which are evoked through the delivery of transcutaneous neuromuscular stimulations (TNS). Most of TMG protocols determine the maximal TNS with a 'current ramp' protocol. Tis methodology entails delivering a series of incremental TNS impulses by increasing amperage (mA) whilst keeping a constant voltage and stimulus length until a maximal muscle contraction is reached – as determined by maximal radial displacement (Dm). Little attention has received in the literature one of the aforementioned variables (i.e. stimulus length) of the TNS protocol. The stimulus length, i.e. the duration of the applied TNS stimulation, influences the magnitude

of the electrical energy delivered to the muscle. Tus, stimulus length influences the amplitude and timing of the TMG waveforms. Unfortunately, a lack of a standardized protocol has led to a variety of TNS stimulus length being reported in humans ranging from 200 µs to 1000 µs in different muscles (Ginz, Zorzato, Iaizzo & Urwyler, 2004; Kimura, Hamada, Massako & Moritani, 2003; Mafuletti et al., 2003). To our knowledge, one of the few studies that have addressed this question on MMG was published more than 10 years ago (McAndrew et al., 2006). These authors tested a wide amount of stimulus length ranging from 50 to 500 µs, at 50-µs increments. They showed that the contractile properties of the muscle were considered stable at pulse durations above 300 µs, which was similar with previously published data for the same muscle (McAndrew et al., 2006). Notably, these authors used a laser MMG sensor while TMG uses a contact-displacement sensor (CDS). In this regard, both laser and CDS offer good-to-excellent reliability, although significant systematic bias was identified with the CDS recording higher mean values (Seidl, Tosovic & Brown, 2017). However, it was previously identified that these differences may not be considered clinically significant. Despite the above, these authors also found that the wide limits of agreement (-19.0ms and 25.2ms) identified between half-relaxation time (Tr) (i.e., the time taken from 90 to 50% of Dm) measures, were considered unreliable from a clinical perspective (Seidl et al., 2017). This finding is consistent with data from a contemporary review on the reliability and measurement error of TMG, which concluded that Tr should not be used for clinical or research purposes (Martín-Rodríguez et al., 2017b). Inter-electrode distance (IED) for electrical stimulation is another key factor in the measurements of any MMG device. Tis parameter has been examined in some studies with several muscles, which have described that IED significantly influences TMG waveform, thus negatively affecting the measure (Wilson et al., 2019b; Wilson et al., 2018; Tous-Fajardo et al., 2010). Despite having studied IED little, the effect of the electrode size on the evoked response has been hardly studied in MMG devices, and only in the field of physical therapy to analyse the thresholds of sensory and motor excitation (Forrester & Petrofsky, 2004; Alon et al., 1994). These studies observed that larger electrodes required greater voltage output but less pulse-charge density than the smaller electrodes. The above, transferred to TMG, means that electrode size could influence the TMG waveform, thus exhibiting different responses of the contractile properties depending on the size. Therefore, it is important to increase the accuracy of TMG assessments while minimizing the measurement error to reach reliable data and compare between studies. It is also important for practitioners for the same issue, so they can be able to accurately and objectively compare several intra- and inter-subjects measurements over time. We hypothesised that the larger the electrode size and stimulus pulse duration

allowed by TMG, the greater the data accuracy and the less the measurement error. Therefore, the aims of this study were: 1) to determine the effect of changing the pulse duration on muscle contractile properties and 2) to discriminate if changing electrode size with different pulse duration affects muscle contractile properties.

Methods

Participants. Thirty-six healthy young and moderately active male volunteers (age 24.8 ± 5.8 years; height 178.2 ± 0.6 cm; body mass 71.8 ± 7.3 kg; self-reported weekly moderate intensity activity 3.5 ± 1.2 h·week⁻¹) who had not suffered muscle or tendon injuries in the previous 6 months participated in the study. The sample size was calculated for each evaluated muscle using the G*POWER software (Heinrich-Heine-Universität Düsseldorf. Germany). The results have showed that for the Dm of the rectus femoris (RF) and vastus medialis (VM), with an alpha of 0.05, a statistical power of 0.80 and an effect size of 0.25, at least 28 and 36 participants were needed respectively. Therefore, a sample size of 36 participants was selected.

Compliance with ethical standards.

All voluntary participants were informed of the research objectives and had the possibility to withdraw at any time from the investigation without any penalty. Informed consent was obtained from all individual participants included in the study. The study was conducted during 4 weeks, according to the Declaration of Helsinki, and the protocol was fully approved by the Ethics Committee of the Catholic University of Valencia (UCV2017-2018-73). The authors declare that they have no conflict of interest.

Experimental design.

A descriptive cross-sectional design was used in order to analyse the effect of the electrode size and the stimulus duration on the parameters obtained with TMG. All participants measurements were made on the same day, in the same room and under the same temperature and humidity conditions $(23.3 \pm 0.6 \,^\circ\text{C}$ and $45 \pm 6.6\%$ respectively) measured with a weather station using an external sensor. The volunteers were instructed to refrain from moderate or heavy physical activities within 72h prior the assessments. Before the data collection, participants were familiar with the electrostimulation stimulus. A 10min period was established lying face up on a stretcher in order to obtain a muscular relaxation state. All subjects were shaved, and the evaluated muscle area was cleaned with alcohol to favour impedance. This was done both in the familiarisation and in the measurement period. New electrodes were used in each measurement using 3.2 or 5 cm self-adhesive circular electrodes. Different measurements were established in the VM and the RF muscles of the dominant leg according to the electrode size $(3.2-5 \,\text{cm})$ and the stimulus length $(0.2, 0.5, \text{ and } 1 \,\text{ms})$, so that 36 measurements were

obtained in each participant, 18 in the RF and 18 in the VM. The total duration of the evaluation was \pm 60 min. The intervention conditions were randomised (see table 12), that is, the muscle evaluation order, the used electrode sequence and the stimulus length was varied. i.e., subject 1 started in the VM with a 5cm electrode and with 1 ms stimulus length, then 5 cm electrode being pulse duration 0.2 ms and finally electrode 5 cm and stimulus length 0.5 ms.

Patch Size		3.2 cm		5 cm							
SUBJECT	Bloc	king sub	jects	SUBJECT	Blocking subjects						
1	А	В	С	1	А	В	С				
2	А	С	В	2	А	С	В				
3	В	А	С	3	В	А	С				
4	В	С	А	4	В	С	А				
5	С	А	В	5	С	А	В				
6	С	В	А	6	С	В	А				

Table 12. Blocking subjects. A: 0.2 milliseconds (ms); B: 0.5 ms; C: 1 ms.

Contractile properties assessment.

TMG was used to assess the neuromuscular properties of both RF and VM muscles of the dominant leg. Measurements were taken under static and relaxed conditions. Prior to performing the measurements, an accurate digital displacement-transducer (GK 40, Panoptik doo, Ljubljana, Slovenia) was perpendicularly positioned at the highest point of the muscle belly. The exact positioning of the electrodes for the RF and VM muscles were in concordance with the recommendations of the SENIAM project (Hermens et al., 2000). RF electrodes were placed at 50% on the line from the anterior superior iliac spine to the superior part of the patella and the VM electrodes were placed at 80% on the line between the anterior superior iliac spine and the joint space in front of the anterior border of the medial ligament. To assure the same electrode placement between the consecutive measurements, the point was marked with a dermatological pen. To elicit the twitch responses, two circular self-adhesive electrodes with difference sizes (3.2 or 5 cm) (Dura-Stick premium, CEFAR-COMPEX, Hannover, Germany) were connected to an electric stimulator (TMG-S1 doo, Ljubljana, Slovenia) and positioned on the muscle surface, following the arrangement of the fibres. Electrodes were placed symmetrically with a IED of approximately 5 cm, placing the positive electrode in the proximal area of the muscle above the measurement point and the negative electrode in the distal area below the measurement point, according to previous investigations (García-García et al., 2016). Both RF and VM muscles were measured in the supine position, with the knee joint

fixed at a 145° knee flexion angle, once again by means of a wedge cushion designed for such purpose. The electrical stimulation was applied with different duration of the stimulus (0.2, 0.5 or 1 ms) with a current amplitude of 100 mA (i.e., single-twitch). Single-twitch is defined as the contractile response to a single electrical impulse and it is a specific type of evoked muscle activity used to characterise the mechanical properties of a muscle or a single motor unit. Although to identify the maximal required stimulus amplitude, and thus the peak muscle response, a progressive incremental approach has been adopted in most studies on the topic (García-García et al., 2019; Macgregor et al., 2018), using in some of them a single-twitch (Rodríguez-Ruiz et al., 2014; García-Manso et al., 2012, 2011). Typically, studies report that the peak response occurs at stimulus amplitudes between 60 and 100 mA, being in larges muscles, such as the lower limbs, much closer to 100mA (Macgregor et al., 2018). Therefore, the decision to use a single-twitch of 100 mA to homogenize the results derived from the different experimental configurations (i.e., electrode size and stimulation pulse duration). A 60 s rest period was allowed between each electrical stimulus to avoid fatigue or post-tetanic activation while a 120 s rest period was established between conditions. All measurements were taken by the same experienced evaluator. In order to ensure the reliability of the TMG assessment, two measurements were taken in each participant in all conditions (stimulus length and electrode size). Between one and two minutes passed between the test-retest of each condition. Each measurement involved recording the following parameters: maximal radial displacement (Dm; mm); contraction time (Tc; ms): Tc as the time from 10% to 90% of Dm; delay time (Td; ms) as the time from onset to 10% of Dm sides of the curve; sustained time (Ts; ms) as the time between 50% of Dm on both the ascending and descending sides of the curve; and Half relaxation time (Tr) was the time from 90% Dm to decline to 50% of the Dm in the relaxation phase (Valencic, & Knez, 1997) (Figure 16).



Figure 16. TMG parameters extracted of displacement-time curve of Vastus Medialis

Statistical analyses.

Normal distribution of all variables was determined by Kolmogorov-Smirnov test, together with the Lilliefors test, in order to verify that the sample distribution was normal, linear and homoscedastic. Relative reliability was quantified by the intraclass correlation coefficient (ICC), along with a 95% confidence interval (CI), respect to the two measurements taken in each participant in all conditions. The ICC was calculated using a two-way-mixed effects and absolute agreement model (McGraw & Wong, 1996). ICC values under 0.50 were rated as low reliability, values between 0.50 and 0.75 indicates moderate reliability, values between 0.75 and 0.90 express good reliability and values greater than 0.90 indicates excellent reliability. Absolute reliability indices were expressed through the standard error of the mean (SEM, SEM%), minimum detectable change (MDC, %MDC) and coefficient of variation (CV) along with the respective 95% CI. The CV was calculated for raw data after being logtransformed (Hopkins, 2000). A CV>10% was interpreted as insufficient absolute reliability (Lohr et al., 2018; Ditroilo et al., 2013; Tous-Fajardo et al., 2010). The SEM is an index for the precision of a measure and reflects the scores consistency within individual subjects (Lohr et al., 2018). The calculation of this index was made according to Weir (2005) as follows equation: (SEM= \sqrt{MSE}), where MSE is the mean square error term from the repeated measures ANOVA. Table 13 and 14.

Table 13 Reliability in tensiomyography parameters for Vastus Medialismuscle (n = 36). Data are mean \pm SD. VM: Vastus Medialis; Dm: displacement; Tc: contraction time; Td: delay time; Tr: half-relaxation time; Ts: sustain time; ms: milliseconds. CV: coefficient of variation; cm: centimeters; ICC: intraclass correlation coefficient; CI: confidence interval; SEM standard error of measurement; MDC: minimum detectable change.

		patch 3.2 cm																
	Stimulus length 0.2							St	s length	0.5			Stimulus length 1					
VM	M±SD	ICC CI 95%	CV %	SEM SEM%	MDC	MDC %	M ± SD	ICC CI 95%	CV %	SEM SEM%	MDC	MDC %	M±SD	ICC CI 95%	CV %	SEM SEM%	MDC	MDC %
Dm (mm)	8.18 ± 1.50	0.97 (0.95- 0.98)	3.0	0.24 ± 2.95	0.67	8.22	8.50 ± 1.51	0.97 (0.95- 0.98)	3.2	0.26 ± 3.10	0.73	8.62	8.60 ± 1.36	0.97 (0.95- 0.98)	3.1	0.23 ± 2.74	0.65	7.62
Tc (ms)	22.52 ± 2.1	0.95 (0.91- 0.97)	2.2	0.49 ± 2.19	1.37	6.09	$\begin{array}{c} 22.17 \pm \\ 2.04 \end{array}$	0.95 (0.91- 0.97)	2.3	0.47 ± 2.11	1.30	5.87	22.21 ± 1.94	0.95 (0.90- 0.97)	2.2	0.43 ± 1.97	1.21	5.46
Td (ms)	22.38 ± 1.25	0.85 (0.75- 0.91)	2.3	0.50 ± 2.23	1.39	6.21	22.23 ± 1.15	0.88 (0.81- 0.94)	1.9	0.39 ± 1.77	1.09	4.92	22.24 ± 1.21	0.87 (0.79- 0.93)	2.2	0.45 ± 2.02	1.25	5.62
Tr (ms)	67.1 ± 39.49	0.76 (0.62- 0.86)	23.1	21.25 ± 31.66	58.92	87.80	75.95 ± 47.31	0.85 (0.75- 0.91)	28.7	19.77 ± 26.03	54.81	72.16	71.09 ± 43.61	0.76 (0.62- 0.86)	36.6	23.69 ± 33.32	65.66	92.37
Ts (ms)	168.6 ± 28.6	0.92 (0.87- 0.96)	4.4	8.10 ± 4.80	22.46	13.32	$\begin{array}{c} 180.5 \pm \\ 27.96 \end{array}$	0.91 (0.86- 0.95)	6.2	8.29 ± 4.59	23.00	12.74	$\begin{array}{c} 175.6 \pm \\ 26.84 \end{array}$	0.90 (0.83- 0.94)	5.4	9.38 ± 5.34	26.01	14.81

		patch 5 cm																
		Stimulus length 0.2						Stimulus length 0.5						Stimulus length 1				
VM	M±SD	ICC CI 95%	CV %	SEM SEM%	MDC	MDC %	M ± SD	ICC CI 95%	CV %	SEM SEM%	MDC	MDC %	M±SD	ICC CI 95%	CV %	SEM SEM%	MDC	MDC %
Dm (mm)	8.65 ± 1.34	0.97 (0.94- 0.98)	3.0	0.25 ± 2.89	0.69	8.04	8.67 ± 1.36	0.96 (0.93- 0.98)	2.9	0.31 ± 3.65	0.88	10.16	8.67 ± 1.30	0.95 (0.91- 0.97)	3.8	$\begin{array}{c} 0.26 \pm \\ 3.06 \end{array}$	0.73	8.51
Tc (ms)	$\begin{array}{c} 21.90 \pm \\ 1.98 \end{array}$	0.97 (0.95- 0.98)	1.6	$\begin{array}{c} 0.33 \pm \\ 1.53 \end{array}$	0.93	4.25	$\begin{array}{c} 21.50 \pm \\ 1.97 \end{array}$	0.95 (0.92- 0.97)	2.0	$\begin{array}{c} 0.43 \pm \\ 2.01 \end{array}$	1.20	5.60	$\begin{array}{c} 21.77 \pm \\ 2.01 \end{array}$	0.95 (0.91- 0.97)	2.4	0.47 ± 2.16	1.30	5.99
Td (ms)	22.21 ± 1.22	0.93 (0.88- 0.96)	1.6	0.33 ± 1.49	0.92	4.15	$\begin{array}{c} 22.08 \pm \\ 1.22 \end{array}$	0.90 (0.83- 0.94)	1.6	0.40 ± 1.84	1.12	5.11	22.06 ± 1.09	0.88 (0.81- 0.93)	1.7	$\begin{array}{c} 0.39 \pm \\ 1.78 \end{array}$	1.09	4.96
Tr (ms)	81.93 ± 47.42	0.85 (0.76- 0.91)	38.1	$\begin{array}{c} 22.69 \pm \\ 27.69 \end{array}$	62.90	76.77	$\begin{array}{c} 75.37 \pm \\ 48.70 \end{array}$	0.89 (0.81- 0.94)	25.8	19.62 ± 26.03	54.38	72.15	62.76 ± 40.17	0.80 (0.68- 0.88)	34.1	21.77 ± 34.68	60.34	96.15
Ts (ms)	175.41 ± 28.64	0.88 (0.81- 0.93)	6.6	10.61 ± 6.04	29.42	16.77	182.65 ± 30.24	0.89 (0.81- 0.94)	5.5	8.36 ± 4.58	23.19	12.69	$\begin{array}{c} 182.58 \\ \pm \ 28.43 \end{array}$	0.90 (0.84- 0.95)	5.7	9.09 ± 4.98	25.20	13.80

The %SEM, which represents the relative amount of measurement error, was calculated according to Wagner et al. (2008) as follows equation: (SEM%=SEM/M × 100), being M the mean of the three TMG measurements. The MDC was calculated as equation: (SEM×1.96× $\sqrt{2}$) (Weir, 2005). MDC% was expressed as equation: (MDC/M × 100), where M is the mean of the three TMG measurements. To analyse the effect of the electrode size (i.e., 3.2 vs. 5 cm) and the stimulus length (i.e., 0.2, 0.5 and 1ms), an analysis of the variance of repeated measures (2 × 3 ANOVA) was used. Bonferroni post hoc test with adjustment for 95% confidence interval was used to compare the main effects and identify significant individual differences. The effect sizes in repeated measures ANOVA were reported as partial eta square (η^2) and interpreted as small (0.01), moderate (0.06), or large (0.14) (Cohen, 1988). An alpha level of *p*<0.05 was considered statistically significant. All data were analysed using SPSS v24.0 for Windows (SPSS Inc., Chicago, IL, USA).

Results

The Dm of the VM moderately increased ($\eta^2 p = 0.089$) as the stimulus length increased, although only significantly between the duration of 0.2 and 0.5 ms (8.41 vs 8.58 mm, 2.0%, p = 0.025) (Figure 17). The Dm of the RF also increased as the stimulus length increased with a large effect size ($\eta 2 p = 0.606$) (Figure 18). In addition, it also increased 10.4% (p = 0.001) between 0.2 and 0.5ms duration, 7.0% (p = 0.001) between 0.5 and 1ms and 18.2% (p = 0.001) between 0.2 and 1 ms.

Table 14 Reliability in tensiomyography parameters for Rectus Femoris muscle (n = 36). Data are mean \pm SD. RF: Rectus Femoris; Dm: displacement; Tc: contraction time; Td: delay time; Tr: half-relaxation time; Ts: sustain time; ms: milliseconds. CV: coefficient of variation; cm: centimeters; ICC: intraclass correlation coefficient; CI: confidence interval; SEM standard error of measurement; MDC: minimum detectable change.

					-	-		pat	ch 3.2	cm		-						
		Stimulus length 0.2						St	imulu	s length	0.5			Stimulus length 1				
RF	M±SD	ICC CI 95%	CV %	SEM SEM%	MDC	MDC %	M ± SD	ICC CI 95%	CV %	SEM SEM%	MDC	MDC %	M±SD	ICC CI 95%	CV %	SEM SEM%	MDC	MDC %
Dm (mm)	7.63 ± 2.10	0.98 (0.97- 0.99)	4.1	0.28 ± 3.77	0.79	10.46	8.6 ± 2.51	0.98 (0.97- 0.99)	3.7	$\begin{array}{c} 0.31 \pm \\ 3.68 \end{array}$	0.88	10.23	9.23 ± 2.19	0.98 (0.97- 0.99)	2.9	0.28 ± 3.09	0.79	8.59
Tc (ms)	26.14 ± 4.79	0.98 (0.96- 0.99)	3.0	0.67 ± 2.58	1.87	7.15	26.38 ± 4.77	0.98 (0.97- 0.99)	2.4	0.57 ± 2.16	1.58	6.00	26.95 ± 4.67	0.98 (0.97- 0.99)	2.4	0.57 ± 2.12	1.59	5.90
Td (ms)	$\begin{array}{c} 24.50 \pm \\ 1.96 \end{array}$	0.94 (0.90- 0.97)	2.3	0.49 ± 2.01	1.35	5.58	$\begin{array}{c} 24.82 \pm \\ 1.90 \end{array}$	0.96 (0.93- 0.98)	1.8	0.40 ± 1.61	1.08	4.46	24.95 ± 1.93	0.93 (0.88- 0.96)	2.7	0.53 ± 2.14	1.48	5.96
Tr (ms)	73.77 ± 39.12	0.86 (0.77- 0.92)	26.2	$\begin{array}{c} 14.92 \pm \\ 20.23 \end{array}$	41.37	56.09	65.21 ± 40.77	0.90 (0.84- 0.95)	31.1	13.26 ± 20.34	36.77	56.38	$\begin{array}{c} 73.52 \pm \\ 52.98 \end{array}$	0.96 (0.92- 0.97)	18.8	$\begin{array}{c} 12.00 \pm \\ 16.32 \end{array}$	33.26	45.24
Ts (ms)	117.77 ± 44.88	0.87 (0.78- 0.92)	21.2	$\begin{array}{c} 16.70 \pm \\ 14.18 \end{array}$	46.29	39.35	$\begin{array}{c} 105.07 \\ \pm \ 44.86 \end{array}$	0.91 (0.86- 0.95)	18.2	13.66 ± 13.00	37.87	36.04	109.3 ± 52.25	0.96 (0.92- 0.97)	12.7	12.54 ± 11.47	34.77	31.79

								pa	tch 5	cm								
		Stimulus length 0.2						Stimulus length 0.5						Stimulus length 1				
RF	M±SD	ICC CI 95%	CV %	SEM SEM%	MDC	MDC %	M±SD	ICC CI 95%	CV %	SEM SEM%	MDC	MDC %	M±SD	ICC CI 95%	CV %	SEM SEM%	MDC	MDC %
Dm (mm)	8.67 ± 2.24	0.98 (0.97- 0.99)	4.2	$\begin{array}{c} 0.35 \pm \\ 4.10 \end{array}$	0.98	11.39	9.53 ± 2.16	0.98 (0.96- 0.99)	4.2	0.31 ± 3.29	0.87	9.15	$\begin{array}{c} 10.04 \pm \\ 2.23 \end{array}$	0.98 (0.96- 0.99)	3.5	$\begin{array}{c} 0.33 \pm \\ 3.37 \end{array}$	0.93	9.36
Tc (ms)	$\begin{array}{c} 26.25 \pm \\ 4.81 \end{array}$	0.97(0.95- 0.98)	2.7	0.87 ± 3.33	2.42	9.24	26.19 ± 4.22	0.98 (0.96- 0.99)	2.6	0.61 ± 2.34	1.70	6.50	$\begin{array}{c} 26.30 \pm \\ 4.12 \end{array}$	0.99 (0.98- 0.99)	2.0	0.45 ± 1.72	1.26	4.79
Td (ms)	$\begin{array}{c} 24.79 \pm \\ 1.96 \end{array}$	0.93 (0.84- 0.96)	2.0	0.53 ± 2.15	1.47	5.96	25.14 ± 1.77	0.94 (0.89- 0.96)	2.0	$\begin{array}{c} 0.45 \pm \\ 1.80 \end{array}$	1.25	5.00	$\begin{array}{c} 25.36 \pm \\ 1.87 \end{array}$	0.94 (0.90- 0.97)	2.3	$\begin{array}{c} 0.46 \pm \\ 1.81 \end{array}$	1.27	5.04
Tr (ms)	75.64 ± 49.14	0.83 (0.73- 0.90)	33.0	$\begin{array}{c} 23.37 \pm \\ 30.89 \end{array}$	64.79	85.66	69.34 ± 53.41	0.82 (0.71- 0.90)	40.9	$\begin{array}{c} 28.75 \pm \\ 41.46 \end{array}$	79.70	114.9 4	$\begin{array}{c} 64.96 \pm \\ 51.58 \end{array}$	0.94 (0.89- 0.96)	22	${19.68 \pm \atop 30.29}$	46.26	71.21
Ts (ms)	$\begin{array}{c} 110.95 \\ \pm \ 45.82 \end{array}$	0.87 (0.79- 0.93)	20.3	$\begin{array}{c} 17.10 \pm \\ 15.41 \end{array}$	47.40	42.72	102.17 ± 52.92	0.83 (0.73- 0.90)	22.9	$\begin{array}{c} 26.90 \pm \\ 26.32 \end{array}$	74.56	72.98	97.92 ± 51.98	0.94 (0.89- 0.96)	14.2	16.94 ± 17.29	46.97	47.97

The use of the largest electrode (5 cm) caused a greater Dm than using the smaller one (3.2 cm) in both the VM (2.8%, p = 0.036) and RF (10.8%, p = 0.001). (Fig. 17-18). In addition, the interaction stimulus length x electrode size moderately ($\eta 2 p = 0.117$) modulated the Dm of the VM, which occurred with all stimulus length (8.18–8.65mm at 0.2ms; 8.50–8.67 mm at 0.5ms; 8.60–8.67 mm at 1ms; p = 0.013). The Td of the VM was only significantly affected by the size of the electrode. The use of a larger electrode (5 cm) caused a lower Td (22.28 vs

22.11ms, 0.7%, p = 0.023). However, the Td of the RF increased as the stimulus length increases with a large effect size ($\eta 2 p = 0.237$). It increased by 1.4% (p = 0.006) between 0.2 and 0.5ms duration, 0.7% (p = 0.036) between 0.5 and 1 ms and 2.1% (p = 0.001) between 0.2 and 1 ms. In addition, the electrode size also modulated the Td of the RF with a large effect size ($\eta 2 p = 0.327$), so that the use of the largest electrode (5 cm) caused longer Td (1.4%, p = 0.001). Table 15.

Discussion

Summarizing, the results of this study show that the stimulus length (duration of the TNS pulse) applied to the RF and VM muscles and the size of the electrode significantly influence the muscle's contractile properties as measured by a single-twitch TMG technique. The main finding is that Dm significantly increases both, as the duration of the TNS increases and with the use of a larger electrode (5 cm) in both muscles. However, Tc and Td are less affected by both conditions and not entirely clear. Our results are consistent with previous reviews on the topic showing that Tr parameter should not be used in research or clinical environments due to their insufficient absolute reliability and high measurement error (Macgregor et al., 2018; Martín-Rodríguez et al., 2017a). Similarly, Ts parameter should not be used in these areas since it has shown a CV higher than 10% (Tous-Fajardo et al., 2012). On the contrary, Dm, Tc and Td parameters have shown a high relative reliability in all muscles and situations evaluated, which is in line with the findings of Lohr et al. (2019), but also an absolute reliability through a low CV, %SEM and %MDC, as opposed to the results of Lohr et al. (2019), which indicates a discordant absolute reliability in the Dm and moderate positive in the Tc. Hence, the effect of stimulus length and electrode size on Tr and Ts was not examined, since these parameters did not meet the reliability requirements. The electrical energy available to stimulate the muscle is influenced by the duration of the applied electrical impulse as shown by the equation (Alon et al., 1994): $E = V \times I \times \Delta t$, where E is the electrical energy, V are volts, I equals current and Δ t is the duration of the TNS pulse. It appears that, at stimulus length below 1ms, there is insufficient electrical energy available to maximally stimulate all motor units (Bickel, Gregory & Dean, 2011). Moreover, given that smaller motor units are easier to stimulate than larger motor units (Milner-Brown, Stein & Yemm, 1973; Henneman, Somjen & Carpenter, 1965), shorter stimulus length appear to favour type I slow twitch muscle fibers over type II fast twitch muscle fibers, which are more difficult to activate (Bickel et al., 2011). Our results match those found by McAndrew et al. (2004), who reported that stimulus length above 0.3 ms provide both a maximal lateral displacement of the muscle's belly and stable measures.

	EFFECT	F	Df	Р	η^2
	stimulus length*	6.812	2	.002	0.163
Tc VM	electrode size *	27.244	1	.001	0.438
	stimulus length x electrode size	1.341	2	.268	0.037
Tc RF	stimulus length	1.526	2	.225	0.042
	electrode size	0.836	1	.367	0.023
	stimulus length x electrode size	1.774	2	.177	0.048
Dm VM	stimulus length*	3.418	2	.038	0.089
	electrode size *	4.757	1	.036	0.120
	stimulus length x electrode size *	4.628	2	.013	0.117
Dm RF	stimulus length*	53.793	2	.001	0.606
	electrode size *	49.926	1	.001	0.588
	stimulus length x electrode size	0.675	2	.513	0.019
Td VM	stimulus length	2.041	2	.138	0.055
	electrode size *	5.694	1	.023	0.140
	stimulus length x electrode size	0.082	2	.922	0.002
Td RF	stimulus length*	10.865	2	.001	0.237
	electrode size *	17.002	1	.001	0.327
	stimulus length x electrode size	0.386	2	.681	0.011

Table 15. Effects of different stimulus length and electrode size on different tensiomography parameters. Interactions between stimulus length (0.2, 0.5 and 1 ms), and electrode size (3.2 vs. 5 cm). p < 0.05



Figure 17. Displacement-time curves of Vastus Medialis of the dominant leg, according to the electrode size (3.2–5 cm) and the stimulus length (0.2, 0.5, and 1ms).

RECTUS FEMORIS



Figure 18. Displacement-time curves of Rectus Femoris of the dominant leg, according to the electrode size (3.2–5 cm) and the stimulus length (0.2, 0.5, and 1ms).

of its contractile properties. Although the previous authors used a laser-based MMG technique, their results can be extrapolated to ours since it has been demonstrated that both laser and CDS shows good-to-excellent reliability for the assessment of muscle contractile properties with no significant differences between them (Seidl et al., 2017). In fact, it should be noted that there are data showing that the radial muscle displacement (Dm) increases linearly with muscle torque up to 68% of maximal voluntary contraction (Pisot et al., 2008). This fact may suggest that, maintaining linearity, using shorter pulse duration (0.2ms) could elicit low torque production, affecting to the wave-form and then to all the TMG parameters.

To our knowledge, to date there has been no study that has analysed the effects of the electrodes size for muscle electrical stimulation measured by TMG. However, it has been previously analysed in transcutaneous electrical nerve stimulation (TENS) (Alon, 1995). This author used 4 types of electrodes: 3×3 , 6×6 , 9×9 and 5×6 cm on the quadriceps with the objective of examining the effects of electrode size at neural and motor level, reaching the conclusion that the larger the electrode, the greater the participation of motor units. Our results seem to be in agreement with those obtained by Alon (1995), with respect to the electrode size, since both VM and RF increased their Dm with the largest electrode. A reasonable explanation is that a larger electrode is able to recruit more motor units on the transverse axis and therefore greater amount of muscle mass displacement directly influences an increase in Dm. However, this hypothesis will still have to be proven. On the other hand, the overall %SEM and %MDC results of both muscles seem to point out, in a general way, that increasing electrode size and pulse duration could increase the data accuracy and minimize the measurement error, although it would be necessary more research with other muscle groups to verify this point. A priori, to minimize measurement errors with TMG parameters, it should be recommended standardizing TMG protocols to develop a TMG standard operating procedure such that experimental studies

may be comparable. In order to do this, based on the present fndings and previous research, it is recommended: (1) to take into account the correct stimulus length (1ms) to maximize Dm; (2) to use electrodes of 5 cm; (3) to use specifc guides for each muscle of inter-electrode distance (Wilson, et al 2019b; Wilson et al., 2018) and (4) additional recommendations for a correct measurement protocol are described in the review of Macgregor et al. (2018). In this line, Lohr et al. (2019), have indicated that it is necessary to have high methodological standards for conduct and reporting TMG studies, and these recommendations could be helpful for this purpose.

Limitations, strengths and practical applications

Finally, it is important pointing that, the maximal TNS voltage required to produce peak lateral displacement of a muscle's belly is unique to each muscle and is determined as the voltage that first produces an undistorted parabolic MMG waveform of maximal amplitude¹⁴. In this regard, we used a unique intensity (100 mA) in our study to be able to homogenize and compare subjects and muscles with each other. This was done because the current and well accepted ramp protocol (García-García et al., 2019; Macgregor et al., 2018), used to decide individual intensity would have made it difficult to compare results between subjects' muscles. This could have been solved by normalizing the values of each parameter of each subject/muscle to, for example, a maximum voluntary contraction. However, we did not have the necessary tools for this purpose, what is a limitation of this study. In addition, a duration of TNS above 1ms could also have been used to try to stimulate larger motor units, but the TMG software did not allow this when the experiment was performed. Furthermore, it has only been evaluated two muscles, both of the lower limb, which may be another limitation of this study. On the other hand, the main strength of the present study is the sample size and the use of several set configurations for both electrode size and stimulus pulse duration, and as well as that the same evaluator with more of 7 years' experience took all measurements. In terms of practical applications, TMG researchers and practitioners can base their measurement protocol on the findings of this study. Our data indicates that a stimulation pulse duration of 1ms together with the election of a 5×5 cm electrode size is necessary to reach a reliable and reproducible assessment of both RF and VM muscles contractile properties. Conversely, the use of a smaller electrode or a stimulus length of less than 1ms would be a risk to guarantee the reproducibility of the measurement taken with TMG.

Conclusions

The duration of the stimulus length applied to the RF and VM muscles and electrode size significantly influence the muscle's contractile properties as measured by a single-twitch TMG technique. In fact, the Dm increases substantially as the duration of the TNS increases and with the use of the larger electrode (5cm) in both muscles. However, Tc and Td are less affected by both conditions and not entirely clear. Therefore, this study indicates that a stimulus pulse duration of 1ms together with the election of a 5×5 cm electrode size is necessary to reach a reliable and reproducible assessment of both rectus and vastus medialis muscles contractile properties.

ESTUDIO II

11. ESTUDIO II

Artículo II

TITLE: ACUTE MECHANICAL, NEUROMUSCULAR AND METABOLIC RESPONSES TO DIFFERENT SET CONFIGURATIONS IN RESISTANCE TRAINING

Journal of Strength Conditioning Research

Abstract

The aim of this study was to investigate the effect of set configuration on mechanical performance, neuromuscular activity, metabolic response, and muscle contractile properties. Sixteen strength-trained men performed 2 training sessions in the squat exercise consisting of (a) 3 sets of 8 repetitions with 5 minutes rest between sets (3×8) and (b) 6 sets of 4 repetitions with 2 minutes rest between sets (6×4) . Training intensity (75% one repetition maximum), total volume (24 repetitions), total rest (10 minutes), and training density were equalized between protocols. A battery of tests was performed before and after each protocol: (a) tensiomyography (TMG), (b) blood lactate and ammonia concentration, (c) countermovement jump, and (d) maximal voluntary isometric contraction in the squat exercise. Force, velocity, and power output values, along with electromyography data, were recorded for every repetition throughout each protocol. The 6×4 protocol resulted in greater mechanical performance (i.e., force, velocity, and power) and lower neuromuscular markers of fatigue (i.e., lower root mean square and higher median frequency) during the exercise compared with 3×8 , particularly for the last repetitions of each set. The 3×8 protocol induced greater lactate and ammonia concentrations, greater reductions in jump height, and greater impairments in TMG-derived velocity of deformation after exercise than 6×4 . Therefore, implementing lower-repetition sets with shorter and more frequent interset rest intervals attenuates impairments in mechanical performance, especially in the final repetitions of each set. These effects may be mediated by lower neuromuscular alterations, reduced metabolic stress, and better maintained muscle contractile properties.

Keywords: rest redistribution, lactate, ammonia, rate of force development, electromyography, tensiomyography.

Link <u>Acute Mechanical</u>, Neuromuscular, and <u>Metabolic Responses to...</u> : The Journal of <u>Strength & Conditioning Research (lww.com)</u>

ACUTE MECHANICAL, NEUROMUSCULAR, AND METABOLIC RESPONSES TO DIFFERENT SET CONFIGURATIONS IN RESISTANCE TRAINING

¹Francisco Piqueras-Sanchiz, ¹Pedro J. Cornejo-Daza,¹⁻²Juan Sánchez-Valdepeñas, ³Beatriz Bachero-Mena, ³Miguel Sánchez-Moreno, ⁴Saúl Martín-Rodríguez, ⁵Oscar García-García & ^{1,2}Fernando Pareja-Blanco*

¹Department of Sports and Computers Sciences, Physical Performance & Sports Research Center, Pablo de Olavide University, Seville, Spain; ²Department of Sports and Computers Sciences, Faculty of Sport Sciences, Pablo de Olavide University, Seville, Spain; ³Department of Physical Education and Sports, University of Seville, Seville, Spain; ⁴Department of Physical Education, University of the Gran Canarian Palms, The Gran Canarian Palms, Spain; and ⁵Laboratory of Sports Performance, Physical Condition and Wellness, Faculty of Education and Sport Sciences, University of Vigo, Pontevedra, Spain

Introduction

Fatigue can be defined as an integral process resulting in a temporal decline in force production capacity (Cairns, Knicker, Thompson, & Sjogaard, 2005). A better understanding of the mechanical and physiological mechanisms underlying fatigue development during resistance training (RT) sessions is essential to improve our knowledge of strength training methodology. Excessive fatigue development during training sessions may be detrimental for athletes focused on maximizing neuromuscular adaptations (Parejo-Blanco et al., 2020; 2017). Shorter set configurations that include rest periods between clusters of repetitions are an effective strategy to attenuate fatigue and maintain mechanical performance (i.e. force production, movement velocity, and, as a consequence, power output) during RT sessions (Tufano, Conlon, Nimphius, Oliver, Kreutzer, & Haff, 2019). In addition, higher blood lactate (González-Hernández, García-Ramos, Castaño-Zambudio, Capelo-Ramírez, Marquez, Boullosa, & Jiménez-Reyes, 2020; García-Ramos, González-Hernández, Baños-Pelegrín, Castaño-Zambudio, Capelo-Ramírez, Boullosa, Haff, & Jiménez-Reyes, 2020; Mora-Custodio, Rodríguez-Rosell, Yáñez-García, Sánchez-Moreno, Pareja-Blanco, & González-Badillo, 2018) and ammonia (Morán-Navarro, Pérez, Mora-Rodríguez, de la Cruz-Sánchez, González-Badillo, Sánchez-Medina, & Pallarés, 2017). concentrations, hormonal response (growth hormone and cortisol) (Tufano, Conlon, Nimphius, Oliver, Kreutzer, & Haff, 2019); Moran-Navarro et al., 2017; Oliver, Kreutzer, Jenke, Phillips, Mitchell, & Jones, 2015), and muscle damage indicators (i.e. creatine kinase) (Moran-Navarro et al., 2017) have been

observed following longer set configurations.

Previous literature analyzing set configuration has focused on the effects on exercise performance itself, with limited studies examining the post-exercise responses to different set configurations. In this regard, longer set configurations are characterized by inducing greater impairments in back squat (SQ) performance (i.e. velocity attained against a given absolute load) and jump height (Mora-Custodio et al., 2018; Moran-Navarro et al., 2017). However, the acute effect of set configuration on relevant markers for the evaluation of fatigue in strength training as maximal isometric force (MIF) or maximal rate of force development (RFDmax) remains unexplored. Likewise, recording electromyography (EMG) activity (amplitude, via root mean square "RMS", and frequency, via median frequency "MDF") may provide a better understanding of the mechanisms behind the changes in mechanical performance, such as muscle activation and neuromuscular fatigue accumulated throughout the training session (6). To date, only one previous study has examined neuromuscular activity during different set configurations involving lower-body muscles (Morales-Artacho et al., 2019), which compared 6 sets of 6 repetitions at 20% of one-repetition maximum (1RM) in loaded jumps, continuously (n = 9) or with a 30-second rest every 2 repetitions (n = 9). These authors observed larger increments in RMS throughout longer set configurations; however, both protocols induced similar decrements in MDF (Morales-Artacho, Garcia-Ramos, Perez-Castilla, Padial, Gomez, Peinado, Perez-Cordoba, & Feriche, 2019). However, the fact that different subjects performed each protocol may have obscured potential differences in the EMG spectral parameters between protocols. Lastly, EMG recordings have limitations during dynamic muscle contractions (Enoka, & Duchateau, 2016). Therefore, it would be reasonable to examine the effects of set configuration on neuromuscular fatigue development during both dynamic and isometric contractions.

Tensiomyography (TMG) has been validated for assessing in-vivo passive muscle contractile properties through simple measurement of the muscle belly radial deformation and the time it takes to occur in response to a single-twitch stimulus (Valencic, & Knez, 1997). This technique allows assessment of the changes in muscle contractile properties induced by fatigue after training sessions (Pereira, Ramirez-Campillo, Martin-Rodriguez, Kobal, Abad, Arruda, Guerriero, & Loturco, 2019; Piqueras-Sanchiz, Martin-Rodriguez, Martinez-Aranda, Lopes, Raya-Gonzalez, Garcia-Garcia, & Nakamura, 2019; de Paula Simola et al., 2015). Despite evidence indicating TMG as a valid and reproducible tool to screen adjustments in skeletal muscle contractile characteristics (Martin-Rodriguez et al., 2017a), to our knowledge, only one study has analyzed the acute effects on TMG outcomes following different set

configurations (Tufano, Omcirk, Malecek, Pisz, Halaj, & Scott, 2020). These authors compared 4 isokinetic unilateral knee extension protocols, 2 different set configurations (4x10 with 95 s of inter-set rest vs. 20x2 with 15 s inter-set rest) at 2 different velocities (60 vs. $360 \circ s^{-1}$), reporting similar post-exercise TMG parameters patterns for all protocols (Tufano et al., 2020). The fact that this study was conducted on an isokinetic device improved the control of confounding variables, but sacrificed the ecological validity of real-life resistance exercises. Therefore, information about the acute effects of set configuration in strength training protocols used in practice on muscle contractile properties is lacking in the literature. In this regard, the SQ is one of the most widely used and effective RT exercises for strengthening the lower limbs and improving athletic performance (Hartmann, Wirth & Klusemann, 2013). In addition, TMG measurements should be accompanied by post-exercise mechanical performance tests in order to better comprehend how these outcomes interact. In light of these considerations, a more detailed knowledge about the integral response (mechanical, neuromuscular and metabolic) to different set configurations would provide coaches and scientists with a better understanding of the effects of set configuration manipulation and could lead to further advances in exercise prescription. Therefore, the purpose of this study was to investigate the effect of set configuration on mechanical performance and neuromuscular activity throughout the SQ training session, as well as the acute mechanical, neuromuscular, metabolic and muscle contractile responses for strength-trained men.

Methods

Experimental Approach to the Problem

A randomized cross-over research design was undertaken to examine the acute mechanical, neuromuscular, metabolic, and muscle contractile properties responses to two different set configurations with a load of 75% 1RM during the SQ exercise: 1) 3 sets of 8 repetitions with 5 min rest between sets (3x8); 2) 6 sets of 4 repetitions with 2 min rest between sets (6x4). Training intensity (75% 1RM), total volume (24 repetitions), total rest (10 min) and, as a consequence, training density (work to rest ratio) were equalized between protocols. Protocols were performed in a random order, separated by a period of 4 days.

In order to compare the mechanical, neuromuscular, metabolic, and muscle contractile properties response, subjects underwent a battery of tests before and after each protocol: 1) TMG measurements; 2) blood lactate and ammonia concentration; 3 countermovement jumps (CMJ); and 4) maximal voluntary isometric contraction (MVIC) in SQ exercise (Figure 19). In addition, to compare the performances attained throughout the session, force, velocity and power output values along with EMG data were recorded for every repetition.

Participants were asked to abstain from any strenuous physical activity for at least 2 days before each trial. Participants were also instructed to maintain their usual diet, although they were asked to refrain from consuming caffeine 12 hours before attending the study. All sessions took place at a neuromuscular research laboratory under the direct supervision of the researchers, at the same time of the day for each subject and under similar environmental conditions (20°C and 60% humidity, approximately).

Subjects

Sixteen strength-trained men (age 23.4 ± 4.4 years; height 1.75 ± 0.05 m; body mass 73.9 ± 9.1 kg) with at least 2 years of RT experience in the SQ exercise (range 2 to 6 years; 1RM strength for the SQ exercise: 105.8 ± 12.1 kg, and 1.44 ± 0.11 normalized per kg of body mass) participated in the study. Subjects were injury-free and were fully informed about the procedures, potential risks and benefits of the study and they all signed a written informed consent prior to the tests. Participants reported they were not taking drugs, medications or dietary supplements known to influence physical performance. The present study was approved by the Research Ethics Committee of University of Vigo (Ref: 03-819), in accordance with the Declaration of Helsinki.

Testing Procedures

Progressive Loading Test

One week before the resistance exercise protocols, a progressive loading test was conducted on a Smith machine (Multipower Fitness Line, Peroga, Murcia, Spain) with no counterweight mechanism in order to obtain the 1RM strength and individualized load-velocity relationships. The SQ was performed with subjects starting from the upright position with the knees and hips fully extended and stance approximately shoulder-width apart, and the barbell resting across the back at the level of the acromion. Each subject descended in a continuous motion as low as possible (35-40° knee flexion), then immediately reversed motion and returned to the upright position. Unlike the eccentric phase that was performed at a controlled mean velocity (0.50-0.65 m \cdot s⁻¹), subjects were required to always execute the concentric phase at maximal intended velocity. Range of movement and velocity values of all repetitions were recorded at 1,000 Hz with a linear velocity transducer (T-Force System Ergotech, Murcia, Spain). Firstly, the participants warmed up by performing 6 repetitions with a 20 kg load. The initial load was set at 30 kg and was progressively increased in 10 kg increments until the mean propulsive velocity (MPV) was $\leq 0.50 \text{ m} \cdot \text{s}^{-1}$. Then, the load was increased with smaller increments (2.5-5.0 kg) for better adjustments. A total of 10.0 ± 1.8 increasing loads were used for each subject. Three repetitions were executed for light loads ($\geq 1.00 \text{ m} \cdot \text{s}^{-1}$), two for medium

loads (1.00-0.80 m·s⁻¹) and one for the heaviest loads (≤ 0.80 m·s⁻¹). Inter-set rests were 3 min for light and medium loads and 5 min for heavy loads. Only the best repetition (i.e. highest MPV) with each load was considered for subsequent analysis. The propulsive phase corresponds to the portion of the concentric action during which the measured acceleration is greater than acceleration due to gravity (-9.81 m·s⁻²) (Sanchez-Medina, Perez & Gonzalez-Badillo, 2017).

Resistance exercise protocol

Figure 19 provides a detailed timeline description of the experimental protocol. In order to minimize the effects of fluid changes caused by walking, participants remained lying down for 10 minutes before starting the TMG measurements and baseline data acquisition. During the time they were lying down, a resting blood sample was collected (lactate and ammonium) and electrode locations (for TMG and EMG) were marked. After baseline TMG and blood lactate measurements were taken, the CMJ and MVIC tests were performed. Then, a standardized SQ warm-up was performed before the resistance exercise protocol, which consisted of: 6-6-4-3-2 SQ repetitions with 20 kg, 40%, 50%, 60% and 70% of 1RM, respectively, with 3 min rest between loads. Relative loads were determined from the individual second-order load-velocity relationship ($r^2 = 0.996 \pm 0.002$) obtained from the Progressive loading test. Subsequently, the corresponding protocol was performed (3x8 with 75% 1RM and 5 min rest between sets vs. 6x4 with 75% 1RM and 2 min rest between sets). The SQ execution technique described in the "Progressive loading test" section was carefully reproduced in all repetitions performed in the study. A force plate (FP-500, Ergotech, Murcia, Spain) synchronized with a linear velocity transducer (T-Force System, Ergotech, Murcia, Spain) was installed on the Smith machine (Multipower Fitness Line, Peroga, Murcia, Spain) to record mean propulsive values of force (MPF), velocity (MPV) and power (MPP) for every repetition. In addition, EMG data (i.e. RMS and MDF) were also recorded throughout the 24 repetitions. Subjects received immediate velocity feedback while being encouraged to perform each repetition at maximal intended velocity. After completing the final repetition (the 24th), the battery of tests was repeated as follows: CMJ (20 s-Post), MVIC (50 s-Post), blood samples (2 min and 30 s-Post) and TMG (5 min-Post), in order to obtain the acute Post-exercise values. This order was chosen in order to minimize the interference between tests and record valid data; i.e. the acute response to mechanical performance (but not high-fatiguing tests), metabolic response about 2-3 min Post-exercise, and TMG after 4-5 min resting. The duration per subject and session was approximately 1 hour.



Figure 19. Schematic representation of the study design including the 2 resistance exercise protocols analyzed and the timeline of the battery of tests before and after the protocols.

Tensiomyography

The contractile properties of the vastus lateralis (VLA) and vastus medialis (VME) muscles of the right leg were assessed using a TMG (TMG-100 System electrostimulator, TMG-BMC, Ljubljana, Slovenia) to determine their response to an electrically-evoked contraction. The electric stimulus was applied through two self-adhesive electrodes (5x5 cm, Dura-Stick® premium, Cefar-Compex, Hanover, Germany) placed at a 5 cm inter-electrode distance. The muscle mechanical response was measured with a digital Dc-Dc transducer Trans-TekR (GK 40, Ljubliana, Slovenia) placed perpendicular to the muscle belly and equidistant from the self- adhesive electrodes at a distance of 25-30 mm. Participants remained lying in a supine position for 10 min before starting the TMG data acquisition and the VLA and VME were marked according to SENIAM indications and location (Hermens, et al., 2000). To ensure the same placement of electrodes between consecutive measurements, the locations were marked on the skin using a permanent marker and participants were advised to keep the marks in place until the second session. Measurements were taken with the athletes in the supine position and the knee joint fixed at an angle of $\sim 140^{\circ}$ using a wedge cushion. Electrical stimulation was applied with a pulse duration of 1 ms and an initial current amplitude of 40 mA, which was progressively increased in 10 mA steps up to the stimulator's maximal output (100 mA). The use of a stimulus pulse of 1 ms, using a 5 x 5 cm electrode (the procedure followed in the present study) has been recently found to be necessary to reach a reliable and reproducible assessment of muscle contractile properties (Piqueras-Sanchiz et al., 2020a). A 10-s rest period was allowed between each electrical stimulus to avoid fatigue or post-tetanic activation. The variables assessed in this study were the maximum radial displacement of the muscle belly (Dm), contraction time (Tc), and delay time (Td). Dm was defined as the peak

amplitude in the displacement-time curve of the tensiomyographical twitch response; Tc was obtained by determining the time interval from 10 to 90% of Dm; and Td was defined as the time between the electrical stimulus and 10% of Dm (Valencic, & Knez, 1997). In addition, the velocity of deformation (Vd) was calculated as: $Dm \cdot (Tc + Td)-1$ (Loturco et al., 2016). Although Vd was originally termed the velocity of contraction, it is now recommended to use the term velocity of deformation, which is mainly dependent on muscle stiffness, in order to avoid confusion with sarcomere shortening velocity (Valenzuela et al., 2018). Mean velocities of muscle contraction (mm \cdot s-1 from the onset of electrical stimulation until 10% (V₁₀) and 90% (V₉₀) of Dm were also recorded using equations developed elsewhere (de Paula Simola et al., 2015). All measurements were carried out by the same experienced evaluator and only the curve with the highest Dm value was considered for further analysis. Test-retest reliabilities for TMG measures, using the intraclass correlation coefficient (ICC) with 95% confidence intervals (CI) and coefficient of variation (CV) values, were as follows: Dm (ICC (95%CI): 0.98 (0.96; 0.99), CV: 5.3%); Tc (ICC (95% CI): 0.98 (0.95; 0.99), CV: 3.2%); Td (ICC (95% CI): 0.95 (0.89; 0.98), CV: 2.3%); Vd (ICC (95% CI): 0.98 (0.97; 0.99), CV: 4.6%), V₁₀ (ICC (95% CI): 0.97 (0.95; 0.99), CV: 5.3%), and V₉₀ (ICC (95% CI): 0.98 (0.97; 0.99), CV: 4.6%). Metabolic variables

After cleaning the skin, 5 and 20 µL of capillary blood from fingertip punctures were used for lactate and ammonia quantification, respectively. Lactate was measured using a portable lactate analyzer (Lactate Pro 2, Arkray, Kyoto, Japan) and ammonia was analyzed using an ammonia checker (Blood Ammonia Meter PocketChem, model BAPA-4140; Arkray, Kyoto, Japan). Both devices were calibrated before each exercise session according to the manufacturer's specifications.

Countermovement jump

An infrared timing system (OptojumpNext, Microgate, Bolzano, Italy) was used for determining jump height. The CMJ was performed with both hands on the waist, while performing a downward movement to about 90° of knee flexion, followed by a maximal vertical jump. All participants were instructed to land in an upright position and to bend the knees after landing. The participants were required to do 2 trials separated by 10 s, and the mean height was determined. A specific warm-up was performed, consisting of 5 min of jogging at a self-selected easy pace, 2 sets of 10 squats without external load, 5 submaximal CMJs and 3 maximal CMJs. Test–retest reliability values were ICC (95% CI): 0.99 (0.97; 0.99) and CV: 1.9%.

Maximal voluntary isometric contraction test

Kinetic and EMG data were measured during an MVIC in the SQ exercise with the participants standing with their knees flexed at 90° ($180^{\circ} =$ full extension) measured with a handheld goniometer. This test was performed on a Smith machine instrumented with two telescopic bar holders, with a precision scale placed at the left and right sides of the Smith machine to precisely replicate the individual positions between trials. The subjects were instructed to maintain a constant minimum pre-tension until the experimenter gave the following verbal instruction: "Push against the ground as hard and as fast as possible" (Rodriguez-Rosell, Pareja-Blanco, Aagaard & Gonzalez-Badillo, 2018). Two 5 s trials, separated by 30 s rest, were performed. The warm-up protocol consisted of two attempts at 70% and 90% of perceived effort with 30 s rest between them.

Kinetic data

External forces were collected at a sampling rate of 1,000 Hz with an 80 x 80 cm dynamometric platform (FP-500; Ergotech, Murcia, Spain) and processed with specific software (T-Force System; Ergotech, Murcia, Spain). Maximal isometric force (MIF) was defined as the maximal strength value attained during the MVIC. RFDmax was calculated as the maximal slope of the force–time curve measured in 20-ms time intervals. The average value of each variable in the two attempts was recorded for further analysis. Test–retest reliability values for MIF and RFDmax were: ICC (95% CI) 261 0.99 (0.97; 0.99) and 0.94 (0.86; 0.97); and CV: 3.4% and 13.8%, respectively.

EMG signal acquisition

Surface EMG electrodes were placed on the same location previously described for TMG measurements. EMG signals were recorded continuously during MVIC testing using a parallel-bar, bipolar surface electromyographic sensor TrignoTM wireless EMG system, with an interelectrode distance of 10 mm, common mode rejection ratio >80 dB, and bandwidth filter between 20 and 450 Hz \pm 10% (Delsys Inc, Natick, MA) <5 μ V peak to peak, and sampling rate was 2,000 Hz. The raw data from the EMG were stored in digital format using EMG works Acquisition software. From each isometric and dynamic contraction the highest averaged (over sliding windows of 500 ms with an overlap of 499 ms) RMS and MDF values for each muscle were recorded. VME and VLA muscle excitation values were averaged, and the average of the two MVICs was calculated for further analysis. The value of the signal from MVIC at Pre-training of each resistance exercise protocol was used to normalize the EMG parameters. Test-retest reliability for RMS measures was: ICC (95% CI): 0.950 (0.90; 0.98) and CV: 7.4%, and for MDF was ICC (95% CI): 0.952 (0.90; 0.98) and CV: 5.3%.

Statistical Analyses

Values are reported as mean \pm standard deviation (SD). Sample size was calculated (using GPower Version 3.1.9.4) introducing the following parameters: effect size (ES) 0.85 and 0.70 for mean velocity and mean power between-protocols comparisons based on a recent meta-analysis comparing different RT set configurations (20); and α error probability (0.05) and power (0.95), which resulted in a sample size of 6 and 14 subjects, respectively. Statistical significance was established at $p \le 0.05$. Test-retest absolute reliability was measured by the standard error of measurement (SEM), which was expressed in relative terms through CV. The SEM was calculated as the root mean square of the total mean square intra-subject. Relative reliability was assessed using the ICC calculated with the one-way random effects model and its 95% CI. A paired sample t-test was conducted to compare the average values attained during each protocol (averaged 24 repetitions). A 2 (protocol) x 24 (repetitions) repeated measures ANOVA with Bonferroni's post-hoc adjustments were performed to compare differences between protocols throughout the repetitions completed. In addition, a 2 (protocol) x 2 (Pre vs. Post) repeated measures ANOVA with Bonferroni's post-hoc comparisons was performed to analyze the acute response to each protocol. In addition, pre-post ES values were calculated using Hedge's on the pooled SD (Hedges & Olkin, 1985) using a purpose-built spreadsheet. The rest of statistical analyses were performed using SPSS software version 20.0 (SPSS Inc., Chicago, IL, USA). Figures were designed using SigmaPlot 12.0 (Systat Software Inc, San Jose, CA, USA).

Results

Descriptive characteristics of the resistance exercise protocol. Table 16 shows the mechanical and neuromuscular characteristics of each protocol. Higher MPF, MPV and MPP values were obtained for the 6x4 configuration compared with 3x8. In addition, the 3x8 protocol exhibited significantly higher RMS during the session than 6x4, with no significant differences for MDF. Figure 20 shows the evolution of mechanical parameters throughout the 24 repetitions for each resistance exercise protocol. Significant "protocol 3 repetitions" interactions (p< 0.001) were observed for all mechanical variables. Performance in these variables progressively decreased throughout the 24 repetitions for both protocols. However, performance in these parameters (i.e., MPF, MPV, and MPP) was higher for the 6 x 4 protocol compared with the 3 x 8 protocol. Figure 21 depicts the development of neuromuscular variables throughout the 24 repetitions for each resistance exercise protocol. Significant "protocol. Significant "protocol. Significant the 3 x 8 protocol. Figure 21 depicts the development of neuromuscular variables throughout the 24 repetitions for each resistance exercise protocol. Significant "protocol. Significant the 3 x 8 protocol. Figure 21 depicts the development of neuromuscular variables throughout the 24 repetitions for each resistance exercise protocol. Significant "protocol x repetitions" interactions were observed for RMS (p< 0.001) and for MDF (p = 0.002). The 3 x 8 configuration resulted in higher RMS and lower MDF values than the 6 x 4 protocol, particularly for the final repetitions of each set.
Table 16. Average values of mechanical performance and neuromuscular parameters attained during the entire protocols (24 repetitions). Data are mean \pm SD, n= 16. 3x8: protocol consisted of performing 3 sets of 8 repetitions with 75% of 1RM; 6x4: protocol consisted of performing 6 sets of 4 repetitions with 75% of 1RM; MPF: mean propulsive force; MPV: mean propulsive velocity; MPP: mean propulsive power. RMS: root mean square averaged from the vastus medialis and vastus lateralis muscles; MDF: median frequency averaged from the vastus medialis and vastus lateralis muscles.

	3x8	6x4	P-value
MPF (N)	842.87 ± 96.40	869.38 ± 116.92	0.03
MPV $(\mathbf{m} \cdot \mathbf{s}^{-1})$	0.59 ± 0.08	0.63 ± 0.08	0.02
MPP (w)	486.36 ± 66.83	524.79 ± 74.46	0.001
RMS (%)	117.45 ± 15.83	105.69 ± 11.58	0.02
MDF (%)	105.13 ± 9.40	108.59 ± 10.69	0.23



Figure 20. Evolution of mechanical performance (A) MPF, B) MPV, and C) MPP) throughout the 24 repetitions for both protocols. Data are mean \pm SD, n= 16. 3x8: protocol consisted of performing 3 sets of 8 repetitions with 75% of 1RM; 6x4: protocol consisted of performing 6 sets of 4 repetitions with 75% of 1RM. Intra-protocol significant differences with respect to the first repetitions: *p< 0.05. Inter-protocol significant differences in each repetition: *p< 0.05.



Figure 21. Evolution of neuromuscular parameters throughout the 24 repetitions for both protocols. A) root mean square averaged from the vastus medialis and vastus lateralis muscles; and B) median frequency averaged from the vastus medialis and vastus lateralis muscles. Data are mean \pm SD, n= 16. 3x8: protocol consisted of performing 3 sets of 8 repetitions with 75% of 1RM; 6x4: protocol consisted of performing 6 sets of 4 repetitions with 75% of 1RM. Data were normalized with respect to the first repetition of each resistance exercise protocol. Within-protocol significant differences with respect to the first repetition: *p< 0.05. Significant differences between protocols at the corresponding repetition: #p< 0.05.

Tensiomyography

No significant "protocol x time" interactions were found for TMG-derived parameters (Table 17). A significant "time effect" was observed for all variables, except for VLA-Tc and VME-Td, with significant decreases in Dm and Vd (in both muscles) and VLA-Td for both protocols at Post. However, the 3x8 protocol showed significantly lower values of VLA-Vd, VLA-V₁₀ and VLA-V90 than 6x4 at Post.

110

Table 17. Effects of different resistance exercise protocols on muscle's contractile properties assessed by tensiomyography. Data are mean \pm SD, n= 16. 3x8: protocol consisted of performing 3 sets of 8 repetitions with 75% of 1RM; 6x4: protocol consisted of performing 6 sets of 4 repetitions with 75% of 1RM; VL: vastus lateralis muscle; VM: vastus medialis muscle; TC: contraction time; TD: delay time; DM: muscle displacement; VC: Velocity of deformation radial (DM / (TC+TD); ES: within protocol effect size from Pre to Post-protocol. Intra-protocol significant differences from Pre to Post-training: *p < 0.05, **p < 0.01, ***p < 0.001. Inter-protocol significant differences from Pre to Post-protocol: *p < 0.05.

							P-value	P-value
	3x8				6x4	time effect	protocol x time	
-	Pre	Post	ES	Pre	Post	ES		
TC-VL (ms)	21.80 ± 3.75	22.66 ± 3.57	0.25	21.48 ± 2.84	22.39 ± 3.37	0.26	0.157	0.95
TC-VM (ms)	21.90 ± 2.83	22.87 ± 3.47	0.32	21.85 ± 2.73	$23.04 \pm 2.93^{*}$	0.39	0.045	0.63
TD-VL (ms)	22.82 ± 1.66	$21.36\pm1.84^*$	-0.87	22.48 ± 1.64	$20.93 \pm 1.43^{**}$	-0.92	0.004	0.89
TD-VM (ms)	22.00 ± 1.36	22.09 ± 2.17	0.06	21.74 ± 1.14	21.35 ± 1.42	-0.24	0.715	0.41
DM-VL (mm)	6.13 ± 1.62	$3.77 \pm 1.57^{***}$	-1.37	6.05 ± 1.98	$4.09 \pm 1.58^{***}$	-1.14	P<0.001	0.29
DM-VM (mm)	8.58 ± 1.63	$6.61 \pm 1.54^{***}$	-1.25	8.35 ± 1.59	$6.70 \pm 1.45^{***}$	-1.05	P<0.001	0.29
VC-VL (mm/ms ⁻¹)	0.138 ± 0.038	$0.084 \pm 0.029^{***}$	-1.71	0.135 ± 0.040	$0.093 \pm 0.030^{***}$	-1.42	P<0.001	0.10
VC-VM (mm/ms ⁻¹)	0.196 ± 0.038	$0.149 \pm 0.039^{***}$	-1.35	0.193 ± 0.035	$0.152 \pm 0.034^{***}$	-1.08	P<0.001	0.39

Metabolic response and jump performance

Significant "protocol x time" interactions were observed for CMJ height and blood lactate and ammonia values (Table 18). The 3x8 protocol induced higher lactate and ammonia concentrations and CMJ height impairments than 6x4 at Post.

Mechanical and neuromuscular response during maximal voluntary isometric contraction

Significant "protocol x time" interactions were noted for neuromuscular parameters (i.e. RMS: p = 0.02; and MDF: p = 0.002) (Fig. 3). However, no "protocol x time" interactions were noted for mechanical outcomes (i.e. MIF and RFDmax) (Table 3). The RMS attained during MVIC decreased for both protocols at Post, although lower values were observed for 6x4. However, the 6x4 protocol induced significant increases in MDF at Post while the 3x8

protocol exhibited significantly lower MDF than 6x4. Moreover, both MIF and RFDmax significantly decreased at Post, with no significant differences between protocols.

Table 18. Mechanical, neuromuscular and metabolic response to the different resistance exercise protocols under study. Data are mean \pm SD, n = 16. 3x8: protocol consisted of performing 3 sets of 8 repetitions with 75% of 1RM; 6x4: protocol consisted of performing 6 sets of 4 repetitions with 75% of 1RM; Lactate: blood lactate concentration; Ammonia: blood ammonia concentration; CMJ: countermovement jump height; MIF: maximal isometric force; RFDmax; maximal rate of force development; RMS: root mean square averaged from the vastus medialis and vastus lateralis muscles; MDF: median frequency averaged from the vastus medialis and vastus lateralis muscles; ES: within-protocol effect size from Pre to Post. Intra-protocol significant differences from Pre to Post: * p < 0.05, ** p < 0.01, *** p < 0.001. Significant differences between protocols at the corresponding time-point: *p < 0.05.

	3x8			6x4			p-value time	p-value
	Pre Post ES		Pre	e Post E		effect	time	
Lactate (mmol·l ⁻¹)	1.7 ± 0.6	$12.0 \pm 3.8^{***\#}$	3.31	1.5 ± 0.7	$8.9 \pm 4.2^{***}$	2.38	<i>p</i> <0.001	0.05
Ammonia (µmol·l ⁻¹)	60.5 ± 16.6	$103.8 \pm 44.9^{***\#}$	1.21	65.1 ± 20.4	76.4 ± 44.1	0.31	0.001	0.02
CMJ (cm)	37.3 ± 4.6	$27.0 \pm 3.8^{***\#}$	-2.56	37.5 ± 3.6	$29.6 \pm 3.4^{***}$	-1.96	<i>p</i> <0.001	0.02
Maximal Voluntary Isometric Contraction								
MIF (N)	1267.7 ± 230.9	1034.5 ± 258.7***	-0.93	1270.0 ± 279.0	1017.3 ± 216.8***	-1.01	<i>p</i> <0.001	0.62
RFDmax (N·s ⁻¹)	4609 ± 1528	$3998 \pm 1431^{**}$	-0.38	5242 ± 2006	$3924 \pm 1224^{**}$	-0.82	0.001	0.09
RMS (%)	100.0	90.1 ± 9.3**#		100.0	$78.8 \pm 14.4^{***}$		<i>p</i> <0.001	0.02
MDF (%)	100.0	$97.4\pm5.9^{\#}$		100.0	$105.0 \pm 5.8^{**}$		0.07	0.003

Discussion

Integral responses to different set configurations, were examined during (mechanical and neuromuscular features) and after exercise (mechanical, neuromuscular, metabolic, and muscle contractile properties). The results indicate that redistributing long sets and inter-set rest periods into shorter but more frequent sets modulates training stimuli. Specifically, shorter set configurations (i.e. 6x4) resulted in greater mechanical performance maintenance and concomitant lower neuromuscular markers of fatigue during the exercise compared to longer set configurations (i.e. 3x8). Longer set configurations also led to greater metabolic responses, along with greater impairments in jump height and muscle deformation velocity (i.e. Vd, V₁₀, and V₉₀) after exercise. Lastly, an overall impairment in isometric force production was observed for both protocols, with no significant differences between them. Therefore,

introducing shorter but more frequent inter-set rests attenuates impairments in mechanical performance, especially in the latter final repetitions of each set, which may be mediated by lower neuromuscular alterations, reduced metabolic stress, and better maintained muscle contractile properties.

Performance as indicated by mechanical parameters (i.e. force, velocity and power) progressively decreased throughout the repetitions for both protocols, although shorter but more frequent sets alleviated fatigue-induced impairments in performance during the session. There is compelling evidence that including rest periods between repetitions or clusters of repetitions is an effective strategy to attenuate fatigue and ameliorate loss of mechanical performance during RT sessions (Jukic, Ramos, Helms, McGuigan, & Tufano, 2020; Latella, Teo, Drinkwater, Kendall, & Haff, 2019; Tufano et al., 2017). However, the mechanisms underlying this phenomenon have not been clearly established. One of the potential mechanisms associated with the higher acute performance attained using shorter set configurations is that the frequent rest periods may allow for greater maintenance of phosphocreatine (PCr) stores, a partial resynthesis of adenosine triphosphate (ATP) and increased metabolite clearance in the working muscles (Tufano et al., 2017; Gorostiaga, Navarro-Amezqueta, Calbet, Hellsten, Cusso, Guerrero, Granados, Gonzalez-Izal, Ibanez, & Izquierdo, 2012). In agreement with previous studies (Moran-Navarro et al., 2017; Gorostiaga, Navarro-Amezqueta, Calbet, Sanchez-Medina, Cusso, Guerrero, Granados, Gonzalez-Izal, Ibanez, & Izquierdo, 2014), the longer set configuration (i.e. 3x8) induced higher blood ammonia levels. An increase in blood ammonia concentrations during high-intensity exercise is interpreted as indicative of accelerated ammonia production by muscles, resulting from the deamination of adenosine monophosphate to inosine monophosphate (Hellsten-Westing, Norman, Balsom & Sjodin, 1993). In this regard, Gorostiaga et al. (2012), observed greater depletion of intramuscular ATP and PCr stores following 5x10 vs. 10x5 with 85% 1RM with 2 min inter-set rests in the leg press exercise, along with concomitant greater power reductions. Likewise, the higher lactate values observed for the longer set configuration suggest a greater reliance on anaerobic glycolysis for energy for these set structures (Jukic et al., 2020; Oliver et al., 2015). Accordingly, the lower blood ammonia and lactate concentrations observed after the shorter set configurations (i.e. 6x4) may indicate better replenishment of ATP and PCr stores, as well as reduced glycolytic requirements within each set, which may result in a greater ability to maintain mechanical performance.

In an attempt to provide a better understanding of the mechanisms behind the different performances achieved with longer and shorter set configurations, we recorded the EMG activity attained in each repetition. Our data suggest that longer set configurations induced higher neuromuscular fatigue (i.e. higher RMS and lower MDF values) during isometric (i.e. MVIC) and dynamic contractions, mainly during the latter final repetitions in each set (Figure 21). In agreement with our findings, Ortega-Becerra, Sanchez-Moreno, & Pareja-Blanco (2020), reported higher RMS and higher MDF values during traditional sets (3 sets of 12 repetitions at 60% of 1RM with inter-set rests of 2 min,) in the bench-press exercise compared to protocols employing similar training intensities and volume but including cluster configurations (30-second rest every 4 or 2 repetitions). Fatigue-induced alterations in neuromuscular markers have been attributed to metabolic by product accumulation (Hunter, Duchateau & Enoka, 2018). Specifically, increased RMS values may be primarily due to increased muscle activation (i.e. recruitment of higher-threshold motor units, motor unit firing frequency and/or changes in intrinsic muscle properties) attempting to compensate for the loss of force in the fatigued state (Hunter et al., 2018; Bigland-Ritchie, Dawson, Johansson & Lippold, 1996), while decreased MDF values may be evoked by reduced action potential conduction velocity associated with a decline in intramuscular pH (Brody, Pollock, Roy, De Luca, & Cel, 1991), changes in action potential shape (Hermens, Bruggen, Baten, Rutten, & Boom, 1992) and decreases in the firing rate of fatigued fast motor units (Bigland-Ritchie, Dawson, Johansson & Lippold, 1996). In this regard, it is fair to assume that interpretation of mechanistic information from EMG data is speculative. Nevertheless, our data suggest that from a neuromuscular standpoint, shorter set configurations also contribute to reduced fatigue development, which may be due to lower metabolic by product accumulation.

Besides the effects of set configuration on exercise performance itself, it is also important to consider the residual mechanical fatigue induced by these training sessions. In line with the fatigue levels observed during the training session, greater reductions in CMJ height were observed following the longer set configuration. Previous studies have also shown smaller CMJ height losses after training sessions with intra-set rest periods compared to traditional structures (Gonzalez-Hernandez et al., 2020; Mora-Custodio et al., 2018). However, the exercise-induced fatigue on isometric strength (i.e. MIF and RFDmax) and several TMGderived parameters (i.e. Dm, Tc, and Td) was similar for both set configurations. Decreases in Dm after RT have been associated with impaired muscle function (Hunter et al., 2017), muscle swelling and exercise induced muscle damage (Harmsen et al., 2019), while it has been suggested that temporal TMG parameters should be treated with caution (Macgregor et al., 2018). On the other hand, the 3x8 protocol induced greater impairments in muscle velocity of deformation (i.e. Vd, V_{10} and V_{90}) than 6x4 (Table 2). The only study that has previously examined the acute effects of different set configurations (4x10 with 95 s of inter-set rest vs. 20x2 with 15 s inter-set rest) on TMG outcomes did not observe significant differences in TMG parameters during post-exercise measurements, although the ES values indicated a lower impaired muscle function for shorter set configurations (Tufano et al., 2019). Therefore, longer set configurations induced greater impairments in neuromuscular ability and muscle function in both voluntary (i.e. squats and jumps) and involuntary (i.e. TMG) actions.

Conclusions

Despite using similar loads (75% 1RM), total volume (24 repetitions), total rest (10 min), and, therefore, training density (work- to- rest ratio), implementing shorter but more frequent inter-set rest intervals allowed for better maintenance of performance, especially in the final repetitions of each set, along with lower alterations of neuromuscular markers of fatigue, a dampened metabolic stress, and a lesser worsening of muscle velocity of deformation. Therefore, rest redistribution may be a viable strategy for maintaining performance during a training session, which may be mediated by lower neuromuscular alterations, reduced metabolic stress and better maintenance of muscle contractile properties.

Practical applications

The present study provides a greater insight for sport professionals about the integral responses (mechanical, neuromuscular and metabolic) to different set configurations, which may allow us to optimize the design of RT programs aimed at enhancing physical performance. Coaches can implement shorter but more frequent rest intervals to spare mechanical performance and alleviate neuromuscular fatigue, while longer set configurations could be used to increase metabolic stress. Therefore, a given set configuration could be chosen beforehand, depending on the specific training goal being pursued. Further studies should analyze the long-term effects of different configurations on mechanical, neuromuscular and hypertrophic adaptations.

ESTUDIO III

12. ESTUDIO III

Artículo III

TITLE: EVOLUTION OF CONTRACTILE PROPERTIES OF THE LOWER LIMB MUSCLES THROUGHOUT A SEASON IN ELITE FUTSAL PLAYERS

The Journal of Sports Medicine and Physical Fitness

Futsal is a team sport involving intermittent technical actions of high intensity, and high physical (strength) and muscular demands. In this regard, the tensiomyography (TMG) is a useful and non-invasive tool for the monitoring and assessment of the muscle's contractile capacity. This study aimed to analyse the changes in the contractile properties produced during the season, as well as to determine the potential cumulative effect of a resistance training (RT) program in futsal players. Fourteen elite futsal players (2 goalkeepers, 4 defenders, 4 wingers and 3 pivots) were assessed by TMG at 11th, 18th, and 28th week of the season. The maximal radial displacement of the muscle belly (Dm); contraction time (Tc); delay time (Td) and radial displacement velocity (90%) Dm (VrD₉₀) were assessed. After the second measurement, a RT program was included in the regular training sessions and focused on the lower body musculature. It was performed during 9 weeks (1-weekly). Finally, a third measurement was performed between 28th-29th weeks. Repeated measures analysis of variance was used to detect in-season changes. Two factors were included: Time (changes detected after resistance training program) was used as the within-subject factor and the specific position was used as the between-subject factor. An increment in Tc for several muscles: biceps femoris (BF; p =0.02), semitendinosus (ST; p = 0.04), adductor longus (AL; p=0.008) and gastrocnemius medialis (GM; p = 0.009) was observed throughout the season. Similarly, significant increments in Dm for GM (p = 0.02) and AL (p = 0.05), as well as increments in Td for BF (p= 0.002) were found. Moreover, no significant changes in VrD_{90} between time points 2-3 (analysis of RT effect) were observed. Additionally, the player's positions reported no significant changes for any of the variables analysed. An increase respect to baseline levels was observed for Tc, Td and Dm during the season. However, the adaptations to contractile properties were muscle specific. In addition, an in-season 9-week RT program (1-weekly), had no significant effects (time points 2-3) on the contractile properties of futsal players. In addition, there were no differences when comparing different positions.

Keywords: futsal, resistance training, muscle contractile properties

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TITLE: EVOLUTION OF CONTRACTILE PROPERTIES OF THE LOWER LIMB MUSCLES THROUGHOUT A SEASON IN ELITE FUTSAL PLAYERS

¹Francisco Piqueras-Sanchiz, ²Luis M. Martínez-Aranda, ^{1,3}Fernando Pareja-Blanco, ⁴David Rodríguez-Ruiz & ⁵Óscar García-García*

¹Faculty of Sports Science, Pablo de Olavide University, Seville, Spain; ²Neuroscience of Human Movement Research Group, Faculty of Sport, Catholic University San Antonio, Murcia, Spain; ³Physical Performance and Sports Research Center, Department of Sports and Computers Sciences, Pablo de Olavide University, Seville, Spain; ⁴Department of Physical Education, University of Las Palmas, Gran Canaria, Spain; ⁵Sport Performance, Physical condition and Wellness Lab, University of Vigo, Pontevedra, Spain.

Introduction

Futsal is growing in popularity and more than one million futsal players have already been registered by national federations worldwide. Futsal demands intermittent high-intensity actions which require high physical, technical and tactical efforts from the players. (Caetano, de Oliveira Bueno, Marche, Nakamura, Cunha, & Moura, 2015; Barbero-Alvarez, Soto, Barbero-Alvarez, & Granda-Vera, 2008). As a consequence, players are required to undertake extensive training during different phases of the season aiming to improve both, endurance and power-speed abilities (Yanci, Castillo, Iturricastillo, Ayarra, & Nakamura, 2017; Miloski, de Freitas, Nakamura, de A Nogueira, & Bara-Filho, 2016). It is not uncommon to observe an impairment of physical performance (e.g., vertical jump height and sprint speed) over periods of high volumes of training and competition likely due to the "interference effect" of aerobic and strength power stimuli (Hickson, 1980). In fact, team sports players including futsal players can suffer from the concurrent training effects and become slower and less powerful during some phases of the preparation (Nakamura, Pereira, Rabelo, Ramirez-Campillo, & Loturco, 2016; Loturco et al., 2016). Likewise, the absence of specific strength training programs can impair sprint and strength performance throughout the competitive season in soccer players (Magal, Smith, Dyer, & Hoffman, 2009), whereas implementing strength training during the competitive season in a group of high-level amateur soccer players, was able to improve the peak strength and the rate of force development without altering sprinting speed (Faude, Roth, Di Giovine, Zahner, Donath, 2013). However, it is not easy to implement physical tests too

frequently in order to monitor relevant fitness changes, as maximal tests can be timedemanding and increase the risk of undesired injuries.

An alternative to the monitoring of the fitness status in team sport players could be the use of the tensiomyography (TMG). TMG is a non-invasive tool that measures muscle belly enlargement in transversal plane during an isometric twitch contraction to provide mechanical indices of the muscle contraction properties that are related to performance changes. Maximal displacement (Dm), delay time (Td) and contraction time (Tc) are among the main TMGderived parameters (Martín-Rodríguez et al., 2017a). These parameters have shown good to excellent reliability and low measurement error (Martín-Rodríguez et al., 2017a). In addition, both Dm and radial displacement velocity until 10% (VrD₁₀) and 90% (VrD₉₀) of Dm have been considered as indirect markers of muscle fatigue (Macgregor et al., 2018; de Paula et al., 2015). For instance, the rectus femoris Dm, which is inversely related to muscle stiffness, was acutely increased after a repeated-sprint ability test protocol in futsal players, indicating reduced muscle stiffness Sánchez-Sánchez, Bishop, García-Unanue, Ubago-Guisado, Hernando, & López-Fernández, 2018. Furthermore, this increasing change in response to the protocol (pre- to post) was negatively correlated to the performance change across the sprints (r=-0.49). In a longitudinal observation, it was found that in soccer players the radial displacement velocity (VrD) was impaired during the inter-season period, concomitant with reductions in sprinting speed (Loturco et al., 2016). Another longitudinal observation in professional soccer players found was that Tc-Td-Dm significantly decreased (~10%) after 10 weeks (García-García et al., 2016). In professional road cyclists, the contractile properties were also analyzed in two periods: preparation and competition, aiming to detect differences, before and after the official competitive season, in the contractile properties of the main musculature involved in this kind of endurance discipline. The Tc values during the competitive period showed marked differences between the knee extensors (large increased) and the knee flexor (large decreased), whereas the Dm did not change (García-García et al., 2013). Taken together, TMG provides some advantages: 1) involuntary contractions, which avoid the influence of factors such as motivation; 2) it does not affect performance for subsequent training sessions; 3) it allows the assessment of individual muscles in the field. These results support the notion that TMG indices can be useful to monitor muscle contraction properties that can influence performance in team sports players. However, these indices have not been investigated yet in professional futsal players, especially in response to a period of resistance training.

Resistance training has been proposed as an effective training method to improve performance in team sports (Suchomel, Nimphius, Bellon, & Stone, 2018). Its implementation

has been shown to enhance the change of direction and speed (Pareja-Blanco, Asián-Clemente, & Sáez de Villarrea, 2021; Tous-Fajardo, Gonzalo-Skok, Arjol-Serrano, & Tesch, 2018), the linear speed and the vertical jump capacity (Pareja-Blanco et al 2021; Gonzalo-Skok, Tous-Fajardo, Valero-Campo, Berzosa, Bataller, Arjol-Serrano, Moras, & Mendez-Villanueva, 2018), in addition to a reduced risk of injuries in team sports players (de Hoyo, Pozzo, Sañudo, Carrasco, Gonzalo-Skok, Domínguez-Cobo, & Morán-Camacho, 2015). In line with this, a recent study has shown a Dm decreased in vastus lateralis, biceps femoris, tibialis anterior, gastrocnemius medialis and gastrocnemius lateralis after eight weeks of plyometric training (Zubac, & Simunic, 2017). In spite of the widely described seasonal changes in physical performance in team sport athletes, less is known about the seasonal changes in the neuromuscular abilities along with the adaptation of contractile properties to systematic resistance training in team sport athletes. Therefore, the aims of the present study were: (I) to analyze the seasonal changes in the contractile properties produced in elite futsal players; (II) to determine the possible cumulative effect of resistance training program on the contractile properties in these futsal players. We hypothesize that there are impairments in the neuromuscular abilities of the main lower limbs muscles which are involved during the season and that, during the resistance training program, those changes will be modulated.

Materials and methods

Study design

A longitudinal and intragroup comparative study was conducted to assess in-season changes in the contractile properties of the musculature evaluated in professional futsal players. Players were divided according to their specific position: goalkeeper, defender, winger and pivot. Three neuromuscular evaluations were conducted during the competitive season using TMG. Data collection started at the 11th week of the season (five pre-season weeks and six competition weeks) with an average of 82 ± 61 competitive min played per evaluated player and an average of accumulated training minutes of $6,703 \pm 653$ minutes since the start of the preseason. Those values were established as a baseline. The second measurement was performed between the 18th19th week of the season (accumulated competitive minutes: 193 ± 82 min and accumulated training: $16,812 \pm 833$ min).

After this second measurement, an intervention based on resistance training program focused on the leg muscles was carried out during nine weeks (one weekly training session). Next, a third measurement was carried out between the 28th-29th week (accumulated competitive minutes: 302 ± 54 and an accumulated training minutes: $24,577 \pm 551$ min) (Figure 22). All players participated on average in ~10 hours of futsal training (5 sessions) plus 1

competitive match per week and the weekly distribution of training contents did not show significant variations throughout the season, with a solid work related to the training load (volume and intensity) between microcycles (Table 19).



Figure 22. Experimental design and testing structure

Table 19. Distribution of the training and competition load in a seasonal microcycle.

	MONDAY	TUESDAY	WEDNESDAY	THURSDAY	FRIDAY	SATURDAY	SUNDAY
MORNING	-	TEC-TAC	-	-	-		
AFTERNOON	PRT + CO + TEC- TAC	CORE + PROP + MAP + TEC-TAC	TEC-TAC	ACTIVATION + TEC-TAC	MATCH	FREE	FREE

PRT: Preventive Resistance Training, CO: Coordination; TEC-TAC: Technical-tactical; CORE: Core training; PROP: Proprioception; MAP: Maximum Aerobic Power.

All evaluations were conducted at the club facilities under the same environmental conditions. Using the TMG, eight muscles were tested in both legs: Adductor Longus (AL), Biceps Femoris (BF), Gastrocnemius Lateralis (GL), Gastrocnemius Medialis (GM), Rectus Femoris (RF), Semitendinosus (ST), Vastus Lateralis (VL) and Vastus Medialis (VM). Before each testing session, players complied with the following pre-test guidelines: not to consume any energy/performance enhancing drinks, supplements or beverages containing caffeine or alcohol 3 hours before testing, as well as not to eat any food at least 2 hours before testing. Evaluations were held on the same weekday, after the recovery day and before the next training session. No player exhibited abnormal fatigue symptoms or muscle damage.

Subjects

Fourteen elite futsal players $(27.2 \pm 3.3 \text{ years}, 74.2 \pm 5.6 \text{ kg} \text{ and } 180.2 \pm 4.8 \text{ cm})$ with no previous injuries during the past three months, and who complied with at least 85% of all training sessions participated in this study. Regarding the position, the players were two goalkeepers, four defenders, four wingers and three pivots. All the players belonged to the same team in the Spanish Professional Futsal League. One player missed too many training sessions or was absent from the post testing session due to injury or illness.

Ethical statement

All participants provided a written consent after being informed about the research process and the potential risks associated with TMG assessment. The research protocol followed the principles of the Declaration of Helsinki regarding biomedical research involving human subjects (64th WMA General Assembly, Fortaleza, Brazil, 2013). The Local Ethical Research Committee, as well as the coaching staff and management board of the professional futsal team approved the study.

Procedures

Resistance training program

The training took place one day per week over a period of 9 weeks. Each session had a duration of 45-60 min and was carried out always during the evening (20:30h-21:30h). Resistance bands, kettlebell, weights and platform as well as a Flywheel device Kbox® (inertia 0.035 kg·m²) were used in this study. All training sessions were preceded by a 10 min standardized dynamic warm-up (jogging, high knee skipping and running, lunges with trunk rotation, straight leg skipping, straight-leg deadlift walk, deep squatting and bilateral jumps), core stability exercises (plank, side plank and bridging) and dorsiflexion. The main exercises in the training program are presented in the figure 23 as follows: unipodal hip extension with resistance band, hip adduction with a resistance band, Kbox inertial bipodal calf, prevention tendinopathy unipodal, half squat, resisted acceleration with elastic bands and jumps in stable and unstable conditions. Two sets of eight repetitions were performed per exercise with 30 s of resting between exercises and 1 min between sets, except to Kbox inertial calf bipodal, in which was performed 2x8 with 60 s resting between exercises and 2 min between sets. All the exercises were executed in the same order in every session. The main researcher controlled every training session, providing verbal encouragement to each participant.

The premises in Kbox exercise were: concentric phase was performed as fast as possible. The eccentric phase of the bipodal calf required decelerating the movement because the flywheel keeps spinning due to its inertia. On the contrary, exercises employing resistance

band were carried out by performing the concentric/eccentric phases at high/low speed respectively.



Figure 23. Exercises performed during training program and workloads. A) Hip extension resistance unipodal band (48kg); B) hip adduction with an elastic unipodal band; C) Kbox inertial bipodal; D) prevention of unipodal tendinophaty (kettlebell 6-8 kg); E) barbell squat (22-26 kg); F) acceleration/deceleration resistance band accelerator (48kg); G) jump stable/unstable conditions.

TMG protocol

The TMG was used to assess the mechanical properties of eight selected muscles. The assessments were performed following the protocol described by García-García et al. (2013). All measurements were performed isometrically in relaxed predefined positions: RF, VL and VM in a supine position with the knee angle set at 30° flexion (where 0° represents the fully extended joint); BF and ST in a prone position with the knee angle set at 5° flexion; GM and GL in a prone position with the ankle in a neutral position; and AL in a side-lying position, with the leg closest to the bed slightly adducted. Foam pads were used to support the joints. Measures of radial muscle belly displacement were acquired through a digital displacement transducer (GK 40, Panoptik d.o.o., Ljubljana, Slovenia) located perpendicular to the thickest part of the muscle belly. These measurements were visually determined through palpation during a voluntary contraction. The self-adhesive electrodes (5x5 cm, Cefar Compex Medical AB Co., Ltd, Malmö, Sweden) were placed symmetrically at 2.5 cm away from the electrodes (Inter-electrode distance, 5 cm, IED), placing the positive electrode in the proximal area of the muscle above the measurement point and the negative electrode in the distal area below the measurement point, according to previous investigations14 and following the protocol suggested by Perotto, Delagi, Lazzeti, & Morrison. (2005). For all measurements, the point of maximum radial muscle belly displacement was determined by obtaining the time-

displacement curve for each muscle, as well as on the basis of low intensity measurements (30 mA) previously obtained by placing the sensor on different points (2-3 mm apart within the area defined by the electrodes), until the maximum radial displacement point was identified. Electrical stimulation was applied using a TMG-S1 stimulator (EMF FURLAN & Co. d.o.o., Ljubljana, Slovenia) with 1 ms monophasic pulse duration and 30 mA initial current amplitude, which progressively augmented in 10 mA until reaching 100 mA (maximal stimulator output). Consecutive stimuli were separated by a 10 s resting period to avoid fatigue or post-tetanic activation (Krizaj et al., 2008). A total of eight curves were recorded for each participant, but only the one with the highest maximum radial displacement was included in the analysis for each assessed muscle (García-García et al., 2017; 2016). All the measurements involved the recording of the following parameters of involuntary isometric contraction produced by the electrical stimulus: Maximum radial muscle belly displacement (Dm) in mm.; Delay time (Td) as the time in milliseconds (ms) from onset to 10% of Dm and; contraction time (Tc) as the time in ms from 10% to 90% of Dm. Other authors²⁵ previously reported the need to normalize the increase in Tc, in order to compare the records obtained from different muscles. Therefore, we used the radial velocity displacement (VrD), in order to have a measure relatively independent of the Dm, calculated by V₉₀, (Raeder et al., 2016) dividing the 90% of Dm by the sum of Tc and Td in mm·s-1 i.e., $V_{90} = VrD_{90} = ((90*Dm/100)) / ((Tc+Td) \cdot 1000).$

In order to analyze the reliability of the relative and absolute TMG parameters, two measurements for each assessment were conducted. The electrode and sensor positions were marked with a dermatological pen on four randomly selected participants for each assessment. These selected participants were re-tested 20-45 min after the initial data collection.

Statistical analysis

Application of the Kolmogorov-Smirnov Test, together with the Lilliefors Test, showed that the sample distribution was normal, linear, and homoscedastic. Relative and absolute reliability analyses were conducted using the data collected from within-day. Relative reliability of TMG parameters was assessed by calculating 2-way mixed effects model and the absolute agreement using the single measurement intra-class correlation coefficient (ICC) (McGraw, & Wong, 1998). In addition, absolute reliability was assessed by the standard error of measurement (SEM), which was expressed in relative terms through the coefficient of variation (CV). Preliminary analysis showed no significant differences (paired t-test) between dominant vs. non-dominant leg in any of the measured variables and thus, the analysis was based on the mean values for both legs. Repeated measures analysis of variance (RM ANOVA) was used to detect in-season changes in the mechanical and neuromuscular characteristics of

professional futsal players. Two factors were included in the RM ANOVA model: Time as changes detected between 1-2 points and 1-3 points (session effect) and changes detected between 2-3 points (resistance training program effect) was used as the within-subject variable and the specific position (goalkeeper, defender, winger and pivot) was used as the between-subject variable. Bonferroni post-hoc tests with 95% adjustment for confidence intervals were used to compare the main effects and identify significant individual differences.

The effect sizes in RM ANOVA were reported as partial eta squared (η 2) and interpreted as small (0.01), moderate (0.06) or large (0.14) (Cohen, 1998). The percentage differences in TMG parameters between the three assessments points were also calculated and interpreted. The significance level was set at 5% (p< 0.05). Statistical analyses were performed using SPSS v.21 (SPSS Inc., Chicago, IL, USA)

Results

The values obtained for Test-retest absolute reliability measured by the CV were between 3.2-5.2%. The ICC values (95% CI) ranged between 0.91-0.98. The evolution of contractile properties is reported in table 20 in terms of Tc, Dm, Td and VrD₉₀. No significant changes were observed between 2-3 time points for any of the studied variables (resistance training effect). A large increment in Tc was observed for several muscles: BF (17.5% between 1-3 time points; p = 0.02; $\eta^2 = 0.20$), ST (8.1% between 1-2 time points; p = 0.04; $\eta^2 = 0.28$), AL (10.1% between 1-2 time points and 18.4% between 1-3 time points; p=0.008; $\eta^2 = 0.38$) and GM (22.4% between 1-3 time points; p = 0.009; $\eta^2 = 0.37$). Larger increments were also observed in Dm for GM (30% between 1-3 time points; p = 0.02; $\eta^2 = 0.30$) and AL (22.4% between 1-3 time points; p = 0.02; $\eta^2 = 0.25$). Additionally, it was observed an increment in Td for BF (9.6% between 1-2 time points; p = 0.002; $\eta^2 = 0.46$), with a large size effect. No significant changes were observed in VrD₉₀ throughout the season, as well as non-significant increments for AL and GL between 2-3 time points (16.8%, p = 0.13; $\eta^2 = 0.16$ and 11.9%, p = 0.055; $\eta^2 = 0.25$, respectively). Finally, no significant differences were observed between player positions in any of the analyzed variables.

Table 20. Evolution of contractile properties throughout a season and after a resistance-training program in elite futsal players. Data are expressed as mean of both legs \pm SD (n=14). VM: vastus medialis; VL: vastus lateralis; RF: rectus femoris; BF: biceps femoris; ST: semitendinosus; AL: adductor longus; GL: gastrocnemius lateralis; GM: gastrocnemius medialis; Tc: contraction time; Dm: maximal radial displacement of the muscle belly; TD: delay time; VrD₉₀: radial displacement velocity at 90% Dm; Time-points 1, 2 and 3: First, second and third measurement. Tc, Td, and are in millisecons (ms), while Dm is in millimeters (mm) and VrD₉₀ in millimeters/milliseconds (mm·s⁻¹). Values are mean of both legs and standard deviation. ^a Significant differences in relation to baseline (Time 1) measurements (p < 0.05).

	-	-		-	-		-	-
TMG Parameters	VM	VL	RF	BF	ST	AL	GL	GM
Tc 1 (ms)	24.1 ± 2.6	21.1 ± 3.5	28.1 ± 3.8	32.0 ± 12.3	37.9 ± 5.1	20.6 ± 4.1	44.8 ± 12.1	26.7 ± 6.2
Tc 2 (ms)	24.3 ± 3.5	22.1 ± 3.3	28.3 ± 3.5	39.2 ± 12.0	41.0 ± 5.7^{a}	22.7 ± 5.1^a	50.9 ± 13.3	31.0 ± 7.0
Tc 3 (ms)	24.3 ± 3.5	21.0 ± 3.9	28.4 ± 2.9	37.6 ± 10.2^a	42.3 ± 7.4	24.4 ± 4.7^a	47.1 ± 13.4	32.7 ± 6.5^a
Dm 1 (mm)	8.5 ± 2.0	4.6 ± 1.0	9.8 ± 3.1	6.9 ± 1.8	9.7 ± 2.0	4.6 ± 2.1	5.8 ± 1.2	3.0 ± 0.7
Dm 2 (mm)	8.5 ± 1.7	4.8 ± 1.2	9.7 ± 2.4	8.1 ± 2.3	10.6 ± 2.0	4.7 ± 2.3	6.2 ± 1.9	3.4 ± 0.9
Dm 3 (mm)	9.1 ± 2.2	4.8 ± 1.6	9.8 ± 1.6	7.4 ± 1.4	10.3 ± 1.7	5.7 ± 1.8^{a}	6.5 ± 1.7	3.9 ± 1.0^{a}
Td 1 (ms)	24.0 ± 1.6	23.7 ± 1.8	27.0 ± 1.8	24.3 ± 2.3	26.2 ± 2.3	24.2 ± 2.1	25.5 ± 2.5	24.8 ± 2.6
Td 2 (ms)	24.0 ± 2.0	24.3 ± 2.0	26.9 ± 1.5	26.8 ± 2.6^a	26.8 ± 2.2	23.7 ± 2.0	26.7 ± 3.4	25.6 ± 2.8
Td 3 (ms)	24.0 ± 2.3	24.0 ± 1.9	26.8 ± 1.7	25.8 ± 2.0	25.9 ± 2.4	24.3 ± 1.8	25.5 ± 2.0	25.4 ± 2.2
VrD90 1 $(mm \cdot s^{-1})$	160.6 ± 39.5	93.8 ± 18.6	163.5 ± 61.1	111.0 ± 18.6	135.4 ± 23.8	95.7 ± 35.3	75.2 ± 14.0	53.1 ± 14.0
VrD90 2 $(mm \cdot s^{-1})$	161.1 ± 37.8	93.1 ± 26.4	161.8 ± 48.1	111.2 ± 22.5	142.0 ± 28.9	93.3 ± 36.4	73.9 ± 20.6	55.9 ± 17.2
VrD90 3 $(mm \cdot s^{-1})$	172.3 ± 45.4	97.8 ± 33.2	160.9 ± 30.0	107.4 ± 23.3	137.9 ± 26.1	109.0 ± 26.3	82.7 ± 21.7	61.5 ± 17.2

Discussion

To the best of our knowledge, this study is the first one analyzing the contractile properties in elite futsal players throughout the precompetition and competition season including a period of nine weeks resistance training program (one weekly training session). The aims of this study were: 1) to analyze the seasonal changes in the contractile properties produced in elite futsal players; 2) to determine the possible cumulative effect of a resistance training program on the contractile properties in those futsal players.

Our findings show that there is an increase in Tc, Dm and Td during the season but not uniformly among all the evaluated muscles. In fact, there is no change in the knee extensor muscles (RF, VL and VM) and therefore, our hypothesis is not fully confirmed. On the other hand, an in-season 9-week resistance training program (one day per week) had no effect on the contractile properties of futsal players. That is to say, no changes were found in any TMG parameters between measurement two, and measurement three. In addition, some of the methodological points regarding the present study should be addressed before further discussion. The absolute and relative intraday reliability values of our TMG assessment and estimation of contractile parameters show good reliability (Atkinson, & Neville, 1998) and they are in agreement with previous studies (Macgregor et al., 2018; Martín-Rodríguez et al., 2017a). Futsal is an intermittent sport where there are often changes of pace and direction preceded by short sprints of high intensity (Girard, Micallef, & Millet, 2011; Spencer, Bishop, Dawson, & Goodman, 2005), which significantly requests the stabilizing role of the evaluated muscles that have undergone changes (BF, ST, AL, GM). In addition, fatigue produces alterations in kinetic (Girard, Micallef, & Millet, 2011) and kinematic parameters (Small, McNaughton, Greig, Lohkamp, & Lovel, 2009) which are directly related to the sprint technique. This situation could explain, in part, the changes found in Tc, Dm and Td in BF, ST, AL and GM. García-Manso et al. (2011) and Rodríguez-Ruiz et al. (2012), suggested that there is an adaptation of TMG parameters in the musculature involved in all technical actions of each analyzed sport modality.

There is a considerable increase of Tc in BF, ST, GM and AL during the season. However, knee extensors showed smalls variations. In line with this observation, it has been reported an increase in the Tc for the knee extensors during the season in professional cyclists (García-García et al., 2013) and a decrease after a training program that mainly involved acceleration speed and agility tasks in professional footballers (García-García et al., 2016). In contrast, the knee flexors (BF) showed an increase in Tc and Td (García-García et al., 2016). Impairments in neuromuscular performance (sprint and change of direction) and in TMG properties throughout a season period have also observed in professional soccer players (Loturco et al., 2016). The high volume of specific training performed by the athletes may be the main reason of these impairments in their contractile properties. These findings seem to indicate that the loads of training and competition cause changes in the contractile properties of the involved muscles.

The Tc values obtained from the professional futsal players during the season were slightly lower than those obtained from professional soccer players also taken during the season (García-García et al., 2016) and lower (i.e. VL 21.1 vs. 26.1 ms) compared to those taken at the beginning of the preseason (García-García et al., 2017). They are also logically lower than those obtained for professional cyclists both in preparation and in competition, with differences of almost 20 ms (García-García et al., 2013). All these data have been obtained by using exactly the same measurement protocol. These differences in the Tc of the muscles from professional, futsal and soccer players and cyclists are in line with the lower Tc found in power athletes compared to endurance athletes (de Paula et al., 2016).

Our results revealed a significant increase of Dm in AL and GM throughout the season. By opposite, García-García et al. (2016) observed a decrease in the Dm values of VL, VM and RF after a training program in professional soccer players, mainly focused on acceleration speed and agility tasks during the season. However, in accordance with our findings, these authors also found an increment in Dm in BF muscle throughout the season Dm has been suggested to be an indirect measurement of muscle stiffness (Simunic et al., 2019; Pisot et al., 2008). Notwithstanding, it remains to establish a clear interpretation of how the muscular stiffness evolves in connection with this parameter throughout the competition season, since it has been interpreted that a major Dm is associated with a decrease in muscle tone³⁷. Indeed, the increase in Dm has been previously associated with a decrease in muscle diameter during muscle atrophy induced by bed rest (Pisot et al., 2008), and moreover, it has been correlated with BF muscle thickness decrease after a 6-degree headdown tilt bed rest during 35 days (Simunic et al., 2019). Therefore, it is reasonable to assume that athletes with lower Dm could achieve better physical performance in explosive tasks compared to their counterparts with higher Dm. The Dm values obtained by futsal players are similar to those obtained from professional footballers during the season (García-García et al., 2016) and slightly lower than those obtained by professional soccer players at the beginning of the season (García-García et al., 2017). Our findings indicate that there is no alteration in the contractile properties of the evaluated muscles in professional futsal players after nine weeks of resistance training performed once a week. In addition, although no significant changes were observed in VrD₉₀ throughout the season, an increment of this variable for AL and GL between 2-3 time points (16.8% and 11.9%, respectively) was observed. Conversely, a RT program (one session per week) for one muscle group during eight weeks has been shown to increase the 1RM from the baseline and the muscle thickness in well-trained men, although the effect is greater when the participants trained more than once per week (Schoenfeld, Ratamess, Peterson, Contreras, & Tiryaki-Sonmez, 2015). Very similar results were obtained by Zaroni, Brigatto, Schoenfeld, Braz, Benvenutti, Germano, Marchetti, Aoki, & Lopes. (2019), also in a training period of 8 weeks. A previous study with TMG (Zubac et al., 2017) found that 8-week plyometric training in three weekly sessions improved jumping performance, which was paralleled by a decrease in Tc and Dm. Similar findings were also reported by Zubac, Paravlić, Koren, Felicita, & Simunic (2019), showing that an eight-week plyometric training (three sessions per week) increased the CMJ height together with an improvement in contractile properties of the leg muscles in seniors. Those findings suggest that there are muscle-specific adaptations to the stimuli induced by both, training and competition throughout the season. The small changes in the muscle

properties induced by the resistance training accomplished in the present study could be explained by the low training frequency (one session per week). This fact, along with the intensity and the volume that were carried out, could be an insufficient stimulus to induce positive adaptations on the contractile properties of the muscles. Moreover, the specific position does not seem to modulate neither the effect of the competition season or the effect of the resistance training program performed once a week, which contrasts with other findings where differences were observed in TMG between the specific positions of volleyball players (Rodríguez-Ruiz et al., 2012) and professional soccer players in pre-season (García-García et al., 2017). However, our results are in agreement with those found by Barbero-Alvarez et al. (2008) and Caetano et al. (2015). since they did not find any significant differences in the covered distance or between the work intensities performed by the different specific positions, highlighting the versatility of futsal players. Limitations of the study

From a general point of view, this study had the following limitations: 1) performance evaluations such as maximal strength, jumping or sprints have not been carried out; 2) analysis of physiological parameters (levels of creatine kinase, ammonium, lactate, haemoglobin, etc.) were also not available; 3) finally, no psychophysiological evaluation, such as rate of perceived exertion was carried out. The analysis of the variables mentioned above would provide a more comprehensive evaluation. Although we recognize that this research is limited, our data open new perspectives for monitoring the muscle adaptations induced throughout the season in athletes with high frequency of competitions.

Conclusions

An increase in Tc, Td and Dm during the season was observed in futsal players, but not uniformly among all the evaluated muscles, since no changes were found for the knee extensors (RF, VL and VM). The change in the contractile properties of the evaluated muscles is not modulated by the specific position of the futsal player. In addition, an in-season 9-week resistance training program, once a week, had no significant effect on the contractile properties of futsal players based on the results. It seems not to be enough to have a differentiated effect probably due to insufficient volume, intensity or frequency. Thus, we propose to increase those training parameters for professional futsal players. The control of mechanical and contractile properties of the player's muscles could be useful to the head and strength and conditioning coaches to individualize the training loads and to control the effects of neuromuscular training throughout the season in professional futsal players, using a non-invasive technique that does not induce fatigue and, consequently, does not alter the rhythm of the workouts during the season.

PERSPECTIVAS DERIVADAS DE LOS TRABAJOS DE LA PRESENTE TESIS DOCTORAL

13. PERSPECTIVAS DERIVADAS DE LOS TRABAJOS DE LA PRESENTE TESIS DOCTORAL

En general, los hallazgos de los trabajos señalan que, desde un punto de vista metodológico, sería conveniente usar una duración del estímulo de 1 ms junto con electrodos de dimensiones 5 × 5 cm para asegurar una mayor fiabilidad en la evaluación de las propiedades contráctiles musculares a través de mediciones con TMG. Con este protocolo de obtención de la curva de desplazamiento-tiempo, se ha observado que un protocolo de entrenamiento de fuerza en el ejercicio de sentadilla, consistente en 3 series de 8 repeticiones con una intensidad del 75% de 1RM y un descanso total de 10 minutos, produce una mayor disminución de la Vrd junto con un mayor deterioro del rendimiento físico y estrés metabólico que un protocolo de 6 series de 4 repeticiones con la misma intensidad, volumen de repeticiones y descanso total. Por tanto, series de entrenamiento más largas, resultarían en mayor fatiga, tanto a nivel metabólico como mecánico, y los parámetros de TMG parecen ser sensibles para detectar las diferencias producidas entre ambos protocolos. Finalmente, parámetros de TMG como Tc, Td y Dm permiten evaluar las adaptaciones y los cambios en el sistema neuromuscular de jugadores profesionales de fútbol sala a lo largo de una temporada, ya que estos son sensibles a un deterioro de las propiedades mecánicas musculares que se dan a lo largo de la misma.

En el primer estudio, y como se había indicado previamente en el planteamiento del problema, no se había realizado ningún trabajo que hubiese evaluado la influencia de la duración del impulso y del tamaño del parche sobre las propiedades contráctiles cuando se miden con TMG. En este sentido, en el Estudio 1 de la presente tesis doctoral se ha podido observar una diferencia de un 12% en el Dm del RF entre el parche de 3.2×3.2 cm versus el parche de 5×5 cm (7.63 versus 8.67 mm) y de un 5% en el Dm del VM entre el parche de 3.2×3.2 cm versus el parche de 5×5 cm (8.18 versus 8.65 mm). Además, el parche 5×5 cm y la duración de estímulo de 1 ms permitieron obtener los valores más altos de fiabilidad en la mayoría de los parámetros de TMG analizados. En base a nuestros resultados, se recomienda la utilización de un parche de 5×5 cm en músculos grandes como el RF, VL o VM y una duración del impulso de 1 ms. Por tanto, parece pertinente que en la parte metodológica de los estudios se especifique qué protocolo se ha utilizado para la medición, mencionando el tipo de parche y la duración del impulso.

Por lo que respecta al segundo estudio, se examinaron las respuestas a nivel de rendimiento mecánico y neuromuscular antes diferentes configuraciones de la serie durante el

entrenamiento de fuerza y se pudo comprobar que la configuración de series más cortas (es decir, 6 x 4) dio lugar a un mejor mantenimiento del rendimiento mecánico y, por consiguiente, se obtuvieron marcadores neuromusculares de fatiga más bajos durante el ejercicio en comparación con las configuraciones de series más largas (es decir, 3 x 8). Con respecto a los parámetros de TMG, se observó un descenso del Dm de un 32% en la configuración 6x4 (pre: 6.05; post: 4.09 mm) versus un descenso del 38% en la configuración 3x8 (pre: 6.13; post: 3.77 mm) para el músculo VL. Por el contrario, el descenso observado en Dm para el músculo fue similar para ambas configuraciones de la serie (6x4: -32%, pre: 8.35; post: 6.70 mm versus 3x8: -33%, pre: 8.58; post: 6.61 mm). Por lo que respecta al estrés metabólico y al rendimiento físico, se observó un descenso de un 21% en el CMJ tras la realización de la configuración 6x4 (pre: 37.5; post: 29.6 cm) versus un descenso de un 28% tras la configuración 3x8 (pre: 37.3; post: 27.0 cm). En relación a la concentración de amonio, se produjo un aumento de un 15% tras la configuración 6x4 (pre: 65.1; post: 76.4 umol·l⁻¹) versus a un incremento del 42% tras la configuración 3x8 (pre: 60.5; post: 103.8 umol· l^{-1}): Finalmente, el lactato también aumentó en mayor medida para la configuración de la serie más larga (se pasó de un 83% en la configuración 6x4 (Pre:1.5 post: 8.9 mmol·l⁻¹) versus un aumento de un 86% en la configuración 3x8 (pre: 1.7; post: $12.0 \text{ mmol} \cdot l^{-1}$).

Por último, en el tercer estudio se observó que hay un deterioro de las propiedades mecánicas musculares, a través de un aumento de Tc, Dm y Td, durante la temporada en jugadores profesionales de fútbol sala, pero no de manera uniforme entre todos los músculos evaluados. De hecho, no hubo cambios significativos en los músculos extensores de la rodilla (RF, VL y VM). Asimismo, la inclusión de un entrenamiento de fuerza de una vez por semana, sirvió para evitar un mayor deterioro en estos valores a final de temporada, pero no para recuperar los valores basales.

CONCLUSIONES

14. CONCLUSIONES

La duración de la longitud del estímulo y el tamaño de los electrodos influyen significativamente en las propiedades contráctiles del músculo, medidas mediante TMG. De hecho, el Dm aumenta sustancialmente a medida que aumenta la duración del estímulo y con el uso del electrodo más grande (5 x 5 cm) en los dos músculos evaluados (RF y VM). Asimismo, el parche 5 x 5 cm y la duración de estímulo de 1 ms permitieron obtener los valores más altos de fiabilidad en la mayoría de los parámetros de TMG analizados.

• Esta conclusión afirma en su totalidad la hipótesis Nº 1.

Stimulus pulse duration and electrode size significantly influence on the contractile properties of the muscle measured by TMG. In fact, Dm increases substantially as stimulus pulse duration rises and with the use of the largest electrode (5 x 5 cm) in both muscles tested (RF and VM). Furthermore, the 5 x 5 cm patch and the stimulus duration of 1 ms provided the highest reliability values for most of the TMG parameters analyzed.

• This conclusion fully affirms hypothesis N° 1.

Desde el punto de vista neuromuscular, las configuraciones de series más cortas durante el entrenamiento de fuerza contribuyen a reducir el desarrollo de la fatiga, evaluado tanto de manera voluntaria, a través de la fuerza en sentadilla y salto vertical, como de manera involuntaria, medida a través de TMG, lo que puede deberse, al menos parcialmente, a una menor acumulación de metabolitos.

• Esta conclusión afirma en su totalidad la hipótesis Nº 2.

From a neuromuscular point of view, shorter set configurations during resistance training contribute to a reduction in the development of fatigue, assessed both voluntarily, through squat strength and vertical jump, and involuntarily, measured through TMG, which may be due, at least partially, to a lower accumulation of metabolites.

• This conclusion fully supports hypothesis N° 2.

Durante las mediciones con TMG, se observó un deterioro de las propiedades mecánicas musculares (es decir, un aumento de Tc, Td y Dm) durante la temporada en los jugadores de fútbol sala, pero no de manera uniforme entre todos los músculos evaluados, ya que no se encontraron cambios en los músculos extensores de la rodilla (RF, VL y VM).

• Esta conclusión afirma en parte la hipótesis Nº 3.

During TMG measurements, a deterioration of muscle mechanical properties (i.e. an increase in Tc, Td and Dm) was observed during the season in futsal players, but not uniformly among all tested muscles, as no changes were found in the knee extensor muscles (RF, VL and VM).

• This conclusion partly affirms hypothesis N° 3.

APLICACIONES PRÁCTICAS

15. APLICACIONES PRÁCTICAS

En la última década, el uso del TMG se ha generalizado, sobre todo debido a la información rápida y sencilla que aporta de musculatura aislada, lo cual podría dar lugar a procesos de entrenamiento y recuperación más individualizados. Sin embargo, para que esta herramienta tenga una aplicación práctica real, es imprescindible que el evaluador tenga experiencia en la medición y el análisis de los resultados. Así, los principales hallazgos indican que los investigadores y profesionales que usen TMG deberían elegir una duración de pulso de estimulación de 1 ms junto con un tamaño de electrodo de 5×5 cm para alcanzar una evaluación fiable y reproducible de las propiedades mecánicas contráctiles. Por el contrario, el uso de un electrodo más pequeño o de una duración del estímulo inferior a 1 ms supondría un riesgo para garantizar la reproducibilidad de la medición realizada con la TMG.

Los resultados del Estudio II proporcionan una mayor visión a los profesionales del deporte sobre respuestas a nivel mecánico, muscular y metabólico ante diferentes configuraciones de la serie durante el entrenamiento de fuerza, lo que puede permitir optimizar el diseño de los programas de entrenamiento destinados a mejorar el rendimiento físico relacionado con la producción de fuerza. Los entrenadores deben implementar intervalos de descanso más cortos, pero más frecuentes para aliviar la fatiga neuromuscular, mientras que las configuraciones de ejercicios más largas producirán un aumento del estrés metabólico. Por lo tanto, se podría elegir de antemano una configuración de series determinada, en función del objetivo específico que se persiga.

Por último, los resultados del Estudio III demuestran que el control de las propiedades mecánicas y contráctiles de los músculos de jugadores profesionales de fútbol sala podría ser útil para los preparadores físicos y especialistas en fuerza de cara a individualizar las cargas de entrenamiento y controlar los efectos del entrenamiento a lo largo de la temporada en los jugadores profesionales de fútbol sala, utilizando una técnica no invasiva que no induce a la fatiga y, en consecuencia, no altera el ritmo de los entrenamientos durante la temporada. De hecho, una de las principales aplicaciones prácticas del uso de la TMG, es que permite valorar el estado muscular del deportista sin inducir fatiga adicional sobre el propio entrenamiento o la competición.
LIMITACIONES DE LOS ESTUDIOS

16. LIMITACIONES DE LOS ESTUDIOS

En relación al primer estudio, las limitaciones fueron:

- 1) Se utilizó una intensidad única (100 mA) para poder homogeneizar y comparar sujetos y músculos entre sí. Esto se hizo porque de lo contrario hubiese sido muy difícil la comparación de los resultados entre los músculos de los sujetos. Esto podría haberse resuelto normalizando los valores de cada parámetro de cada sujeto/músculo mediante por ejemplo, una contracción voluntaria máxima. Sin embargo, no se disponía de las herramientas necesarias para ello, lo que constituye una limitación de este estudio. Además, una duración del impulso superior a 1 ms también podría haberse utilizado para tratar de estimular UMs más grandes, pero el software de TMG no lo permitía cuando se realizó el experimento.
- Además, sólo se han evaluado dos músculos, ambos de la extremidad inferior y ninguno de la extremidad superior. Sería interesante incluir músculos más pequeños para ver si los resultados observados serían extrapolables a este tipo de musculatura.

Con respecto al segundo estudio:

- El tiempo transcurrido entre que acaban el protocolo de fuerza y la evaluación con TMG (5 minutos), así como las distintas pruebas de rendimiento físico realizadas en ambos protocolos tras finalizar la prueba (3 saltos verticales, 2 contracciones isométricas máximas y 3 repeticiones ante el 60% 1RM en sentadilla), podrían explicar la ausencia de diferencias significativas entre protocolos para los diferentes parámetros de TMG evaluados, en línea con Tufano et al. (2020). No obstante, el hecho de que el protocolo de evaluación fuese similar para ambas configuraciones de la serie hace que la posible influencia de todas estas mediciones sobre los parámetros de TMG fuese similar para ambos protocolos de entrenamiento.
- Solo se analizó una intensidad del entrenamiento (75% de 1RM) y un ejercicio (sentadilla completa). Sería interesante ver si los resultados observados también serían reproducibles ante intensidades o ejercicios diferentes a los examinados.
- 3) Sólo hubo un momento de medición (a los 5 minutos). Igual si se hubiesen realizado diferentes momentos de medición tras el protocolo se hubiese obtenido más información con respecto a la respuesta de las propiedades mecánicas musculares ante este tipo de

esfuerzos. En este sentido, es importante saber cómo los deportistas recuperan ante distintos esfuerzos para de este modo favorecer los procesos de recuperación

Y por último, las limitaciones del tercer estudio fueron:

1) No se realizaron evaluaciones del rendimiento, como la fuerza máxima, saltos o sprints, ya que no se disponía en el momento de material que permitiese una evaluación precisa del rendimiento en dichos tests.

2) Por último, no se realizó ninguna evaluación psicofisiológica como el índice de esfuerzo percibido para tener información de la carga acumulada por los deportistas a lo largo de la temporada. Aunque reconocemos que esta investigación es limitada, nuestros datos abren nuevas perspectivas para el seguimiento de las adaptaciones musculares inducidas a lo largo de la temporada en deportistas con alta frecuencia de competiciones..

FUTURAS LINEAS DE INVESTIGACIÓN

17. FUTURAS LÍNEAS DE INVESTIGACIÓN

Las futuras líneas de investigación en TMG podrían centrarse en:

- Ahondar en las posibles relaciones existentes entre diferentes parámetros de TMG y porcentajes de fibras musculares. Actualmente hay solo 3 estudios que hayan analizado la correlación entre el porcentaje de fibras tipo I y el parámetro Tc (Simunic et al., 2011; Dahmane et al., 2005; Dahmane et al., 2001). Por lo que sería interesante realizar estudios en los que se observe de manera indirecta la correlación entre el porcentajes de fibras musculares y los parámetros tensiomiográficos.
- Tratar de confirmar los estudios existentes en la literatura científica sobre atrofia e hipertrofia muscular. Con respecto a la atrofia, solo hay dos estudios que han investigación la posible relación entre el Dm y la pérdida de masa muscular (Simunic et al., 2019;. Y en relación a la hipertrofia, también solo hay dos estudios (Kojić, Mandić, & Ilić, 2021; Wilson et al., 2019b) en los que se analicen las adaptaciones en la musculatura tras un programa de entrenamiento de fuerza. Por lo que se hace necesario conocer más a fondo si los cambios en Dm guardan relación con los cambios producidos en el área de sección transversal muscular.
- Cambiar el paradigma de interpretación en la TMG. Es decir, analizar la respuesta muscular a TMG en su totalidad (sumatorio), no músculo a músculo. De esta manera, empezar a realizar sumatorios de los músculos medidos y compararlos con diferentes test de rendimiento físico, como el CMJ, para ver la posible relación existente entre una prueba de rendimiento y las propiedades contráctiles musculares medidas con TMG en su conjunto.

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18. REFERENCIAS

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ANEXO I CONSENTIMIENTO INFORMADO

CONSENTIMIENTO INFORMADO

DECLARACIÓN DE CONSENTIMIENTO INFORMADO

Yo, D		,	mayor	de	edad,	con	D.N.I.
	, domicilio en					у	N.º de
Teléfono							

DECLARO:

Que he sido informado por FRANCISCO JOSÉ PIQUERAS SANCHIZ sobre las posibles consecuencias de la realización de los test físicos de velocidad, saltos y sentadilla completa, así como de los riesgos potenciales y molestias que podrían derivarse de los mismos, a la vez que he podido realizar todas las preguntas que he considerado necesarias, respondiéndome a todas ellas de manera comprensible para mí.

También me ha informado de mi derecho a rechazar el tratamiento o revocar este consentimiento.

Por lo tanto, CONSIENTO en someterme a los protocolos indicados.

Si mi participación puede ser de utilidad científica y para tal fin se publican artículos científicos, autorizo la utilización de los datos reportados durante mis pruebas siempre y cuando se me garantice el más absoluto respeto a mi intimidad y anonimato.

Firma del sujeto

Firma del responsable del estudio

En Sevilla, a _____de _____de 20____

ANEXO II PUBLICACIÓN CIENTÍFICA Y DIVULGACIÓN DE RESULTADOS

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Mechanomyographic Measures of Muscle Contractile Properties are Influenced by Electrode Size and Stimulation Pulse Duration

Francisco Piqueras-Sanchiz¹, Saúl Martín-Rodríguez², Fernando Pareja-Blanco¹, Luis Baraja-Vegas³, Jorge Blázquez-Fernández⁴, Iker J. Bautista⁴ & Óscar García-García⁵⊠

The aim was to determine the effects of changing pulse duration and electrode size on muscle contractile properties. Thirty-six healthy young male participated in the study (age 24.8 \pm 5.8 years; height 178.2 ± 0.6 cm; body mass 71.8 ± 7.3 kg; self-reported weekly moderate intensity activity 3.5 ± 1.2 h·week⁻¹). Tensiomyography was used to assess rectus femoris (RF) and vastus medialis (VM) muscles neuromuscular properties of the dominant leg according to the electrode size (3.2-5 cm) and the stimulus length (0.2, 0.5, and 1 ms). Maximal radial displacement (Dm); Contraction time (Tc); Delay time (Td); Sustained time (Ts) and Half relaxation time (Tr) were measured. Relative and absolute reliability was quantified. To analyze the effects of the electrode and the stimulus length, a repeated-measures analysis of variance was used. Dm and Tc parameters showed for both muscles an excellent relative (0.95-0.99) and absolute reliability (1.6-4.2%). However, Ts and Tr showed low values of absolute reliability (4.4–40.9%). The duration of the stimulus length applied to the RF and VM and electrode size significantly influences muscle's contractile properties (p < 0.05; $\eta_p^2 = 0.09 - 0.60$). The Dm increases substantially as the duration of the stimulus increases and with the use of the larger electrode in both muscles. However, Tc and Td are less affected by both conditions and not entirely clear. Practically, our study suggests that a stimulus pulse duration of 1 ms together with a 5 imes 5 cm electrode is necessary to reach a reliable and reproducible assessment of both RF and VM muscles contractile properties.

Mechanomyography (MMG) is a set of different methods to record mechanical properties, such as muscle's belly displacement and contraction time (Tc), in response to either voluntary or electrically stimulated muscle contraction¹. Among the different MMG methods as vibromyography or sonomyography, tensiomyography (TMG) has received great attention in the last decade by the scientific and clinical community². This method has been utilized in a variety of applications, including estimating myosin heavy chain composition³, determining muscle fiber type populations^{4,5}, detecting stiffness or atrophy^{6,7}, assessing athlete's performance⁸, evaluating muscle fatigue in laboratory or field conditions² or identifying muscle dysfunctions and treatment-effects^{9,10}, among other functions.

TMG is one of the MMG techniques that records muscle contractile properties in response to electrically stimulated muscle contractions, which are evoked through the delivery of transcutaneous neuromuscular stimulations (TNS). Most of TMG protocols determine the maximal TNS with a 'current ramp' protocol. This methodology entails delivering a series of incremental TNS impulses by increasing amperage (mA) whilst keeping a constant voltage and stimulus length until a maximal muscle contraction is reached – as determined by maximal radial displacement (Dm). Little attention has received in the literature one of the aforementioned variables (i.e. stimulus length) of the TNS protocol. The stimulus length, i.e. the duration of the applied TNS stimulation, influences the magnitude of the electrical energy delivered to the muscle. Thus, stimulus length influences the amplitude and timing of the TMG waveforms. Unfortunately, a lack of a standardized protocol has led to a variety

¹Physical Performance & Sports Research Center, Universidad Pablo de Olavide, Seville, Spain. ²Department of Physical Education, University of Las Palmas de Gran Canaria, Las Palmas de Gran Canaria, Spain. ³Department of Physiotherapy, Catholic University of Valencia, Valencia, Spain. ⁴Faculty of Physiotherapy and Podology, Catholic University of Valencia, Spain. ⁵Laboratory of Sports Performance, Physical Condition and Wellness, Faculty of Education and Sport Sciences, Universidade de Vigo, Pontevedra, Spain. ^{Se}e-mail: oscargarcia@uvigo.es

of TNS stimulus length being reported in humans ranging from 200 μ s to 1000 μ s in different muscles¹¹⁻¹³. To our knowledge, one of the few studies that have addressed this question on MMG was published more than 10 years ago¹⁴. These authors tested a wide amount of stimulus length ranging from 50 to 500 μ s, at 50- μ s increments. They showed that the contractile properties of the muscle were considered stable at pulse durations above 300 μ s, which was similar with previously published data for the same muscle¹⁴. Notably, these authors used a laser MMG sensor while TMG uses a contact-displacement sensor (CDS). In this regard, both laser and CDS offer good-to-excellent reliability, although significant systematic bias was identified with the CDS recording higher mean values¹⁵. However, it was previously identified that these differences may not be considered clinically significant. Despite the above, these authors also found that the wide limits of agreement (-19.0 ms and 25.2 ms) identified between half-relaxation time (Tr) (i.e., the time taken from 90 to 50% of Dm) measures, were considered unreliable from a clinical perspective¹⁵. This finding is consistent with data from a contemporary review on the reliability and measurement error of TMG, which concluded that Tr should not be used for clinical or research purposes¹⁶.

Inter-electrode distance (IED) for electrical stimulation is another key factor in the measurements of any MMG device. This parameter has been examined in some studies with several muscles, which have described that IED significantly influences TMG waveform, thus negatively affecting the measure^{17–19}. Despite having studied IED little, the effect of the electrode size on the evoked response has been hardly studied in MMG devices, and only in the field of physical therapy to analyse the thresholds of sensory and motor excitation^{20,21}. These studies observed that larger electrodes required greater voltage output but less pulse-charge density than the smaller electrodes. The above, transferred to TMG, means that electrode size could influence the TMG waveform, thus exhibiting different responses of the contractile properties depending on the size.

Therefore, it is important to increase the accuracy of TMG assessments while minimizing the measurement error to reach reliable data and compare between studies. It is also important for practitioners for the same issue, so they can be able to accurately and objectively compare several intra- and inter-subjects measurements over time.

We hypothesised that the larger the electrode size and stimulus pulse duration allowed by TMG, the greater the data accuracy and the less the measurement error. Therefore, the aims of this study were: 1) to determine the effect of changing the pulse duration on muscle contractile properties and 2) to discriminate if changing electrode size with different pulse duration affects muscle contractile properties.

Methods

Participants. Thirty-six healthy young and moderately active male volunteers (age 24.8 ± 5.8 years; height 178.2 ± 0.6 cm; body mass 71.8 ± 7.3 kg; self-reported weekly moderate intensity activity 3.5 ± 1.2 h·week⁻¹) who had not suffered muscle or tendon injuries in the previous 6 months participated in the study.

The sample size was calculated for each evaluated muscle using the G*POWER software (Heinrich-Heine-Universität Düsseldorf. Germany). The results have showed that for the Dm of the rectus femoris (RF) and vastus medialis (VM), with an alpha of 0.05, a statistical power of 0.80 and an effect size of 0.25, at least 28 and 36 participants were needed respectively. Therefore, a sample size of 36 participants was selected.

Compliance with ethical standards. All voluntary participants were informed of the research objectives and had the possibility to withdraw at any time from the investigation without any penalty. Informed consent was obtained from all individual participants included in the study. The study was conducted during 4 weeks, according to the Declaration of Helsinki, and the protocol was fully approved by the Ethics Committee of the Catholic University of Valencia (UCV2017-2018-73). The authors declare that they have no conflict of interest.

Experimental design. A descriptive cross-sectional design was used in order to analyse the effect of the electrode size and the stimulus duration on the parameters obtained with TMG. All participants measurements were made on the same day, in the same room and under the same temperature and humidity conditions $(23.3 \pm 0.6 \text{ }^{\circ}\text{C} \text{ and } 45 \pm 6.6\% \text{ respectively})$ measured with a weather station using an external sensor. The volunteers were instructed to refrain from moderate or heavy physical activities within 72 h prior the assessments.

Before the data collection, participants were familiar with the electrostimulation stimulus. A 10 min period was established lying face up on a stretcher in order to obtain a muscular relaxation state. All subjects were shaved and the evaluated muscle area was cleaned with alcohol to favour impedance. This was done both in the familiarisation and in the measurement period. New electrodes were used in each measurement using 3.2 or 5 cm self-adhesive circular electrodes. Different measurements were established in the VM and the RF muscles of the dominant leg according to the electrode size (3.2-5 cm) and the stimulus length (0.2, 0.5, and 1 ms), so that 36 measurements were obtained in each participant, 18 in the RF and 18 in the VM. The total duration of the evaluation was \pm 60 min. The intervention conditions were randomised (see Table 1), that is, the muscle evaluation order, the used electrode sequence and the stimulus length was varied. i.e., subject 1 started in the VM with a 5 cm electrode and with 1 ms stimulus length, then 5 cm electrode being pulse duration 0.2 ms and finally electrode 5 cm and stimulus length 0.5 ms.

Contractile properties assessment. TMG was used to assess the neuromuscular properties of both RF and VM muscles of the dominant leg. Measurements were taken under static and relaxed conditions. Prior to performing the measurements, an accurate digital displacement-transducer (GK 40, Panoptik doo, Ljubljana, Slovenia) was perpendicularly positioned at the highest point of the muscle belly. The exact positioning of the electrodes for the RF and VM muscles were in concordance with the recommendations of the SENIAM project²². RF electrodes were placed at 50% on the line from the anterior superior iliac spine to the superior part of the patella and the VM electrodes were placed at 80% on the line between the anterior superior iliac spine and the joint space in front of the anterior border of the medial ligament. To assure the same electrode placement between

Electrode Size	3.2 cm	n			5 cm				
SUBJECT	Block subje	ing cts		SUBJECT	Blocking subjects				
1	A	В	С	1	А	В	С		
2	A	С	В	2	А	С	В		
3	В	А	С	3	В	A	С		
4	В	С	Α	4	В	С	A		
5	С	А	В	5	С	A	В		
6	С	В	Α	6	С	В	А		

Table 1. Blocking subjects. A: 0.2 milliseconds (ms); B: 0.5 ms; C: 1 ms.



Figure 1. TMG parameters extracted of displacement-time curve of Vastus Medialis.

the consecutive measurements, the point was marked with a dermatological pen. To elicit the twitch responses, two circular self-adhesive electrodes with difference sizes (3.2 or 5 cm) (Dura-Stick premium, CEFAR-COMPEX, Hannover, Germany) were connected to an electric stimulator (TMG-S1 doo, Ljubljana, Slovenia) and positioned on the muscle surface, following the arrangement of the fibres. Electrodes were placed symmetrically with a IED of approximately 5 cm, placing the positive electrode in the proximal area of the muscle above the measurement point and the negative electrode in the distal area below the measurement point, according to previous investigations²³. Both RF and VM muscles were measured in the supine position, with the knee joint fixed at a 145° knee flexion angle, once again by means of a wedge cushion designed for such purpose. The electrical stimulation was applied with different duration of the stimulus (0.2, 0.5 or 1 ms) with a current amplitude of 100 mA (i.e., single-twitch). Single-twitch is defined as the contractile response to a single electrical impulse and it is a specific type of evoked muscle activity used to characterise the mechanical properties of a muscle or a single motor unit. Although to identify the maximal required stimulus amplitude, and thus the peak muscle response, a progressive incremental approach has been adopted in most studies on the topic^{2,8}, using in some of them a single-twitch²⁴⁻²⁶. Typically, studies report that the peak response occurs at stimulus amplitudes between 60 and 100 mA, being in larges muscles, such as the lower limbs, much closer to 100 mA². Therefore, the decision to use a single-twitch of 100 mA to homogenize the results derived from the different experimental configurations (i.e., electrode size and stimulation pulse duration). A 60 s rest period was allowed between each electrical stimulus to avoid fatigue or post-tetanic activation while a 120 s rest period was established between conditions. All measurements were taken by the same experienced evaluator.

In order to ensure the reliability of the TMG assessment, two measurements were taken in each participant in all conditions (stimulus length and electrode size). Between one and two minutes passed between the test-retest of each condition.

Each measurement involved recording the following parameters: maximal radial displacement (Dm; mm); contraction time (Tc; ms): Tc as the time from 10% to 90% of Dm; delay time (Td; ms) as the time from onset to 10% of Dm sides of the curve; sustained time (Ts; ms) as the time between 50% of Dm on both the ascending and descending sides of the curve; and Half relaxation time (Tr) was the time from 90% Dm to decline to 50% of the Dm in the relaxation phase²⁷ (Fig. 1).

Statistical analyses. Normal distribution of all variables was determined by Kolmogorov-Smirnov test, together with the Lilliefors test, in order to verify that the sample distribution was normal, linear and homoscedastic. Relative reliability was quantified by the intraclass correlation coefficient (ICC), along with a 95% confidence interval (CI), respect to the two measurements taken in each participant in all conditions. The ICC was calculated using a two-way-mixed effects and absolute agreement model²⁸. ICC values under 0.50 were rated as low reliability, values between 0.50 and 0.75 indicates moderate reliability, values between 0.75 and 0.90 express good reliability and values greater than 0.90 indicates excellent reliability. Absolute reliability indices were expressed through the standard error of the mean (SEM, SEM%), minimum detectable change (MDC, %MDC) and coefficient of variation (CV) along with the respective 95% CI. The CV was calculated for raw data after being log-transformed²⁹. A CV > 10% was interpreted as insufficient absolute reliability^{17,30,31}. The SEM is an index for

	Electrode 3.2 cm																	
		Stimulus length 0.2					Stimulus	length 0.	5				Stimulus length 1					
VM	$M \pm SD$	ICC CI 95%	CV%	SEM SEM%	MDC	MDC%	M±SD	ICC CI 95%	CV%	SEM SEM%	MDC	MDC%	M±SD	ICC CI 95%	CV%	SEM SEM%	MDC	MDC%
Dm (mm)	8.18 ± 1.50	0.97 (0.95- 0.98)	3.0	$\begin{array}{c} 0.24 \pm \\ 2.95 \end{array}$	0.67	8.22	8.50 ± 1.51	0.97 (0.95- 0.98)	3.2	$\begin{array}{c} 0.26 \pm \\ 3.10 \end{array}$	0.73	8.62	$\begin{array}{c} 8.60 \pm \\ 1.36 \end{array}$	0.97 (0.95- 0.98)	3.1	$\begin{array}{c} 0.23 \pm \\ 2.74 \end{array}$	0.65	7.62
Tc (ms)	22.52 ± 2.10	0.95 (0.91- 0.97)	2.2	$\begin{array}{c} 0.49 \pm \\ 2.19 \end{array}$	1.37	6.09	$\begin{array}{c} 22.17 \pm \\ 2.04 \end{array}$	0.95 (0.91- 0.97)	2.3	$\begin{array}{c} 0.47 \pm \\ 2.11 \end{array}$	1.30	5.87	$\begin{array}{c} 22.21 \pm \\ 1.94 \end{array}$	0.95 (0.90- 0.97)	2.2	$\begin{array}{c} 0.43 \pm \\ 1.97 \end{array}$	1.21	5.46
Td (ms)	22.38 ± 1.25	0.85 (0.75- 0.91)	2.3	$\begin{array}{c} 0.50 \pm \\ 2.23 \end{array}$	1.39	6.21	22.23 ± 1.15	0.88 (0.81- 0.94)	1.9	0.39 ± 1.77	1.09	4.92	$\begin{array}{c} 22.24 \pm \\ 1.21 \end{array}$	0.87 (0.79- 0.93)	2.2	$\begin{array}{c} 0.45 \pm \\ 2.02 \end{array}$	1.25	5.62
Tr (ms)	67.10 ± 39.49	0.76 (0.62- 0.86)	23.1	$\begin{array}{c} 21.25\pm\\ 31.66\end{array}$	58.92	87.80	75.95± 47.31	0.85 (0.75- 0.91)	28.7	$\begin{array}{c} 19.77 \pm \\ 26.03 \end{array}$	54.81	72.16	$\begin{array}{c} 71.09 \pm \\ 43.61 \end{array}$	0.76 (0.62- 0.86)	36.6	$\begin{array}{c} 23.69 \pm \\ 33.32 \end{array}$	65.66	92.37
Ts (ms)	168.60 ± 28.60	0.92 (0.87- 0.96)	4.4	$\begin{array}{c} 8.10\pm\\ 4.80\end{array}$	22.46	13.32	$\begin{array}{c} 180.5\pm\\ 27.96\end{array}$	0.91 (0.86- 0.95)	6.2	$\begin{array}{c} 8.29 \pm \\ 4.59 \end{array}$	23.00	12.74	$\begin{array}{c} 175.6\pm\\ 26.84\end{array}$	0.90 (0.83- 0.94)	5.4	$\begin{array}{c} 9.38 \pm \\ 5.34 \end{array}$	26.01	14.81
	Electrode 5 cm																	
Dm (mm)	$\textbf{8.65} \pm \textbf{1.34}$	0.97 (0.94- 0.98)	3.0	$\begin{array}{c} 0.25 \pm \\ 2.89 \end{array}$	0.69	8.04	8.67 ± 1.36	0.96 (0.93- 0.98)	2.9	$\begin{array}{c} 0.31 \pm \\ 3.65 \end{array}$	0.88	10.16	$\begin{array}{c} 8.67 \pm \\ 1.30 \end{array}$	0.95 (0.91- 0.97)	3.8	$\begin{array}{c} 0.26 \pm \\ 3.06 \end{array}$	0.73	8.51
Tc (ms)	21.90 ± 1.98	0.97 (0.95- 0.98)	1.6	0.33 ± 1.53	0.93	4.25	21.50 ± 1.97	0.95 (0.92- 0.97)	2.0	$\begin{array}{c} 0.43 \pm \\ 2.01 \end{array}$	1.20	5.60	$\begin{array}{c} 21.77 \pm \\ 2.01 \end{array}$	0.95 (0.91- 0.97)	2.4	$\begin{array}{c} 0.47 \pm \\ 2.16 \end{array}$	1.30	5.99
Td (ms)	22.21 ± 1.22	0.93 (0.88- 0.96)	1.6	0.33 ± 1.49	0.92	4.15	22.08 ± 1.22	0.90 (0.83- 0.94)	1.6	$\begin{array}{c} 0.40 \pm \\ 1.84 \end{array}$	1.12	5.11	22.06 ± 1.09	0.88 (0.81- 0.93)	1.7	$\begin{array}{c} 0.39 \pm \\ 1.78 \end{array}$	1.09	4.96
Tr (ms)	81.93 ± 47.42	0.85 (0.76- 0.91)	38.1	22.69 ± 27.69	62.90	76.77	75.37 ± 48.70	0.89 (0.81- 0.94)	25.8	$\begin{array}{c} 19.62 \pm \\ 26.03 \end{array}$	54.38	72.15	62.76 ± 40.17	0.80 (0.68- 0.88)	34.1	21.77± 34.68	60.34	96.15
Ts (ms)	175.41 ± 28.64	0.88 (0.81- 0.93)	6.6	$\begin{array}{c} 10.61 \pm \\ 6.04 \end{array}$	29.42	16.77	182.65 ± 30.24	0.89 (0.81- 0.94)	5.5	8.36 ± 4.58	23.19	12.69	$\begin{array}{c} 182.58 \\ \pm 28.43 \end{array}$	0.90 (0.84- 0.95)	5.7	9.09 ± 4.98	25.20	13.80

Table 2. Reliability in tensiomyography parameters for Vastus Medialis muscle (n = 36). Data are mean \pm SD.VM: Vastus Medialis; Dm: displacement; Tc: contraction time; Td: delay time; Tr: half-relaxation time; Ts:sustain time; ms: milliseconds. CV: coefficient of variation; cm: centimeters; ICC: intraclass correlationcoefficient; CI: confidence interval; SEM standard error of measurement; MDC: minimum detectable change.

the precision of a measure and reflects the scores consistency within individual subjects³¹. The calculation of this index was made according Weir³² as follows equation: (SEM = $\sqrt{MS_E}$), where MS_E is the mean square error term from the repeated measures ANOVA. The %SEM, which represents the relative amount of measurement error, was calculated according to Wagner *et al.*³³ as follows equation: (SEM% = SEM/ $M \times 100$), being M the mean of the three TMG measurements. The MDC was calculated as equation: (SEM × 1.96 × $\sqrt{2}$)³². MDC% was expressed as equation: (MDC/ $M \times 100$)³⁰, where M is the mean of the three TMG measurements. To analyse the effect of the electrode size (i.e., 3.2 vs. 5 cm) and the stimulus length (i.e., 0.2, 0.5 and 1 ms), an analysis of the variance of repeated measures (2 × 3 ANOVA) was used. Bonferroni post hoc test with adjustment for 95% confidence interval was used to compare the main effects and identify significant individual differences. The effect sizes in repeated measures ANOVA were reported as partial eta square (η^2_p) and interpreted as small (0.01), moderate (0.06), or large (0.14)³⁴. An alpha level of p < 0.05 was considered statistically significant. All data were analysed using SPSS v24.0 for Windows (SPSS Inc., Chicago, IL, USA).

Results

Dm and Tc parameters showed an excellent relative and absolute reliability with both electrode sizes and with all intensities for both muscles (Tables 2 and 3). Similarly, Td exhibited an excellent relative and absolute reliability in all conditions, except for the 3.2 cm electrode applied to the VM where the relative reliability was good. Ts showed a good to excellent relative reliability in both muscles, together with a good absolute reliability in the VM in all conditions. However, Ts exhibited an insufficient absolute reliability in the RF for all the conditions. Lastly, Tr showed insufficient absolute reliability for all the conditions for both muscles and a moderate to excellent relative reliability for both muscles. Based on the reliability results, Tr and Ts parameters were omitted in the ANOVA due to the insufficient absolute reliability.

As observed in Table 4, the stimulus length and the electrode size modulated the Tc of the VM with a large effect size ($\eta^2_p = 0.163 - 0.438$). However, none of the conditions modulated the Tc of the RF. In the VM, a smaller electrode size caused a longer Tc response (22.30 vs 21.72 ms; 2.6%, p = 0.001 for 3.2 vs. 5 cm, respectively). In addition, a shorter stimulus length caused a longer Tc between 0.2 and 0.5 ms (22.21 vs 21.83 ms; 1.7%, p = 0.001). However, there were no differences in the Tc between stimuli neither of 0.5 and 1 ms nor between 0.2 and 1 ms.

	Electrode 3.2 cm																	
		Stimulus les	ngth 0.2					Stimulus length 0.5					Stimulus length 1					
RF	$M \pm SD$	ICC CI 95%	CV%	SEM SEM%	MDC	MDC%	M±SD	ICC CI 95%	CV%	SEM SEM%	MDC	MDC%	$M \pm SD$	ICC CI 95%	CV%	SEM SEM%	MDC	MDC%
Dm (mm)	7.63 ± 2.10	0.98 (0.97- 0.99)	4.1	$\begin{array}{c} 0.28 \pm \\ 3.77 \end{array}$	0.79	10.46	$\begin{array}{c} 8.60 \pm \\ 2.51 \end{array}$	0.98 (0.97- 0.99)	3.7	$\begin{array}{c} 0.31 \pm \\ 3.68 \end{array}$	0.88	10.23	$\begin{array}{c} 9.23 \pm \\ 2.19 \end{array}$	0.98 (0.97- 0.99)	2.9	$\begin{array}{c} 0.28 \pm \\ 3.09 \end{array}$	0.79	8.59
Tc (ms)	26.14 ± 4.79	0.98 (0.96- 0.99)	3.0	$\begin{array}{c} 0.67 \pm \\ 2.58 \end{array}$	1.87	7.15	$\begin{array}{c} 26.38\pm\\ 4.77\end{array}$	0.98 (0.97- 0.99)	2.4	$\begin{array}{c} 0.57 \pm \\ 2.16 \end{array}$	1.58	6.00	$\begin{array}{c} 26.95 \pm \\ 4.67 \end{array}$	0.98 (0.97- 0.99)	2.4	0.57 ± 2.12	1.59	5.90
Td (ms)	24.50 ± 1.96	0.94 (0.90- 0.97)	2.3	$\begin{array}{c} 0.49 \pm \\ 2.01 \end{array}$	1.35	5.58	$\begin{array}{c} 24.82 \pm \\ 1.90 \end{array}$	0.96 (0.93- 0.98)	1.8	$\begin{array}{c} 0.40 \pm \\ 1.61 \end{array}$	1.08	4.46	$\begin{array}{c} 24.95 \pm \\ 1.93 \end{array}$	0.93 (0.88- 0.96)	2.7	$\begin{array}{c} 0.53 \pm \\ 2.14 \end{array}$	1.48	5.96
Tr (ms)	$\begin{array}{c} 73.77 \pm \\ 39.12 \end{array}$	0.86 (0.77- 0.92)	26.2	$\begin{array}{c} 14.92 \pm \\ 20.23 \end{array}$	41.37	56.09	65.21 ± 40.77	0.90 (0.84- 0.95)	31.1	$\begin{array}{c} 13.26 \pm \\ 20.34 \end{array}$	36.77	56.38	$\begin{array}{c} 73.52 \pm \\ 52.98 \end{array}$	0.96 (0.92- 0.97)	18.8	$\begin{array}{c} 12.00 \pm \\ 16.32 \end{array}$	33.26	45.24
Ts (ms)	117.77 ± 44.88	0.87 (0.78- 0.92)	21.2	$\begin{array}{c} 16.70 \pm \\ 14.18 \end{array}$	46.29	39.35	$\begin{array}{c}105.07\\\pm44.86\end{array}$	0.91 (0.86- 0.95)	18.2	13.66 ± 13.00	37.87	36.04	$\begin{array}{c}109.30\\\pm\ 52.25\end{array}$	0.96 (0.92- 0.97)	12.7	$12.54 \pm \\ 11.47$	34.77	31.79
	Electrode 5 ci	m																
Dm (mm)	8.67 ± 2.24	0.98 (0.97- 0.99)	4.2	$\begin{array}{c} 0.35 \pm \\ 4.10 \end{array}$	0.98	11.39	$\begin{array}{c} 9.53 \pm \\ 2.16 \end{array}$	0.98 (0.96- 0.99)	4.2	0.31 ± 3.29	0.87	9.15	$\begin{array}{c} 10.04 \pm \\ 2.23 \end{array}$	0.98 (0.96- 0.99)	3.5	0.33 ± 3.37	0.93	9.36
Tc (ms)	26.25 ± 4.81	0.97 (0.95- 0.98)	2.7	0.87 ± 3.33	2.42	9.24	$\begin{array}{c} 26.19\pm\\ 4.22\end{array}$	0.98 (0.96- 0.99)	2.6	$\begin{array}{c} 0.61 \pm \\ 2.34 \end{array}$	1.70	6.50	$\begin{array}{c} 26.30\pm\\ 4.12\end{array}$	0.99 (0.98- 0.99)	2.0	0.45 ± 1.72	1.26	4.79
Td (ms)	24.79 ± 1.96	0.93 (0.84- 0.96)	2.0	$\begin{array}{c} 0.53 \pm \\ 2.15 \end{array}$	1.47	5.96	25.14± 1.77	0.94 (0.89- 0.96)	2.0	$\begin{array}{c} 0.45 \pm \\ 1.80 \end{array}$	1.25	5.00	$\begin{array}{c} 25.36 \pm \\ 1.87 \end{array}$	0.94 (0.90- 0.97)	2.3	0.46 ± 1.81	1.27	5.04
Tr (ms)	$\begin{array}{c} 75.64 \pm \\ 49.14 \end{array}$	0.83 (0.73- 0.90)	33.0	$\begin{array}{c} 23.37 \pm \\ 30.89 \end{array}$	64.79	85.66	69.34 ± 53.41	0.82 (0.71- 0.90)	40.9	$\begin{array}{c} 28.75\pm\\ 41.46\end{array}$	79.70	114.94	$\begin{array}{c} 64.96 \pm \\ 51.58 \end{array}$	0.94 (0.89- 0.96)	22	${}^{19.68\pm}_{30.29}$	46.26	71.21
Ts (ms)	110.95 ± 45.82	0.87 (0.79- 0.93)	20.3	17.10 ± 15.41	47.40	42.72	$\begin{array}{c}102.17\\\pm52.92\end{array}$	0.83 (0.73- 0.90)	22.9	26.90 ± 26.32	74.56	72.98	$\begin{array}{r} 97.92 \pm \\ 51.98 \end{array}$	0.94 (0.89- 0.96)	14.2	16.94 ± 17.29	46.97	47.97

Table 3. Reliability in tensiomyography parameters for Rectus Femoris muscle (n = 36). Data are mean \pm SD. RF: Rectus Femoris; Dm: displacement; Tc: contraction time; Td: delay time; Tr: half-relaxation time; Ts: sustain time; ms: milliseconds. CV: coefficient of variation; cm: centimeters; ICC: intraclass correlation coefficient; CI: confidence interval; SEM standard error of measurement; MDC: minimum detectable change.

The Dm of the VM moderately increased ($\eta^2_{\rm p}$ =0.089) as the stimulus length increased, although only significantly between the duration of 0.2 and 0.5 ms (8.41 vs 8.58 mm, 2.0%, p = 0.025) (Fig. 2). The Dm of the RF also increased as the stimulus length increased with a large effect size ($\eta^2_{\rm p}$ =0.606) (Fig. 3). In addition, it also increased 10.4% (p = 0.001) between 0.2 and 0.5 ms duration, 7.0% (p = 0.001) between 0.5 and 1 ms and 18.2% (p = 0.001) between 0.2 and 1 ms. The use of the largest electrode (5 cm) caused a greater Dm than using the smaller one (3.2 cm) in both the VM (2.8%, p = 0.036) and RF (10.8%, p = 0.001). In addition, the interaction stimulus length x electrode size moderately ($\eta^2_{\rm p}$ =0.117) modulated the Dm of the VM, which occurred with all stimulus length (8.18–8.65 mm at 0.2 ms; 8.50–8.67 mm at 0.5 ms; 8.60–8.67 mm at 1 ms; p = 0.013).

The Td of the VM was only significantly affected by the size of the electrode. The use of a larger electrode (5 cm) caused a lower Td (22.28 vs 22.11 ms, 0.7%, p = 0.023). However, the Td of the RF increased as the stimulus length increases with a large effect size ($\eta^2_p = 0.237$). It increased by 1.4% (p = 0.006) between 0.2 and 0.5 ms duration, 0.7% (p = 0.036) between 0.5 and 1 ms and 2.1% (p = 0.001) between 0.2 and 1 ms. In addition, the electrode size also modulated the Td of the RF with a large effect size ($\eta^2_p = 0.327$), so that the use of the largest electrode (5 cm) caused longer Td (1.4%, p = 0.001).

Discussion

Summarizing, the results of this study show that the stimulus length (duration of the TNS pulse) applied to the RF and VM muscles and the size of the electrode significantly influence the muscle's contractile properties as measured by a single-twitch TMG technique. The main finding is that Dm increases significantly both, as the duration of the TNS increases and with the use of a larger electrode (5 cm) in both muscles. However, Tc and Td are less affected by both conditions and not entirely clear.

Our results are consistent with previous reviews on the topic showing that Tr parameter should not be used in research or clinical environments due to their insufficient absolute reliability and high measurement error^{2,16}. Similarly, Ts parameter should not be used in these areas since it has shown a CV higher than 10%¹⁷. On the contrary, Dm, Tc and Td parameters have shown a high relative reliability in all muscles and situations evaluated, which is in line with the findings of Lohr *et al.*³⁵ but also an absolute reliability through a low CV, %SEM and %MDC, as opposed to the results of Lohr *et al.*³⁵ which indicates a discordant absolute reliability in the Dm and moderate positive in the Tc. Hence, the effect of stimulus length and electrode size on Tr and Ts was not examined, since these parameters did not meet the reliability requirements.

The electrical energy available to stimulate the muscle is influenced by the duration of the applied electrical impulse as shown by the equation²⁰: $(E = V \times I \times \Delta t)$, where E is the electrical energy, V are volts, I equals current and Δ t is the duration of the TNS pulse. It appears that, at stimulus length below 1 ms, there is insufficient electrical energy available to maximally stimulate all motor units³⁶. Moreover, given that smaller motor units are easier to stimulate than larger motor units^{37,38}, shorter stimulus length appear to favor type I slow twitch muscle fibers over type II fast twitch muscle fibers, which are more difficult to activate³⁶. Our results match those found

	EFFECT	F	Df	Р	η^2
	stimulus length*	6.812	2	0.002	0.163
Tc VM	electrode size *	27.244	1	0.001	0.438
	stimulus length $ imes$ electrode size	1.341	2	0.268	0.037
	stimulus length	1.526	2	0.225	0.042
Tc RF	electrode size	0.836	1	0.367	0.023
	stimulus length $ imes$ electrode size	1.774	2	0.177	0.048
Dm VM	stimulus length*	3.418	2	0.038	0.089
	electrode size *	4.757	1	0.036	0.120
	stimulus length $ imes$ electrode size *	4.628	2	0.013	0.117
	stimulus length*	53.793	2	0.001	0.606
Dm RF	electrode size *	49.926	1	0.001	0.588
	stimulus length $ imes$ electrode size	0.675	2	0.513	0.019
	stimulus length	2.041	2	0.138	0.055
Td VM	electrode size *	5.694	1	0.023	0.140
	stimulus length $ imes$ electrode size	0.082	2	0.922	0.002
	stimulus length*	10.865	2	0.001	0.237
Td RF	electrode size *	17.002	1	0.001	0.327
	stimulus length \times electrode size	0.386	2	0.681	0.011

Table 4. Effects of different stimulus length and electrode size on different tensiomography parameters.Interactions between stimulus length (0.2, 0.5 and 1 ms), and electrode size (3.2 vs. 5 cm). *p < 0.05.</td>



Figure 2. Displacement-time curves of Vastus Medialis of the dominant leg, according to the electrode size (3.2–5 cm) and the stimulus length (0.2, 0.5, and 1 ms).



Figure 3. Displacement-time curves of Rectus Femoris of the dominant leg, according to the electrode size (3.2–5 cm) and the stimulus length (0.2, 0.5, and 1 ms).

by McAndrew *et al.*¹⁴, who reported that stimulus length above 0.3 ms provide both a maximal lateral displacement of the muscle's belly and stable measures of its contractile properties. Although the previous authors used a laser-based MMG technique, their results can be extrapolated to ours since it has been demonstrated that both laser- and CDS shows good-to-excellent reliability for the assessment of muscle contractile properties with no significant differences between them¹⁵. In fact, it should be noted that there are data showing that the radial muscle displacement (Dm) increases linearly with muscle torque up to 68% of maximal voluntary contraction⁶. This fact may suggest that, maintaining linearity, using shorter pulse duration (0.2 ms) could elicit low torque production, affecting to the wave-form and then to all the TMG parameters. To our knowledge, to date there has been no study that has analysed the effects of the electrodes size for muscle electrical stimulation measured by TMG. However, it has been previously analysed in transcutaneous electrical nerve stimulation (TENS)³⁹. This author used 4 types of electrodes: 3×3 , $6 \times 69 \times 9$ and 5×16.2 cm on the quadriceps with the objective of examining the effects of electrode size at neural and motor level, reaching the conclusion that the larger the electrode, the greater the participation of motor units. Our results seem to be in agreement with those obtained by Alon³⁹ with respect to the electrode size, since both VM and RF increased their Dm with the largest electrode. A reasonable explanation is that a larger electrode is able to recruit more motor units on the transverse axis and therefore greater amount of muscle mass displacement directly influences an increase in Dm. However, this hypothesis will still have to be proven.

On the other hand, the overall %SEM and %MDC results of both muscles seem to point out, in a general way, that increasing electrode size and pulse duration could increase the data accuracy and minimize the measurement error, although it would be necessary more research with other muscle groups to verify this point. *A priori*, to minimize measurement errors with TMG parameters, it should be recommended standardizing TMG protocols to develop a TMG standard operating procedure such that experimental studies may be comparable. In order to do this, based on the present findings and previous research, it is recommended: (1) to take into account the correct stimulus length (1 ms) to maximize Dm; (2) to use electrodes of 5 cm; (3) to use specific guides for each muscle of inter-electrode distance^{18,19}; and (4) additional recommendations for a correct measurement protocol are described in the review of Macgregor *et al.*². In this line, Lohr *et al.*³⁵ have indicated that it is necessary to have high methodological standards for conduct and reporting TMG studies, and these recommendations could be helpful for this purpose.

Limitations, strengths and practical applications

Finally, it is important pointing that, the maximal TNS voltage required to produce peak lateral displacement of a muscle's belly is unique to each muscle and is determined as the voltage that first produces an undistorted parabolic MMG waveform of maximal amplitude¹⁴. In this regard, we used a unique intensity (100 mA) in our study to be able to homogenize and compare subjects and muscles with each other. This was done because the current and well accepted ramp protocol^{2,8}, used to decide individual intensity would have made it difficult to compare results between subjects' muscles. This could have been solved by normalizing the values of each parameter of each subject/muscle to, for example, a maximum voluntary contraction. However, we did not have the necessary tools for this purpose, what is a limitation of this study. In addition, a duration of TNS above 1 ms could also have been used to try to stimulate larger motor units, but the TMG software did not allow this when the experiment was performed. Furthermore, it has only been evaluated two muscles, both of the lower limb, which may be another limitation of this study. On the other hand, the main strength of the present study is the sample size and the use of several set configurations for both electrode size and stimulus pulse duration, and as well as that the same evaluator with more of 7 years' experience took all measurements. In terms of practical applications, TMG researchers and practitioners can base their measurement protocol on the findings of this study. Our data indicates that a stimulation pulse duration of 1 ms together with the election of a 5 \times 5 cm electrode size is necessary to reach a reliable and reproducible assessment of both RF and VM muscles contractile properties. Conversely, the use of a smaller electrode or a stimulus length of less than 1 ms would be a risk to guarantee the reproducibility of the measurement taken with TMG.

Conclusions

The duration of the stimulus length applied to the RF and VM muscles and electrode size significantly influence the muscle's contractile properties as measured by a single-twitch TMG technique. In fact, the Dm increases substantially as the duration of the TNS increases and with the use of the larger electrode (5 cm) in both muscles. However, Tc and Td are less affected by both conditions and not entirely clear. Therefore this study indicates that a stimulus pulse duration of 1 ms together with the election of a 5 \times 5 cm electrode size is necessary to reach a reliable and reproducible assessment of both rectus and vastus medialis muscles contractile properties.

Data availability

The TMG recordings utilized in the current study will be uploaded to an open access server and will be available to anyone who would like to re-analyse them.

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Author contributions

F.P.S., F.P.B., I.J.B. and O.G.G. conceived, coordinated, and designed the study. S.M.R. made suggestions that improved the design and wrote the manuscript together with F.P.S. in equal conditions. L.B.V., J.B.F. and F.P.S. carried out the experimental part. O.G.G. and I.J.B. analyzed data. F.P.B. and O.G.G. contributed to edit and review manuscript while helped in the drafting of the manuscript. All authors have read and approved the current manuscript.

Competing interests

The authors declare no competing interests.

Additional information

Correspondence and requests for materials should be addressed to Ó.G.-G.

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Acute Mechanical, Neuromuscular, and Metabolic Responses to Different Set Configurations in Resistance Training

Francisco Piqueras-Sanchiz,¹ Pedro J. Cornejo-Daza,¹ Juan Sánchez-Valdepeñas,^{1,2} Beatriz Bachero-Mena,³ Miguel Sánchez-Moreno,³ Saúl Martín-Rodríguez,⁴ Óscar García-García,⁵ and Fernando Pareja-Blanco^{1,2}

¹Department of Sports and Computers Sciences, Physical Performance & Sports Research Center, Pablo de Olavide University, Seville, Spain; ²Department of Sports and Computers Sciences, Faculty of Sport Sciences, Pablo de Olavide University, Seville, Spain; ³Department of Physical Education and Sports, University of Seville, Seville, Spain; ⁴Department of Physical Education, University of the Gran Canarian Palms, The Gran Canarian Palms, Spain; and ⁵Laboratory of Sports Performance, Physical Condition and Wellness, Faculty of Education and Sport Sciences, University of Vigo, Pontevedra, Spain

Abstract

Piqueras-Sanchiz, F, Cornejo-Daza, PJ, Sánchez-Valdepeñas, J, Bachero-Mena, B, Sánchez-Moreno, M, Martín-Rodríguez, S, García-García, O, and Pareja-Blanco, F. Acute Mechanical, Neuromuscular, and Metabolic Responses to Different Set Configurations in Resistance Training. J Strength Cond Res XX(X): 000–000, 2021—The aim of this study was to investigate the effect of set configuration on mechanical performance, neuromuscular activity, metabolic response, and muscle contractile properties. Sixteen strength-trained men performed 2 training sessions in the squat exercise consisting of (a) 3 sets of 8 repetitions with 5 minutes rest between sets (3 \times 8) and (b) 6 sets of 4 repetitions with 2 minutes rest between sets (6 \times 4). Training intensity (75% one repetition maximum), total volume (24 repetitions), total rest (10 minutes), and training density were equalized between protocols. A battery of tests was performed before and after each protocol: (a) tensiomyography (TMG), (b) blood lactate and ammonia concentration, (c) countermovement jump, and (d) maximal voluntary isometric contraction in the squat exercise. Force, velocity, and power output values, along with electromyography data, were recorded for every repetition throughout each protocol. The 6 × 4 protocol resulted in greater mechanical performance (i.e., force, velocity, and power) and lower neuromuscular markers of fatigue (i.e., lower root mean square and higher median frequency) during the exercise compared with 3×8 , particularly for the last repetitions of each set. The 3 × 8 protocol induced greater lactate and ammonia concentrations, greater reductions in jump height, and greater impairments in TMG-derived velocity of deformation after exercise than 6×4 . Therefore, implementing lower-repetition sets with shorter and more frequent interset rest intervals attenuates impairments in mechanical performance, especially in the final repetitions of each set. These effects may be mediated by lower neuromuscular alterations, reduced metabolic stress, and better maintained muscle contractile properties.

Key Words: rest-redistribution, lactate, ammonia, rate of force development, electromyography, tensiomyography

Introduction

Fatigue can be defined as an integral process resulting in a temporal decline in force production capacity (3). A better understanding of the mechanical and physiological mechanisms underlying fatigue development during resistance training (RT) sessions is essential to improve our knowledge of strength training methodology. Excessive fatigue development during training sessions may be detrimental for athletes focused on maximizing neuromuscular adaptations (29,30). Shorter set configurations that include rest periods between clusters of repetitions are an effective strategy to attenuate fatigue and maintain mechanical performance (i.e., force production, movement velocity, and, as a consequence, power output) during RT sessions (36). In addition, higher blood lactate (7,8,24) and ammonia (26) concentrations, hormonal response (growth hormone and cortisol) (26,27,37), and muscle damage indicators (i.e., creatine kinase) (26) have been observed after longer set configurations.

Previous literature analyzing set configuration has focused on the effects on exercise performance itself, with limited studies examining the postexercise responses to different set configurations. In this regard, longer set configurations are characterized by inducing greater impairments in back squat (SQ) performance (i.e., velocity attained against a given absolute load) and jump height (24,26). However, the acute effect of set configuration on relevant markers for the evaluation of fatigue in strength training as maximal isometric force (MIF) or maximal rate of force development (RFDmax) remains unexplored. Likewise, recording electromyography (EMG) activity (amplitude, through root mean square "RMS," and frequency, through median frequency "MDF") may provide a better understanding of the mechanisms behind the changes in mechanical performance, such as muscle activation and neuromuscular fatigue accumulated throughout the training session (6). To date, only one previous study has examined neuromuscular activity during different set configurations involving lower-body muscles (25), which compared 6 sets

Address correspondence to Fernando Pareja Blanco, fparbla@upo.es. Journal of Strength and Conditioning Research 00(00)/1–9 © 2021 National Strength and Conditioning Association

of 6 repetitions at 20% of 1 repetition maximum (1RM) in loaded jumps, continuously (n = 9) or with a 30-second rest every 2 repetitions (n = 9). These authors observed larger increments in RMS throughout longer set configurations; however, both protocols induced similar decrements in MDF (25). However, the fact that different subjects performed each protocol may have obscured potential differences in the EMG spectral parameters between protocols. Finally, EMG recordings have limitations during dynamic muscle contractions (5). Therefore, it would be reasonable to examine the effects of set configuration on neuromuscular fatigue development during both dynamic and isometric contractions.

Tensiomyography (TMG) has been validated for assessing in vivo passive muscle contractile properties through simple measurement of the muscle belly radial deformation and the time it takes to occur in response to a single-twitch stimulus (39). This technique allows the assessment of the changes in muscle contractile properties induced by fatigue after training sessions (4,31,32). Despite evidence indicating TMG as a valid and reproducible tool to screen adjustments in skeletal muscle contractile characteristics (23), to the best of our knowledge, only one study has analyzed the acute effects on TMG outcomes after different set configurations (38). These authors compared 4 isokinetic unilateral knee extension protocols, 2 different set configurations (4 \times 10 with 95 seconds of interset rest vs. 20 \times 2 with 15 seconds of interset rest) at 2 different velocities (60 vs. $360^{\circ} \cdot s^{-1}$), reporting similar postexercise TMG parameter patterns for all protocols (38). The fact that this study was conducted on an isokinetic device improved the control of confounding variables but sacrificed the ecological validity of real-life resistance exercises. Therefore, information about the acute effects of set configuration in strength training protocols used in practice on muscle contractile properties is lacking in the literature. In this regard, the SQ is one of the most widely used and effective RT exercises for strengthening the lower limbs and improving athletic performance (12). In addition, TMG measurements should be accompanied by postexercise mechanical performance tests to better comprehend how these outcomes interact. In light of these considerations, a more detailed knowledge about the integral response (mechanical, neuromuscular, and metabolic) to different set configurations would provide coaches and scientists with a better understanding of the effects of set configuration manipulation and could lead to further advances in exercise prescription. Therefore, the purpose of this study was to investigate the effect of set configuration on mechanical performance and neuromuscular activity throughout the SQ training session, as well as the acute mechanical, neuromuscular, metabolic, and muscle contractile responses for strengthtrained men.

Methods

Experimental Approach to the Problem

A randomized cross-over research design was undertaken to examine the acute mechanical, neuromuscular, metabolic, and muscle contractile properties responses to 2 different set configurations with a load of 75% 1RM during the SQ exercise: (a) 3 sets of 8 repetitions with 5 minutes rest between sets (3×8) and (b) 6 sets of 4 repetitions with 2 minutes rest between sets (6×4). Training intensity (75% 1RM), total volume (24 repetitions), total rest (10 minutes), and, as a consequence, training density (work-to-rest ratio) were equalized between protocols. Protocols were performed in a random order, separated by a period of 4 days. To compare the mechanical, neuromuscular, metabolic, and muscle contractile properties response, subjects underwent a battery of tests before and after each protocol: (a) TMG measurements, (b) blood lactate and ammonia concentration, (c) countermovement jump (CMJ), and (d) maximal voluntary isometric contraction (MVIC) in SQ exercise (Figure 1). In addition, to compare the performances attained throughout the session, force, velocity, and power output values along with EMG data were recorded for every repetition.

Subjects were asked to abstain from any strenuous physical activity for at least 2 days before each trial. Subjects were also instructed to maintain their usual diet, although they were asked to refrain from consuming caffeine 12 hours before attending the study. All sessions took place at a neuromuscular research laboratory under the direct supervision of the researchers, at the same time of the day for each subject and under similar environmental conditions (20° C and 60% humidity, approximately).

Subjects

Sixteen strength-trained men (Age range: 18–35 years; age 23.4 \pm 4.4 years; height 1.75 \pm 0.05 m; and body mass 73.9 \pm 9.1 kg; mean \pm *SD*) with at least 2 years of RT experience in the SQ exercise (range 2–6 years; 1RM strength for the SQ exercise: 105.8 \pm 12.1 kg; and 1.44 \pm 0.11 normalized per kg of body mass) participated in the study. Subjects were injury-free and were fully informed about the procedures, potential risks, and benefits of the study, and they all signed a written informed consent form before the tests. Subjects reported they were not taking drugs, medications, or dietary supplements known to influence physical performance. This study was approved by the Research Ethics Committee of the University of Vigo (Ref: 03-819), in accordance with the Declaration of Helsinki.

Procedures

Progressive Loading Test. One week before the resistance exercise protocols, a progressive loading test was conducted on a Smith machine (Multipower Fitness Line, Peroga, Murcia, Spain) with no counterweight mechanism to obtain the 1RM strength and individualized load-velocity relationships. The SQ was performed with subjects starting from the upright position with the knees and hips fully extended and stance approximately shoulder-width apart, and the barbell resting across the back at the level of the acromion. Each subject descended in a continuous motion as low as possible (~35-40° knee flexion) and then immediately reversed motion and returned to the upright position. Unlike the eccentric phase that was performed at a controlled mean velocity (~0.50-0.65 $m \cdot s^{-1}$), subjects were required to always execute the concentric phase at maximal intended velocity. Range of movement and velocity values of all repetitions were recorded at 1,000 Hz with a linear velocity transducer (T-Force System, Ergotech, Murcia, Spain). First, the subjects warmed up by performing 6 repetitions with a 20 kg load. The initial load was set at 30 kg and was progressively increased in 10 kg increments until the mean propulsive velocity (MPV) was $\leq 0.50 \text{ m} \cdot \text{s}^{-1}$. Then, the load was increased with smaller increments (2.5-5.0 kg) for better adjustments. A total of 10.0 ± 1.8 increasing loads were used for each subject. Three repetitions were executed for light loads ($\geq 1.00 \text{ m} \cdot \text{s}^{-1}$), 2 for medium loads (1.00–0.80 m $\cdot \text{s}^{-1}$), and one for the heaviest loads ($\leq 0.80 \text{ m} \cdot \text{s}^{-1}$). Interset rests

179



were 3 minutes for light and medium loads and 5 minutes for heavy loads. Only the best repetition (i.e., highest MPV) with each load was considered for subsequent analysis. The propulsive phase corresponds to the portion of the concentric action during which the measured acceleration is greater than acceleration due to gravity $(-9.81 \text{ m} \cdot \text{s}^{-2})$ (35).

Resistance Exercise Protocol. Figure 1 provides a detailed timeline description of the experimental protocol. To minimize the effects of fluid changes caused by walking, subjects remained lying down for 10 minutes before starting the TMG measurements and baseline data acquisition. During the time they were lying down, a resting blood sample was collected (lactate and ammonium) and electrode locations (for TMG and EMG) were marked. After baseline TMG and blood lactate measurements were taken, the CMJ and MVIC tests were performed. Then, a standardized SQ warm-up was performed before the resistance exercise protocol, which consisted of: 6-6-4-3-2 SQ repetitions with 20 kg, 40, 50, 60, and 70% of 1RM, respectively, with 3 minutes rest between loads. Relative loads were determined from the individual second-order load-velocity relationship (R^2) $= 0.996 \pm 0.002$) obtained from the progressive loading test. Subsequently, the corresponding protocol was performed (3×8) with 75% 1RM and 5 minutes rest between sets vs. 6×4 with 75% 1RM and 2 minutes rest between sets). The SQ execution technique described in the "progressive loading test" section was carefully reproduced in all repetitions performed in the study. A force plate (FP-500, Ergotech) synchronized with a linear velocity transducer (T-Force System, Ergotech) was installed on the Smith machine (Multipower Fitness Line, Peroga, Murcia, Spain) to record mean propulsive values of force (MPF), velocity (MPV), and power (MPP) for every repetition. In addition, EMG data (i.e., RMS and MDF) were also recorded throughout the 24 repetitions. Subjects received immediate velocity feedback while being encouraged to perform each repetition at maximal intended velocity. After completing the final repetition (the 24th), the battery of tests was repeated as follows: CMJ (20 seconds Post), MVIC (50 seconds Post), blood samples (2 minutes and 30 seconds Post), and TMG (5 minutes Post) to obtain the acute postexercise values. This order was chosen to minimize the interference between tests and record valid data; i.e., the acute response to mechanical performance (but not highfatiguing tests), metabolic response about 2-3 minutes postexercise, and TMG after 4-5 minutes resting. The duration per subject and session was approximately 1 hour.

Tensiomyography. The contractile properties of the vastus lateralis (V_{LA}) and vastus medialis (V_{ME}) muscles of the right leg were assessed using a TMG device (TMG-100 System electrostimulator; TMG-BMC, Ljubljana, Slovenia) to determine their response to an electrically evoked contraction. The electric stimulus was applied through 2 self-adhesive electrodes (5 \times 5 cm, Dura-Stick premium; Cefar-Compex, Hanover, Germany) placed at a 5-cm interelectrode distance. The muscle mechanical response was measured with a digital DC-DC transducer Trans-TekR (GK 40, Ljubliana, Slovenia) placed perpendicular to the muscle belly and equidistant from the self-adhesive electrodes at a distance of 25-30 mm. Subjects remained lying in a supine position for 10 minutes before starting the TMG data acquisition, and the V_{LA} and V_{ME} were marked according to SENIAM indications and location (16). To ensure the same placement of electrodes between consecutive measurements, the locations were marked on the skin using a permanent marker and subjects were advised to keep the marks in place until the second session. Measurements were taken with the athletes in the supine position and the knee joint fixed at an angle of ~140° using a wedge cushion. Electrical stimulation was applied with a pulse duration of 1 ms and an initial current amplitude of 40 mA, which was progressively increased in 10 mA steps up to the stimulator's maximal output (100 mA). The use of a stimulus pulse of 1 ms, using a 5×5 -cm electrode (the procedure followed in this study) has been recently found to be necessary to reach a reliable and reproducible assessment of muscle contractile properties (33). A 10-second rest period was allowed between each electrical stimulus to avoid fatigue or posttetanic activation. The variables assessed in this study were the maximum radial displacement of the muscle belly (Dm), contraction time (Tc), and delay time (Td). Dm was defined as the peak amplitude in the displacement-time curve of the tensiomyographical twitch response; Tc was obtained by determining the time interval from 10 to 90% of Dm; and Td was defined as the time between the electrical stimulus and 10% of Dm (39). In addition, the velocity of deformation (Vd) was calculated as follows: Dm $(Tc + Td)^{-1}$ (21). Although Vd was originally termed the velocity of contraction, it is now recommended to use the term velocity of deformation, which is mainly dependent on muscle stiffness, to avoid confusion with sarcomere shortening velocity (40). Mean velocities of muscle contraction $(\text{mm}\cdot\text{s}^{-1})$ from the onset of electrical stimulation until 10% (V10) and 90% (V90) of Dm were also recorded using equations developed elsewhere (4). All measurements were performed by the same experienced evaluator and only the curve with the highest

180
Table 1
Mechanical and neuromuscular characteristics of each resistance
exercise protocol (average of 24 repetitions).*†

	3 × 8	6 × 4	р
MPF (N)	842.9 ± 96.4	869.4 ± 116.9	0.03
MPV (m·s ^{−1})	0.59 ± 0.08	0.63 ± 0.08	0.02
MPP (w)	486.4 ± 66.8	524.8 ± 74.5	0.001
RMS (%)	122.4 ± 15.8	108.9 ± 11.1	0.01
MDF (%)	108.4 ± 9.6	110.5 ± 10.4	0.45

*3 × 8 = protocol consisted of performing 3 sets of 8 repetitions with 75% of 1RM; $6 \times 4 =$ protocol consisted of performing 6 sets of 4 repetitions with 75% of 1RM; MPF = mean propulsive force; MPV = mean propulsive velocity; MPP = mean propulsive power; RMS = root mean square averaged from the vastus medialis and vastus lateralis muscles; MDF = median frequency averaged from the vastus medialis and vastus lateralis muscles.

 $\pm Data are mean \pm SD$, n = 16.

Dm value was considered for further analysis. Test-retest reliabilities for TMG measures, using the intraclass correlation coefficient (ICC) with 95% confidence intervals (CIs) and coefficient of variation (CV) values, were as follows: Dm (ICC [95% CI]: 0.98 [0.96–0.99], CV: 5.3%); Tc (ICC [95% CI]: 0.98 [0.95–0.99], CV: 3.2%); Td (ICC [95% CI]: 0.95 [0.89–0.98], CV: 2.3%); Vd (ICC [95% CI]: 0.98 [0.97–0.99], CV: 4.6%), V10 (ICC [95% CI]: 0.97 [0.95–0.99], CV: 5.3%), and V90 (ICC [95% CI]: 0.98 [0.97–0.99], CV: 4.6%).

Metabolic Variables. After cleaning the skin, 5 and 20 μL of capillary blood from fingertip punctures were used for lactate and ammonia quantification, respectively. Lactate was measured using a portable lactate analyzer (Lactate Pro 2, Arkray, Kyoto, Japan), and ammonia was analyzed using an ammonia checker (Blood Ammonia Meter PocketChem, model BA PA-4140; Arkray, Kyoto, Japan). Both devices were calibrated before each exercise session according to the manufacturer's specifications.

Countermovement Jump. An infrared timing system (OptojumpNext, Microgate, Bolzano, Italy) was used for determining jump height. The CMJ was performed with both hands on the waist while performing a downward movement to about 90° of knee flexion, followed by a maximal vertical jump. All subjects were instructed to land in an upright position and to bend the knees after landing. The subjects were required to do 2 trials separated by 10 seconds, and the mean height was determined. A specific warm-up was performed, consisting of 5 minutes of jogging at a self-selected easy pace, 2 sets of 10 squats without external load, 5 submaximal CMJs, and 3 maximal CMJs. Testretest reliability values were ICC (95% CI): 0.99 (0.97–0.99) and CV: 1.9%.

Maximal Voluntary Isometric Contraction Test. Kinetic and EMG data were measured during an MVIC in the SQ exercise with the subjects standing with their knees flexed at 90° (180° = full extension) measured with a handheld goniometer. This test was performed on a Smith machine instrumented with 2 telescopic bar holders, with a precision scale placed at the left and right sides of the Smith machine to precisely replicate the individual positions between trials. The subjects were instructed to maintain a constant minimum pretension until the experimenter gave the following verbal instruction: "Push against the ground as hard and as fast as possible" (34). Two 5 seconds trials, separated by 30 seconds rest, were performed. The warm-up protocol

consisted of 2 attempts at 70 and 90% of perceived effort with 30 seconds rest between them.

Kinetic Data. External forces were collected at a sampling rate of 1,000 Hz with an 80 \times 80-cm dynamometric platform (FP-500; Ergotech) and processed with specific software (T-Force System; Ergotech). Maximal isometric force was defined as the maximal strength value attained during the MVIC. RFDmax was calculated as the maximal slope of the force-time curve measured in 20-ms time intervals. The average value of each variable in the 2 attempts was recorded for further analysis. Test-retest reliability values for MIF and RFDmax were as follows: ICC (95%CI): 0.99 (0.97; 0.99) and 0.94 (0.86; 0.97) and CV: 3.4 and 13.8%, respectively.

Electromyography Signal Acquisition. Surface EMG electrodes were placed on the same location previously described for TMG measurements. Electromyography signals were recorded continuously during MVIC testing using a parallel-bar, bipolar surface electromyographic sensor Trigno wireless EMG system, with an interelectrode distance of 10 mm, common mode rejection ratio >80 dB, and bandwidth filter between 20 and 450Hz \pm 10% (Delsys, Inc., Natick, MA). Baseline noise was $<5 \mu$ V peak to peak, and sampling rate was 2,000 Hz. The raw data from the EMG were stored in digital format using EMGworks Acquisition software (Delsys, Inc.). From each isometric and dynamic contraction the highest averaged (over sliding windows of 500 ms with an overlap of 499 ms) RMS and MDF values for each muscle were recorded. V_{ME} and V_{LA} muscle excitation values were averaged, and the average of the 2 MVICs was calculated for further analysis. The value of the signal from MVIC at pretraining of each resistance exercise protocol was used to normalize the EMG parameters. The test-retest reliability for RMS measures was ICC (95% CI): 0.95 (0.90-0.98) and CV: 7.4% and for MDF was ICC (95% CI): 0.95 (0.90-0.98) and CV: 5.3%.

Statistical Analyses

Values are reported as mean \pm SD. Sample size was calculated (using GPower version 3.1.9.4) introducing the following parameters: effect size (ES) 0.85 and 0.70 for mean velocity and mean power between-protocol comparisons based on a recent meta-analysis comparing different RT set configurations (20) and α error probability (0.05) and power (0.95), which resulted in a sample size of 6 and 14 subjects, respectively. Statistical significance was established at $p \le 0.05$. The test-retest absolute reliability was measured by the SEM, which was expressed in relative terms through CV. The SEM was calculated as the root mean square of the total mean square intrasubject. The relative reliability was assessed using the ICC calculated with the oneway random-effects model and its 95% CI. A paired sample ttest was conducted to compare the average values attained during each protocol (averaged 24 repetitions). A 2 (protocol) \times 24 (repetitions) repeated measures analysis of variance (ANOVA) with Bonferroni's post hoc adjustments was performed to compare differences between protocols throughout the repetitions completed. In addition, a 2 (protocol) \times 2 (Pre vs. Post) repeated measures ANOVA with Bonferroni's post hoc comparisons was performed to analyze the acute response to each protocol. In addition, pre-post ES values were calculated using Hedge's g on the pooled SD (13) using a purpose-built spreadsheet. The rest of statistical analyses were performed



Figure 2. Evolution of mechanical performance throughout the 24 repetitions for both protocols. A) Mean propulsive force; B) mean propulsive velocity; and C) mean propulsive power. Data are mean \pm *SD*, $n = 16.3 \times 8$: protocol consisted of performing 3 sets of 8 repetitions with 75% of 1RM; 6×4 : protocol consisted of performing 6 sets of 4 repetitions with 75% of 1RM. Within-protocol significant differences with respect to the first repetition: *p < 0.05. Significant differences between protocols at the corresponding repetition: #p < 0.05.

using SPSS software version 20.0 (SPSS, Inc., Chicago, IL). Figures were designed using SigmaPlot 12.0 (Systat Software, Inc., San Jose, CA).

RESULTS

Descriptive Characteristics of the Resistance Exercise Protocol. Table 1 shows the mechanical and neuromuscular characteristics of each protocol. Higher MPF, MPV, and MPP values were obtained for the 6×4 configuration compared with 3×8 . In addition, the 3×8 protocol exhibited significantly higher RMS during the session than the 6×4 protocol, with no significant differences for MDF. Figure 2 shows the evolution of mechanical parameters throughout the 24 repetitions for each resistance exercise protocol. Significant "protocol × repetitions" interactions (p < 0.001) were observed for all mechanical variables. Performance in these variables progressively decreased throughout the 24 repetitions for both protocols. However, performance in these parameters (i.e., MPF, MPV, and MPP) was higher for the 6 × 4 protocol compared with the 3 × 8 protocol. Figure 3 depicts the development of neuromuscular variables throughout the 24 repetitions for each resistance exercise protocol. Significant "protocol × repetitions" interactions were observed for RMS (p = 0.001) and for MDF (p =



averaged from the vastus medialis and vastus lateralis muscles; and B) median frequency averaged from the vastus medialis and vastus lateralis muscles. Data are mean \pm SD, $n = 16.3 \times 8$: protocol consisted of performing 3 sets of 8 repetitions with 75% of 1RM; $6 \times 4 =$ protocol consisted of performing 6 sets of 4 repetitions with 75% of 1RM. Data were normalized with respect to the first repetition of each resistance exercise protocol. Within-protocol significant differences with respect to the first repetition: *p < 0.05. Significant differences between protocols at the corresponding repetition: #p < 0.05.

0.002). The 3 \times 8 configuration resulted in higher RMS and lower MDF values than the 6 \times 4 protocol, particularly for the final repetitions of each set.

Tensiomyography. No significant "protocol × time" interactions were found for TMG-derived parameters (Table 2). A significant "time effect" was observed for all variables, except for V_{LA}-Tc and V_{ME}-Td, with significant decreases in Dm and Vd (in both muscles) and V_{LA}-Td for both protocols at Post. However, the 3 × 8 protocol showed significantly lower values of V_{LA}-Vd, V_{LA}-V10, and V_{LA}-V90 than the 6 × 4 protocol at Post.

Metabolic Response and Jump Performance. Significant "protocol × time" interactions were observed for CMJ height and blood lactate and ammonia values (Table 3). The 3×8 protocol induced higher lactate and ammonia concentrations and CMJ height impairments than the 6×4 protocol at Post.

Mechanical and Neuromuscular Response during Maximal Voluntary Isometric Contraction. Significant "protocol × time" interactions were noted for neuromuscular parameters (i.e., RMS: p = 0.02 and MDF: p = 0.002) (Figure 3). However, no "protocol × time" interactions were noted for mechanical outcomes (i.e., MIF and RFDmax) (Table 3). The RMS attained during MVIC decreased for both protocols at Post, although lower values were observed for the 6×4 protocol. However, the 6×4 protocol induced significant increases in MDF at Post while the 3 \times 8 protocol exhibited significantly lower MDF than the 6×4 protocol. Moreover, both MIF and RFDmax significantly decreased at Post, with no significant differences between protocols.

Discussion

Integral responses to different set configurations were examined during (mechanical and neuromuscular features) and after exercise (mechanical, neuromuscular, metabolic, and muscle contractile properties). The results indicate that redistributing long sets and interset rest periods into shorter but more frequent sets modulates training stimuli. Specifically, shorter set configurations (i.e., 6×4) resulted in greater mechanical performance maintenance and concomitant lower neuromuscular markers of fatigue during the exercise compared with longer set configurations (i.e., 3×8). Longer set configurations also led to greater metabolic responses, along with greater impairments in jump height and muscle deformation velocity (i.e., Vd, V10, and V90) after exercise. Finally, an overall impairment in isometric force production was observed for both protocols, with no significant differences between them.

Table 2
iffects of different resistance exercise protocols on muscles' contractile properties assessed by tensiomyography.**

	3 × 8			6 × 4				
	Pre	Post	ES	Pre	Post	ES	p-value time effect	p -value protocol \times time
VLA-Tc (ms)	21.80 ± 3.75	22.66 ± 3.57	0.25	21.48 ± 2.84	22.39 ± 3.37	0.26	0.16	0.95
VME-Tc (ms)	21.90 ± 2.83	22.87 ± 3.47	0.32	21.85 ± 2.73	23.04 ± 2.93‡	0.39	0.04	0.63
VLA-Td (ms)	22.82 ± 1.66	21.36 ± 1.84‡	-0.87	22.48 ± 1.64	$20.93 \pm 1.43^{\$}$	-0.92	0.004	0.89
VME-Td (ms)	22.00 ± 1.36	22.09 ± 2.17	0.06	21.74 ± 1.14	21.35 ± 1.42	-0.24	0.72	0.41
VLA-Dm (mm)	6.13 ± 1.62	3.77 ± 1.57	-1.37	6.05 ± 1.98	4.09 ± 1.58	-1.14	< 0.001	0.29
VME-Dm (mm)	8.58 ± 1.63	6.61 ± 1.54	-1.25	8.35 ± 1.59	6.70 ± 1.45	-1.05	< 0.001	0.29
VLA-Vd (mm·ms ⁻¹)	0.138 ± 0.038	0.084 ± 0.029,¶	-1.71	0.135 ± 0.040	0.093 ± 0.030	-1.42	< 0.001	0.10
VME-Vd (mm·ms ⁻¹)	0.196 ± 0.038	0.149 ± 0.039	-1.35	0.193 ± 0.035	0.152 ± 0.034	-1.08	< 0.001	0.39
VLA-V10 (mm·ms ⁻¹)	0.027 ± 0.007	0.017 ± 0.006,¶	-1.49	0.027 ± 0.008	0.020 ± 0.007	-0.91	< 0.001	0.12
VME-V10 (mm·ms ⁻¹)	0.039 ± 0.007	0.030 ± 0.008	-1.16	0.038 ± 0.007	0.032 ± 0.007 §	-0.83	< 0.001	0.38
VLA-V90 (mm·ms ⁻¹)	0.124 ± 0.033	0.076 ± 0.026,¶	-1.57	0.123 ± 0.036	0.084 ± 0.028	-1.18	< 0.001	0.13
VME-V90 (mm·ms ⁻¹)	0.177 ± 0.034	0.134 ± 0.035	-1.21	0.173 ± 0.031	0.136 ± 0.031	-1.16	< 0.001	0.38

 $*3 \times 8 =$ protocol consisted of performing 3 sets of 8 repetitions with 75% of 1RM; $6 \times 4 =$ protocol consisted of performing 6 sets of 4 repetitions with 75% of 1RM; $V_{LA} =$ vastus lateralis muscle; $V_{ME} =$ vastus medialis muscle; $T_C =$ contraction time; Td = delay time; Dm = muscle displacement; Vd = velocity of deformation radial (Dm/(Tc + Td); V90 = velocity of deformation radial to 90% of Dm; V10 = velocity of deformation radial to 10% of Dm; ES = within-protocol effect size from Pre to Post.

+Data are mean \pm *SD*, n = 16.

 \pm Intraprotocol significant differences from Pre to Post: p < 0.05.

§Intraprotocol significant differences from Pre to Post: p < 0.01.

Intraprotocol significant differences from Pre to Post: p < 0.001.

¶Significant differences between protocols: p < 0.05

Therefore, introducing shorter but more frequent interset rests attenuates impairments in mechanical performance, especially in the final repetitions of each set, which may be mediated by lower neuromuscular alterations, reduced metabolic stress, and better maintained muscle contractile properties.

Performance as indicated by mechanical parameters (i.e., force, velocity, and power) progressively decreased throughout the repetitions for both protocols, although shorter but more frequent sets alleviated fatigue-induced impairments in performance during the session. There is compelling evidence that including rest periods between repetitions or clusters of repetitions is an effective strategy to attenuate fatigue and ameliorate loss of mechanical performance during this phenomenon have not been clearly established. One of the potential mechanisms associated with the higher acute

performance attained using shorter set configurations is that the frequent rest periods may allow for greater maintenance of phosphocreatine (PCr) stores, a partial resynthesis of adenosine triphosphate (ATP), and increased metabolite clearance in the working muscles (9,36). In agreement with previous studies (10,26), the longer set configuration (i.e., 3×8) induced higher blood ammonia levels. An increase in blood ammonia concentrations during high-intensity exercise is interpreted as indicative of accelerated ammonia production by muscles, resulting from the deamination of adenosine monophosphate to inosine monophosphate (14). In this regard, Gorostiaga et al. (9) observed greater depletion of intramuscular ATP and PCr stores after 5 \times 10 vs. 10 \times 5 with 85% 1RM with 2 minutes interset rests in the leg press exercise, along with concomitant greater power reductions. Likewise, the higher lactate values observed for the longer set configuration suggest

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Mechanical,	neuromuscular,	and metabolic	response to the	different resistar	nce exercise protoco	is under study.*†
,						

	3 × 8			6 × 4				
	Pre	Post	ES	Pre	Post	ES	p time effect	p protocol × time
Metabolic response and jump								
performance								
Lactate (mmol·I ⁻¹)	1.7 ± 0.6	12.0 ± 3.8§	3.31	1.5 ± 0.7	8.9 ± 4.2§	2.38	< 0.001	0.05
Ammonia (µmol·l ⁻¹)	60.5 ± 16.6	103.8 ± 44.9§	1.21	65.1 ± 20.4	76.4 ± 44.1	0.31	0.001	0.02
CMJ (cm)	37.3 ± 4.6	27.0 ± 3.8§	-2.56	37.5 ± 3.6	29.6 ± 3.4 §	-1.96	< 0.001	0.02
Maximal voluntary isometric contraction								
MIF (N)	1,267.7 ±	1,034.5 ±	-0.93	1,270.0 ± 279.0	1,017.3 ± 216.8§	-1.01	< 0.001	0.62
RFDmax (N·s ⁻¹)	$4,609 \pm 1,528$	238.79 3,998 ± 1431 [‡]	-0.38	5,242 ± 2006	3,924 ± 1224§	-0.82	0.001	0.09

 $*3 \times 8 =$ protocol consisted of performing 3 sets of 8 repetitions with 75% of 1RM; $6 \times 4 =$ protocol consisted of performing 6 sets of 4 repetitions with 75% of 1RM; Lactate = blood lactate concentration; Ammonia = blood ammonia concentration; CMJ = countermovement jump height; MIF = maximal isometric force; RFDmax = maximal rate of force development; ES = within-protocol effect size from Pre to Post

+Data are mean \pm SD, n = 16.

‡Intraprotocol significant differences from Pre to Post: p < 0.01.

§Intraprotocol significant differences from Pre to Post: p < 0.001

Significant differences between protocols at the corresponding time point: p < 0.05.

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a greater reliance on anaerobic glycolysis for energy for these set structures (19,27). Accordingly, the lower blood ammonia and lactate concentrations observed after the shorter set configurations (i.e., 6×4) may indicate better replenishment of ATP and PCr stores, as well as reduced glycolytic requirements within each set, which may result in a greater ability to maintain mechanical performance.

In an attempt to provide a better understanding of the mechanisms behind the different performances achieved with longer and shorter set configurations, we recorded the EMG activity attained in each repetition. Our data suggest that longer set configurations induced higher neuromuscular fatigue (i.e., higher RMS and lower MDF values) during isometric (i.e., MVIC) and dynamic contractions, mainly during the final repetitions in each set (Figure 3). In agreement with our findings, Ortega-Becerra et al. (28) reported higher RMS and higher MDF values during traditional sets (3 sets of 12 repetitions at 60% of 1RM with interset rests of 2 minutes) in the bench press exercise compared with protocols using similar training intensities and volume but including cluster configurations (30-second rest every 4 or 2 repetitions). Fatigue-induced alterations in neuromuscular markers have been attributed to metabolic byproduct accumulation (18). Specifically, increased RMS values may be primarily due to increased muscle activation (i.e., recruitment of higherthreshold motor units, motor unit firing frequency, or changes in intrinsic muscle properties) attempting to compensate for the loss of force in the fatigued state (1,18), whereas decreased MDF values may be evoked by reduced action potential conduction velocity associated with a decline in intramuscular pH (2), changes in action potential shape (15), and decreases in the firing rate of fatigued fast motor units (1). In this regard, it is fair to assume that interpretation of mechanistic information from EMG data is speculative. Nevertheless, our data suggest that from a neuromuscular standpoint, shorter set configurations also contribute to reduced fatigue development, which may be due to lower metabolic byproduct accumulation.

Besides the effects of set configuration on exercise performance itself, it is also important to consider the residual mechanical fatigue induced by these training sessions. In line with the fatigue levels observed during the training session, greater reductions in CMJ height were observed after the longer set configuration. Previous studies have also shown smaller CMJ height losses after training sessions with intraset rest periods compared with traditional structures (8,24). However, the exercise-induced fatigue on isometric strength (i.e., MIF and RFDmax) and several TMG-derived parameters (i.e., Dm, Tc, and Td) was similar for both set configurations. Decreases in Dm after RT have been associated with impaired muscle function (17), muscle swelling, and exercise-induced muscle damage (11), whereas it has been suggested that temporal TMG parameters should be treated with caution (22). On the other hand, the 3×8 protocol induced greater impairments in muscle velocity of deformation (i.e., Vd, V10, and V90) than the 6×4 protocol (Table 2). The only study that has previously examined the acute effects of different set configurations (4 \times 10 with 95 seconds of interset rest vs. 20×2 with 15 seconds interset rest) on TMG outcomes did not observe significant differences in TMG parameters during postexercise measurements, although the ES values indicated a lower impaired muscle function for shorter set configurations (38). Therefore, longer set configurations induced greater impairments in neuromuscular ability and muscle function in both voluntary (i.e., squats and jumps) and involuntary (i.e., TMG) actions.

Despite using similar loads (75% 1RM), total volume (24 repetitions), total rest (10 minutes), and, therefore, training density (work-to-rest ratio), implementing shorter but more frequent interset rest intervals allowed for better maintenance of performance, especially in the final repetitions of each set, along with lower alterations of neuromuscular markers of fatigue, a dampened metabolic stress, and a lesser worsening of muscle velocity of deformation. Therefore, rest-redistribution may be a viable strategy for maintaining performance during a training session, which may be mediated by lower neuromuscular alterations, reduced metabolic stress, and better maintenance of muscle contractile properties.

Practical Applications

This study provides a greater insight for sport professionals about the integral responses (mechanical, neuromuscular, and metabolic) to different set configurations, which may allow us to optimize the design of RT programs aimed at enhancing physical performance. Coaches can implement shorter but more frequent rest intervals to spare mechanical performance and alleviate neuromuscular fatigue, whereas longer set configurations could be used to increase metabolic stress. Therefore, a given set configuration could be chosen beforehand, depending on the specific training goal being pursued. Further studies should analyze the long-term effects of different configurations on mechanical, neuromuscular, and hypertrophic adaptations.

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186

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ORIGINAL ARTICLE EXERCISE PHYSIOLOGY AND BIOMECHANICS

Evolution of contractile properties of the lower limb muscles throughout a season in elite futsal players

Francisco PIQUERAS-SANCHIZ¹, Luis M. MARTÍNEZ-ARANDA², Fernando PAREJA-BLANCO^{1,3}, David RODRÍGUEZ-RUIZ⁴, Óscar GARCÍA-GARCÍA⁵*

¹Faculty of Sports Science, Pablo de Olavide University, Seville, Spain; ²Neuroscience of Human Movement Research Group, Faculty of Sport, Catholic University San Antonio, Murcia, Spain; ³Physical Performance and Sports Research Center, Department of Sports and Computers Sciences, Pablo de Olavide University, Seville, Spain; ⁴Department of Physical Education, University of Las Palmas, Gran Canaria, Spain; ⁵Sport Performance, Physical condition and Wellness Lab, University of Vigo, Pontevedra, Spain

*Corresponding author: Óscar García-García, Sport Performance, Physical condition and Wellness Lab. University of Vigo, Campus A Xunqueira s/n 36005, Pontevedra, Spain. E-mail: oscargarcia@uvigo.es

ABSTRACT

BACKGROUND: Futsal is a team sport involving intermittent technical actions of high intensity, and high physical (strength) and muscular demands. In this regard, the tensiomyography (TMG) is a useful and non-invasive tool for the monitoring and assessment of the muscle's contractile capacity. This study aimed to analyze the changes in the contractile properties produced during the season, as well as to determine the potential cumulative effect of a resistance training (RT) program in futsal players.

METHODS: Fourteen elite futsal players (2 goalkeepers, 4 defenders, 4 wingers and 3 pivots) were assessed by TMG at 11th, 18th, and 28th week of the season. The maximal radial displacement of the muscle belly (Dm); contraction time (Tc); delay time (Td) and radial displacement velocity (90%) Dm (VrD90) were assessed. After the second measurement, a RT program was included in the regular training sessions and focused on the lower body musculature. It was performed during 9 weeks (1-weekly). Finally, a third measurement was performed between 28th-29th weeks. Repeated measures analysis of variance was used to detect in-season changes. Two factors were included: Time (changes detected after resistance training program) was used as the within-subject factor and the specific position was used as the between-subject factor. RESULTS: An increment in Tc for several muscles: biceps femoris (BF; P=0.02), semitendinosus (ST; P=0.04), adductor longus (AL; P=0.008) and gastrocnemius medialis (GM; P=0.009) was observed throughout the season. Similarly, significant increments in Dm for GM (P=0.02) and AL (P=0.05), as well as increments in Td for BF (P=0.002) were found. Moreover, no significant changes for any of the variables analyzed. CONCLUSIONS: An increase respect to baseline levels was observed for Tc, Td and Dm during the season. However, the adaptations to contractile properties were muscle specific. In addition, an in-season 9-week RT program (1-weekly), had no significant effects (time points 2-3) on the contractile properties of futual players. In addition, there were no differences when comparing different positions.

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KEY WORDS: Resistance training; Football; Exercise.

Futsal is growing in popularity and more than one million futsal players have already been registered by national federations worldwide. Futsal demands intermittent high-intensity actions which require high physical, technical and tactical efforts from the players.^{1, 2} As a consequence, players are required to undertake extensive training during different phases of the season aiming to

improve both, endurance and power-speed abilities.^{3, 4} It is not uncommon to observe an impairment of physical performance (*e.g.*, vertical jump height and sprint speed) over periods of high volumes of training and competition likely due to the "interference effect" of aerobic and strengthpower stimuli.⁵ In fact, team sports players including futsal players can suffer from the concurrent training effects

Vol. 60 - No. 7

PIQUERAS-SANCHIZ

RESISTANCE TRAINING PROGRAM IN FUTSAL PLAYERS

and become slower and less powerful during some phases of the preparation.^{6, 7} Likewise, the absence of specific strength training programs can impair sprint and strength performance throughout the competitive season in soccer players,⁸ whereas implementing strength training during the competitive season in a group of high-level amateur soccer players, was able to improve the peak strength and the rate of force development without altering sprinting speed.⁹ However, it is not easy to implement physical tests too frequently in order to monitor relevant fitness changes, as maximal tests can be time-demanding and increase the risk of undesired injuries.

An alternative to the monitoring of the fitness status in team sport players could be the use of the tensiomyography (TMG). TMG is a non-invasive tool that measures muscle belly enlargement in transversal plane during an isometric twitch contraction to provide mechanical indices of the muscle contraction properties that are related to performance changes. Maximal displacement (Dm), delay time (Td) and contraction time (Tc) are among the main TMG-derived parameters.¹⁰ These parameters have shown good to excellent reliability and low measurement error.10 In addition, both Dm and radial displacement velocity until 10% (VrD10) and 90% (VrD90) of Dm have been considered as indirect markers of muscle fatigue^{11, 12} For instance, the rectus femoris Dm, which is inversely related to muscle stiffness, was acutely increased after a repeated-sprint ability test protocol in futsal players, indicating reduced muscle stiffness.13 Furthermore, this increasing change in response to the protocol (pre- to post) was negatively correlated to the performance change across the sprints (r=-0.49). In a longitudinal observation, it was found that in soccer players the radial displacement velocity (VrD) was impaired during the inter-season period, concomitant with reductions in sprinting speed.⁶ Another longitudinal observation in professional soccer players found was that Tc-Td-Dm significantly decreased $(\sim 10\%)$ after 10 weeks.¹⁴ In professional road cyclists, the contractile properties were also analyzed in two periods: preparation and competition, aiming to detect differences, before and after the official competitive season, in the contractile properties of the main musculature involved in this kind of endurance discipline. The Tc values during the competitive period showed marked differences between the knee extensors (large increased) and the knee flexor (large decreased), whereas the Dm did not change.¹⁵ Taken together, TMG provides some advantages: 1) involuntary contractions, which avoid the influence of factors such as motivation; 2) it does not affect performance for subsequent training sessions; 3) it allows the assessment of individual muscles in the field.¹⁰ These results support the notion that TMG indices can be useful to monitor muscle contraction properties that can influence performance in team sports players. However, these indices have not been investigated yet in professional futsal players, especially in response to a period of resistance training.

Resistance training has been proposed as an effective training method to improve performance in team sports.¹⁶ Its implementation has been shown to enhance the change of direction and speed,^{17, 18} the linear speed and the vertical jump capacity,^{17, 19} in addition to a reduced risk of injuries in team sports players.²⁰ In line with this, a recent study has shown a Dm decreased in vastus lateralis, biceps femoris, tibialis anterior, gastrocnemius medialis and gastrocnemius lateralis after eight weeks of plyometric training.²¹ In spite of the widely described seasonal changes in physical performance in team sport athletes, less is known about the seasonal changes in the neuromuscular abilities along with the adaptation of contractile properties to systematic resistance training in team sport athletes. Therefore, the aims of the present study were: (I) to analyze the seasonal changes in the contractile properties produced in elite futsal players; (II) to determine the possible cumulative effect of resistance training program on the contractile properties in these futsal players. We hypothesize that there are impairments in the neuromuscular abilities of the main lower limbs muscles which are involved during the season and that, during the resistance training program, those changes will be modulated.

Materials and methods

Study design

A longitudinal and intragroup comparative study was conducted to assess in-season changes in the contractile properties of the musculature evaluated in professional futsal players. Players were divided according to their specific position: goalkeeper, defender, winger and pivot. Three neuromuscular evaluations were conducted during the competitive season using TMG. Data collection started at the 11th week of the season (five pre-season weeks and six competition weeks) with an average of 82±61 competitive min played per evaluated player and an average of accumulated training minutes of 6,703±653 minutes since the start of the preseason. Those values were established as a baseline. The second measurement was performed between the 18th-19th week of the season (accumulated competitive minutes: 193±82 min and accumulated training: 16,812±833 min).

July 2020

PIQUERAS-SANCHIZ



Figure 1.-Experimental design and testing structure.

After this second measurement, an intervention based on resistance training program focused on the leg muscles was carried out during nine weeks (one weekly training session). Next, a third measurement was carried out between the 28th-29th week (accumulated competitive minutes: 302 ± 54 and an accumulated training minutes: $24,577\pm551$ min) (Figure 1). All players participated on average in ~10 hours of futsal training (5 sessions) plus 1 competitive match per week and the weekly distribution of training contents did not show significant variations throughout the season, with a solid work related to the training load (volume and intensity) between microcycles (Table I).

All evaluations were conducted at the club facilities under the same environmental conditions. Using the TMG, eight muscles were tested in both legs: Adductor Longus (AL), Biceps Femoris (BF), Gastrocnemius Lateralis (GL), Gastrocnemius Medialis (GM), Rectus Femoris (RF), Semitendinosus (ST), Vastus Lateralis (VL) and Vastus Medialis (VM). Before each testing session, players complied with the following pretest guidelines: not to consume any energy/performance enhancing drinks, supplements or beverages containing caffeine or alcohol 3 hours before testing, as well as not to eat any food at least 2 hours before testing. Evaluations were held on the same weekday, after the recovery day and before the next training session. No player exhibited abnormal fatigue symptoms or muscle damage.

Subjects

Fourteen elite futsal players $(27.2\pm3.3 \text{ years}, 74.2\pm5.6 \text{ kg})$ and $180.2\pm4.8 \text{ cm}$ with no previous injuries during the past three months, and who complied with at least 85% of all training sessions participated in this study. Regarding the position, the players were two goalkeepers, four defenders, four wingers and three pivots. All the players belonged to the same team in the Spanish Professional Futsal League. One player missed too many training sessions or was absent from the post testing session due to injury or illness.

Ethical statement

All participants provided a written consent after being informed about the research process and the potential risks associated with TMG assessment. The research protocol followed the principles of the Declaration of Helsinki regarding biomedical research involving human subjects (64th WMA General Assembly, Fortaleza, Brazil, 2013). The Local Ethical Research Committee, as well as the coaching staff and management board of the professional futsal team approved the study.

Procedures

Resistance training program

The training took place one day per week over a period of 9 weeks. Each session had a duration of 45-60 min and was carried out always during the evening (20:30h-21:30h). Resistance bands, kettlebell, weights and platform as well as a Flywheel device Kbox® (inertia 0.035 kg·m²) were used in this study. All training sessions were preceded by a 10 min standardized dynamic warm-up (jogging, high knee skipping and running, lunges with trunk rotation, straightleg skipping, straight-leg deadlift walk, deep squatting and bilateral jumps), core stability exercises (plank, side plank and bridging) and dorsiflexion. The main exercises in the training program are presented in the Figure 2 as follows: unipodal hip extension with resistance band, hip adduction with a resistance band, Kbox inertial bipodal calf, prevention tendinopathy unipodal, half squat, resisted acceleration with elastic bands and jumps in stable and unstable

TABLE I.—Distribution of the training and competition load in a seasonal microcycle.

	Monday	Tuesday	Wednesday	Thursday	Friday	Saturday	Sunday		
Morning	-	TEC-TAC	-	-	-				
Afternoon	PRT + CO+ TEC- TAC	CORE + PROP + MAP + TEC-TAC	TEC-TAC	ACTIVATION + TEC-TAC	MATCH	FREE	FREE		
PRT: Preventive resistance training; CO: coordination; TEC-TAC: technical-tactical; CORE: core training; PROP: proprioception; MAP: maximum aerobic power.									

PIQUERAS-SANCHIZ



Figure 2.—Exercises performed during training program and workloads. A) Hip extension resistance unipodal band (48 kg~); B) hip adduction with an elastic unipodal band); C) kbox inertial bipodal; D) prevention of unipodal tendinopathy (kettlebell) (6-8 kg); E) barbell squat (22-26 kg); F) acceleration/ deceleration resistance band accelerator (48 kg~); G) Jump stable/unstable conditions.

conditions. Two sets of eight repetitions were performed per exercise with 30 s of resting between exercises and 1 min between sets, except to Kbox inertial calf bipodal, in which was performed 2x8 with 60 s resting between exercises and 2 min between sets. All the exercises were executed in the same order in every session. The main researcher controlled every training session, providing verbal encouragement to each participant.

The premises in Kbox exercise were: concentric phase was performed as fast as possible. The eccentric phase of

the bipodal calf required decelerating the movement because the flywheel keeps spinning due to its inertia. On the contrary, exercises employing resistance band were carried out by performing the concentric/eccentric phases at high/low speed respectively.

TMG protocol

The TMG was used to assess the mechanical properties of eight selected muscles. The assessments were performed following the protocol described by García-García et al.15 All measurements were performed isometrically in relaxed predefined positions: RF, VL and VM in a supine position with the knee angle set at 30° flexion (where 0° represents the fully extended joint); BF and ST in a prone position with the knee angle set at 5° flexion; GM and GL in a prone position with the ankle in a neutral position; and AL in a side-lying position, with the leg closest to the bed slightly adducted. Foam pads were used to support the joints. Measures of radial muscle belly displacement were acquired through a digital displacement transducer (GK 40, Panoptik d.o.o., Ljubljana, Slovenia) located perpendicular to the thickest part of the muscle belly. These measurements were visually determined through palpation during a voluntary contraction. The self-adhesive electrodes (5x5 cm, Cefar-Compex Medical AB Co., Ltd, Malmö, Sweden) were placed symmetrically at 2.5 cm away from the electrodes (Inter-electrode distance, 5 cm, IED), placing the positive electrode in the proximal area of the muscle above the measurement point and the negative electrode in the distal area below the measurement point, according to previous investigations¹⁴ and following the protocol suggested by Perotto et al.²² For all measurements, the point of maximum radial muscle belly displacement was determined by obtaining the time-displacement curve for each muscle, as well as on the basis of low intensity measurements (30 mA) previously obtained by placing the sensor on different points (2-3 mm apart within the area defined by the electrodes), until the maximum radial displacement point was identified. Electrical stimulation was applied using a TMG-S1 stimulator (EMF FURLAN & Co. d.o.o., Ljubljana, Slovenia) with 1 ms monophasic pulse duration and 30 mA initial current amplitude, which progressively augmented in 10 mA until reaching 100 mA (maximal stimulator output). Consecutive stimuli were separated by a 10 s resting period to avoid fatigue or post-tetanic activation.²³ A total of eight curves were recorded for each participant, but only the one with the highest maximum radial displacement was included in the analysis for each assessed muscle.14, 24

All the measurements involved the recording of the

RESISTANCE TRAINING PROGRAM IN FUTSAL PLAYERS

PIQUERAS-SANCHIZ

following parameters of involuntary isometric contraction produced by the electrical stimulus: Maximum radial muscle belly displacement (Dm) in mm.; Delay time (Td) as the time in milliseconds (ms) from onset to 10% of Dm and; contraction time (Tc) as the time in ms from 10% to 90% of Dm. Other authors²⁵ previously reported the need to normalize the increase in Tc, in order to compare the records obtained from different muscles. Therefore, we used the radial velocity displacement (VrD), in order to have a measure relatively independent of the Dm, calculated by V₉₀,²⁶ dividing the 90% of Dm by the sum of Tc and Td in mm·s-¹ *i.e.*, V₉₀ = VrD90= ((90**Dm*/100)) / ((Tc+Td)·1000)

In order to analyze the reliability of the relative and absolute TMG parameters, two measurements for each assessment were conducted. The electrode and sensor positions were marked with a dermatological pen on four randomly selected participants for each assessment. These selected participants were re-tested 20-45 min after the initial data collection.

Statistical analysis

Application of the Kolmogorov-Smirnov Test, together with the Lilliefors Test, showed that the sample distribution was normal, linear, and homocedastic. Relative and absolute reliability analyses were conducted using the data collected from within-day. Relative reliability of TMG parameters was assessed by calculating 2-way mixed effects model and the absolute agreement using the single measurement intra-class correlation coefficient (ICC).²⁷ In addition, absolute reliability was assessed by the stan-

dard error of measurement (SEM), which was expressed in relative terms through the coefficient of variation (CV). Preliminary analysis showed no significant differences (paired *t*-test) between dominant vs. non-dominant leg in any of the measured variables and thus, the analysis was based on the mean values for both legs. Repeated measures analysis of variance (RM ANOVA) was used to detect in-season changes in the mechanical and neuromuscular characteristics of professional futsal players. Two factors were included in the RM ANOVA model: Time as changes detected between 1-2 points and 1-3 points (session effect) and changes detected between 2-3 points (resistance training program effect) was used as the within-subject variable and the specific position (goalkeeper, defender, winger and pivot) was used as the between-subject variable. Bonferroni post-hoc tests with 95% adjustment for confidence intervals were used to compare the main effects and identify significant individual differences.

The effect sizes in RM ANOVA were reported as partial eta squared (η^2) and interpreted as small (0.01), moderate (0.06) or large (0.14).²⁸ The percentage differences in TMG parameters between the three assessments points were also calculated and interpreted. The significance level was set at 5% (P<0.05). Statistical analyses were performed using SPSS v.21 (SPSS Inc., Chicago, IL, USA).

Results

The values obtained for Test-retest absolute reliability measured by the CV were between 3.2-5.2%. The ICC values (95% CI) ranged between 0.91-0.98.

TABLE II.—Evolution of contractile properties throughout a season and after a resistance-training program in elite futsal players.

TMG Parameters	VM	VL	RF	BF	ST	AL	GL	GM
Tc 1 (ms)	24.1±2.6	21.1±3.5	28.1±3.8	32.0±12.3	37.9±5.1	20.6±4.1	44.8±12.1	26.7±6.2
Tc 2 (ms)	24.3±3.5	22.1±3.3	28.3±3.5	39.2±12.0	41.0±5.7 ^a	22.7±5.1ª	50.9±13.3	31.0±7.0
Tc 3 (ms)	24.3±3.5	21.0±3.9	28.4±2.9	37.6±10.2 ^a	42.3±7.4	24.4±4.7 ^a	47.1±13.4	32.7±6.5ª
Dm 1 (mm)	8.5±2.0	4.6±1.0	9.8±3.1	6.9±1.8	9.7±2.0	4.6±2.1	5.8±1.2	3.0±0.7
Dm 2 (mm)	8.5±1.7	4.8±1.2	9.7±2.4	8.1±2.3	10.6 ± 2.0	4.7±2.3	6.2±1.9	3.4±0.9
Dm 3 (mm)	9.1±2.2	4.8±1.6	9.8±1.6	7.4±1.4	10.3±1.7	5.7±1.8 ^a	6.5±1.7	3.9±1.0 ^a
Td 1 (ms)	24.0±1.6	23.7±1.8	27.0±1.8	24.3±2.3	26.2±2.3	24.2±2.1	25.5±2.5	24.8±2.6
Td 2 (ms)	24.0±2.0	24.3±2.0	26.9±1.5	26.8±2.6a	26.8±2.2	23.7±2.0	26.7±3.4	25.6±2.8
Td 3 (ms)	24.0±2.3	24.0±1.9	26.8±1.7	25.8±2.0	25.9±2.4	24.3±1.8	25.5±2.0	25.4±2.2
VrD90 1 (mm·s-1)	160.6±39.5	93.8±18.6	163.5±61.1	111.0±18.6	135.4±23.8	95.7±35.3	75.2±14.0	53.1±14.0
VrD90 2 (mm·s-1)	161.1±37.8	93.1±26.4	161.8 ± 48.1	111.2±22.5	142.0 ± 28.9	93.3±36.4	73.9±20.6	55.9±17.2
VrD90 3 (mm·s-1)	172.3±45.4	97.8±33.2	160.9±30.0	107.4±23.3	137.9±26.1	109.0±26.3	82.7±21.7	61.5±17.2

Data are expressed as mean of both legs±SD (N.=14).

VM: vastus medialis; VL: vastus lateralis; $\hat{R}F$: rectus femoris; BF: biceps femoris; ST: semitendinosus; AL: adductor longus; GL: gastrocnemius lateralis; GM: gastrocnemius medialis; Tc: contraction time; Dm: maximal radial displacement of the muscle belly; TD: delay time; VrD_{90} : radial displacement velocity at 90% Dm; Time-points 1, 2 and 3: First, second and third measurement. Tc, Td, and are in millisecons (ms), while Dm is in millimeters (mm) and VrD_{90} in millimeters/milliseconds (mm:s⁻¹). Values are mean of both legs and standard deviation. ^aSignificant differences in relation to baseline (Time 1) measurements (P<0.05). PIQUERAS-SANCHIZ

RESISTANCE TRAINING PROGRAM IN FUTSAL PLAYERS

The evolution of contractile properties is reported in Table II in terms of Tc, Dm, Td and VrD90. No significant changes were observed between 2-3 time points for any of the studied variables (resistance training effect). A large increment in Tc was observed for several muscles: BF (17.5% between 1-3 time points; P=0.02; η^2 =0.20), ST (8.1% between 1-2 time points; P=0.04; n²=0.28), AL (10.1% between 1-2 time points and 18.4% between 1-3 time points; P=0.008; η^2 =0.38) and GM (22.4% between 1-3 time points; P=0.009; η^2 =0.37). Larger increments were also observed in Dm for GM (30% between 1-3 time points; P=0.02; n²=0.30) and AL (22.4% between 1-3 time points; P=0.05; n²=0.25). Additionally, it was observed an increment in Td for BF (9.6% between 1-2 time points; P=0.002; η^2 =0.46), with a large size effect. No significant changes were observed in VrD₉₀ throughout the season, as well as non-significant increments for AL and GL between 2-3 time points (16.8%, P=0.13; n²=0.16 and 11.9%, P=0.055; n²=0.25, respectively). Finally, no significant differences were observed between player positions in any of the analyzed variables.

Discussion

To the best of our knowledge, this study is the first one analyzing the contractile properties in elite futsal players throughout the precompetition and competition season including a period of nine weeks resistance training program (one weekly training session). The aims of this study were: 1) to analyze the seasonal changes in the contractile properties produced in elite futsal players; 2) to determine the possible cumulative effect of a resistance training program on the contractile properties in those futsal players.

Our findings show that there is an increase in Tc, Dm and Td during the season but not uniformly among all the evaluated muscles. In fact, there is no change in the knee extensor muscles (RF, VL and VM) and therefore, our hypothesis is not fully confirmed. On the other hand, an in-season 9-week resistance training program (one day per week) had no effect on the contractile properties of futsal players. That is to say, no changes were found in any TMG parameters between measurement two, and measurement three. In addition, some of the methodological points regarding the present study should be addressed before further discussion. The absolute and relative intraday reliability values of our TMG assessment and estimation of contractile parameters show good reliability,²⁹ and they are in agreement with previous studies.^{10, 30}

Futsal is an intermittent sport where there are often

changes of pace and direction preceded by short sprints of high intensity^{31, 32} which significantly requests the stabilizing role of the evaluated muscles that have undergone changes (BF, ST, AL, GM). In addition, fatigue produces alterations in kinetic³¹ and kinematic parameters³³ which are directly related to the sprint technique. This situation could explain, in part, the changes found in Tc, Dm and Td in BF, ST, AL and GM. García-Manso *et al.*³⁴ and Rodríguez-Ruiz *et al.*³⁵ suggested that there is an adaptation of TMG parameters in the musculature involved in all technical actions of each analyzed sport modality.

There is a considerable increase of Tc in BF, ST, GM and AL during the season. However, knee extensors showed smalls variations. In line with this observation, it has been reported an increase in the Tc for the knee extensors during the season in professional cyclists¹⁵ and a decrease after a training program that mainly involved acceleration speed and agility tasks in professional footballers.¹⁴ In contrast, the knee flexors (BF) showed an increase in Tc and Td.14 Impairments in neuromuscular performance (sprint and change of direction) and in TMG properties throughout a season period have also observed in professional soccer players.⁶ The high volume of specific training performed by the athletes may be the main reason of these impairments in their contractile properties. These findings seem to indicate that the loads of training and competition cause changes in the contractile properties of the involved muscles.

The Tc values obtained from the professional futsal players during the season were slightly lower than those obtained from professional soccer players also taken during the season¹⁴ and lower (*i.e.* VL 21.1 *vs.* 26.1 ms) compared to those taken at the beginning of the preseason.²⁴ They are also logically lower than those obtained for professional cyclists both in preparation and in competition, with differences of almost 20 ms.¹⁵ All these data have been obtained by using exactly the same measurement protocol. These differences in the Tc of the muscles from professional, futsal and soccer players and cyclists are in line with the lower Tc found in power athletes compared to endurance athletes.³⁶

Our results revealed a significant increase of Dm in AL and GM throughout the season. By opposite, García-García *et al.*¹⁴ observed a decrease in the Dm values of VL, VM and RF after a training program in professional soccer players, mainly focused on acceleration speed and agility tasks during the season. However, in accordance with our findings, these authors also found an increment in Dm in BF muscle throughout the season.

RESISTANCE TRAINING PROGRAM IN FUTSAL PLAYERS

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Dm has been suggested to be an indirect measurement of muscle stiffness.^{37, 38} Notwithstanding, it remains to establish a clear interpretation of how the muscular stiffness evolves in connection with this parameter throughout the competition season, since it has been interpreted that a major Dm is associated with a decrease in muscle tone.³⁷ Indeed, the increase in Dm has been previously associated with a decrease in muscle diameter during muscle atrophy induced by bed rest,37 and moreover, it has been correlated with BF muscle thickness decrease after a 6-degree headdown tilt bed rest during 35 days.38 Therefore, it is reasonable to assume that athletes with lower Dm could achieve better physical performance in explosive tasks compared to their counterparts with higher Dm. The Dm values obtained by futsal players are similar to those obtained from professional footballers during the season¹⁴ and slightly lower than those obtained by professional soccer players at the beginning of the season.24

Our findings indicate that there is no alteration in the contractile properties of the evaluated muscles in professional futsal players after nine weeks of resistance training performed once a week. In addition, although no significant changes were observed in VrD90 throughout the season, an increment of this variable for AL and GL between 2-3 time points (16.8% and 11.9%, respectively) was observed. Conversely, a RT program (one session per week) for one muscle group during eight weeks has been shown to increase the 1RM from the baseline and the muscle thickness in well-trained men, although the effect is greater when the participants trained more than once per week.³⁹ Very similar results were obtained by Zaroni et al.⁴⁰ also in a training period of 8 weeks. A previous study with TMG²¹ found that 8-week plyometric training in three weekly sessions improved jumping performance, which was paralleled by a decrease in Tc and Dm. Similar findings were also reported by Zubac et al.,41 showing that an eight-week plyometric training (three sessions per week) increased the CMJ height together with an improvement in contractile properties of the leg muscles in seniors. Those findings suggest that there are muscle-specific adaptations to the stimuli induced by both, training and competition throughout the season. The small changes in the muscle properties induced by the resistance training accomplished in the present study could be explained by the low training frequency (one session per week). This fact, along with the intensity and the volume that were carried out, could be an insufficient stimulus to induce positive adaptations on the contractile properties of the muscles.

Moreover, the specific position does not seem to mod-

ulate neither the effect of the competition season or the effect of the resistance training program performed once a week, which contrasts with other findings where differences were observed in TMG between the specific positions of volleyball players³⁵ and professional soccer players in pre-season.²⁴ However, our results are in agreement with those found by Barbero-Alvarez *et al.*¹ and Caetano *et al.*² since they did not found any significant differences in the covered distance or between the work intensities performed by the different specific positions, highlighting the versatility of futsal players.

Limitations of the study

From a general point of view, this study had the following limitations: 1) performance evaluations such as maximal strength, jumping or sprints have not been carried out; 2) analysis of physiological parameters (levels of creatine kinase, ammonium, lactate, hemoglobin, etc.) were also not available; 3) finally, no psychophysiological evaluation, such as rate of perceived exertion was carried out. The analysis of the variables mentioned above would provide a more comprehensive evaluation. Although we recognize that this research is limited, our data open new perspectives for monitoring the muscle adaptations induced throughout the season in athletes with high frequency of competitions.

Conclusions

An increase in Tc, Td and Dm during the season was observed in futsal players, but not uniformly among all the evaluated muscles, since no changes were found for the knee extensors (RF, VL and VM). The change in the contractile properties of the evaluated muscles is not modulated by the specific position of the futsal player. In addition, an in-season 9-week resistance training program, once a week, had no significant effect on the contractile properties of futsal players based on the results. It seems not to be enough to have a differentiated effect probably due to insufficient volume, intensity or frequency. Thus, we propose to increase those training parameters for professional futsal players. The control of mechanical and contractile properties of the player's muscles could be useful to the head and strength and conditioning coaches to individualize the training loads and to control the effects of neuromuscular training throughout the season in professional futsal players, using a non-invasive technique that does not induce fatigue and, consequently, does not alter the rhythm of the workouts during the season.

PIOUERAS-SANCHIZ

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RESISTANCE TRAINING PROGRAM IN FUTSAL PLAYERS

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Vol. 60 - No. 7