

The anatomical, electrophysiological and histological observations of muscle contraction units in rabbits: a new perspective on nerve injury and regeneration

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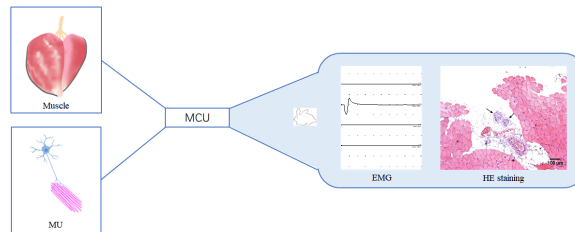
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Ting-Min Xu^{1,2,#}, Bo Chen^{2,3,4,#}, Zong-Xue Jin⁵, Xiao-Feng Yin^{1,2}, Pei-Xun Zhang^{1,2}, Bao-Guo Jiang^{1,2,3,4,*}

Graphical Abstract *Structural and functional integrity of the muscle contraction units*



Abstract

In the conventional view a muscle is composed of intermediate structures before its further division into microscopic muscle fibers. Our experiments in mice have confirmed this intermediate structure is composed of the lamella cluster formed by motor endplates, the innervating nerve branches and the corresponding muscle fibers, which can be viewed as an independent structural and functional unit. In this study, we verified the presence of these muscle construction units in rabbits. The results showed that the muscular branch of the femoral nerve sent out 4–6 nerve branches into the quadriceps and the tibial nerve sent out 4–7 nerve branches into the gastrocnemius. When each nerve branch of the femoral nerve was stimulated from the most lateral to the medial, the contraction of the lateral muscle, intermediate muscle and medial muscle of the quadriceps could be induced by electrically stimulating at least one nerve branch. When stimulating each nerve branch of the tibial nerve from the lateral to the medial, the muscle contraction of the lateral muscle 1, lateral muscle 2, lateral muscle 3 and medial muscle of the gastrocnemius could be induced by electrically stimulating at least one nerve branch. Electrical stimulation of each nerve branch resulted in different electromyographical waves recorded in different muscle subgroups. Hematoxylin-eosin staining showed most of the nerve branches around the neuromuscular junctions consisted of one individual neural tract, a few consisted of two or more neural tracts. The muscles of the lower limb in the rabbit can be subdivided into different muscle subgroups, each innervated by different nerve branches, thereby allowing much more complex muscle activities than traditionally stated. Together, the nerve branches and the innervated muscle subgroups can be viewed as an independent structural and functional unit. This study was approved by the Animal Ethics Committee of Peking University People's Hospital (approval No. 2019PHE027) on October 20, 2019.

Key Words: anatomy; electrophysiology; femoral nerve; hematoxylin-eosin staining; gastrocnemius; motor endplate; muscle contraction unit; peripheral nerve; quadriceps; rabbit; skeletal muscle; tibial nerve

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Introduction

By mediating voluntary muscle movement, neuromuscular junctions play a key role in physiology (Rudolf et al., 2019; Lazaridis and Tzartos, 2020; Barrantes, 2021). The skeletal muscles innervated by the peripheral nerves coordinate to produce various complex and fine movements. Traditionally, the motor unit was thought to be the basic unit of muscle activity (Buchthal and Schmalbruch, 1980; Heckman and Enoka, 2012; Farina et al., 2016; Mani et al., 2018; Piasecki et al., 2018) and that the motor endplates (MEPs), formed

by the terminal branches of motor nerves and the surface of muscle fibers, were uniformly distributed throughout the skeletal muscle (Jang et al., 2016; Liu et al., 2019; Rudolf et al., 2019; Chan et al., 2020; Chen et al., 2020). Excitation of a motor neuron generated electrical signals that induced muscle contraction through MEPs. The theory was that their summation resulted in a simple total contraction or relaxation of the muscle. Rather than that of a genetically fixed tissue specializing in producing tension, skeletal muscle is now considered a phenotypically plastic tissue that responds structurally and functionally to the nature of the

¹Department of Trauma and Orthopedics, Peking University People's Hospital, Beijing, China; ²Key Laboratory of Trauma and Neural Regeneration (Peking University), Ministry of Education, Beijing, China; ³Trauma Center, Peking University People's Hospital, Beijing, China; ⁴National Trauma Medical Center, Beijing, China; ⁵Department of Rehabilitation, Peking University People's Hospital, Beijing, China

*Correspondence to: Bao-Guo Jiang, MD, jiangbaoguo@vip.sina.com.

<https://orcid.org/0000-0001-8436-5266> (Bao-Guo Jiang)

#Both authors contributed equally to this work.

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demands placed on it (Lindstedt, 2016). The complexity of the real muscle activities require further macroscopic study of its nerve innervation pattern to guide clinical treatment (Rudolf et al., 2019). The muscles around a joint, such as the infraspinatus muscle (Fabrizio and Clemente, 2014), participate in the formation and regulation of various complex joint activities, and their origins and terminations are not distributed at a certain anatomical point. The traditional opinion that “the coordination between muscle groups” explains complex limb movements limits the exploration of other possibilities. Certain clinical problems cannot be fully explained by traditional opinions, for example, when only a part of a muscle is injured, the electromyography (EMG) recorded by traditional methods could appear normal.

It is possible that there is a structure that forms an intermediate link between the large-scale skeletal muscle and the microscale motor unit. In our previous studies, *in vivo* injection of fluorescent α -bungarotoxin was combined with the 3-Dimensional Imaging of Solvent-Cleared Organs protocol and ultramicroscopy to determine the 3D distributions of the MEPs in different skeletal muscles. Our results showed that the MEPs in these muscles were not evenly distributed, rather, they were distributed in lamellar clusters (Yin et al., 2019). Our electrophysiological experiments showed that contraction of different muscle subgroups of the gastrocnemius could be induced by electrically stimulating each nerve branch of the tibial nerve individually. Therefore, we proposed that the MEPs, the innervating nerve branches and corresponding muscle fibers can be regarded as an independent structural and functional unit, which we termed muscle contraction unit (MCU). Using a denervated and nerve-repairing mice model, we found that after denervation, the originally lamellar-cluster distributed MEPs in the gastrocnemius gradually disintegrated and diffused. After subsequent nerve-repairing, the diffused MEPs gradually recovered to the original lamellar-cluster form (Yin et al., 2019).

In this study, the anatomy of the nerve branches around the neuromuscular junctions in the muscles of the lower limb in rabbits was investigated. Electrophysiological experiments and hematoxylin-eosin (HE) staining were performed to assess the MCU in rabbits.

Materials and Methods

Animals

Five adult female New Zealand rabbits (about 2.5-kg weight, 10 weeks old) were all purchased from Beijing Vital River Laboratory Animal Technology Co., Ltd. (animal production license No. SCXK (Jing) 2016-0006 and animal usage license No. SYXK (Jing) 2017-0022) and kept in the animal rooms of Peking University People’s Hospital (22–26°C at room temperature, 12/12-hour in light/dark cycle and 60–65% relative humidity), food and water were available *ad libitum*. The animal experiment design was approved by the Animal Ethics Committee of Peking University People’s Hospital (approval No. 2019PHE027) on October 20, 2019.

Anatomy

The rabbits were anesthetized by isoflurane (RWD Life science Co., Ltd., Shenzhen, Guangdong Province, China) inhalation. After anesthetization, the left lower limb was shaved and sterilized. Then, the left quadriceps, gastrocnemius and their innervating nerves (the muscular branch of the femoral nerve and tibial nerve) were exposed carefully. The number of nerve branches around the neuromuscular junction into the muscle was recorded.

EMG

Paired recording electrodes (Xi’an Friendship Medical Electronics Co., Ltd., Xi’an, Shaanxi Province, China) were placed in the distal and proximal part of each muscle

subgroup, with the reference electrode placed in the gluteus maximus. Parameters (Medlec Synergy, Oxford Instruments, Surrey, UK) were set as 0.09–0.12 mA in current strength to elicit an AP, five pulses per second in frequency, 0.1 ms in duration (Dario et al., 2001; Ujiie et al., 2017) We tried a range of stimuli strengths and chose the same strength to stimulate each nerve branch and recorded the action potential elicited in the muscle subgroups.

HE staining

The rabbits were euthanized by excessive CO₂ inhalation. The tissues (about 1 cm × 1 cm × 0.5 cm in size) around each neuromuscular junction into the muscle were dissected and placed in 4% paraformaldehyde for 12 hours. Then the fixed tissues were rinsed with phosphate buffered saline twice, passed through gradient alcohols for dehydration and xylene for transparency. The tissue was then paraffin-embedded before being cut into 5- μ m-thick sections (Fischer et al., 2008). After HE staining, the sections were embedded with neutral resin and then observed under an optical microscope (Leica, Wetzlar, Germany).

Results

Anatomical characteristics of femoral nerve and tibial nerve in rabbit

The regional anatomy showed that the muscular branch of the femoral nerve sent out 4–6 nerve branches innervating the quadriceps; the tibial nerve sent out 4–7 nerve branches innervating the gastrocnemius (**Figure 1A** and **B**). The number of nerve branches of the muscular branch of the femoral nerve and the tibial nerve was shown in **Table 1**.

Table 1 | The number of nerve branches of the muscular branch of the femoral nerve

Nerve	Number of nerve branches	Number of animals
The muscular branch of the femoral nerve	4	1
	5	2
	6	2
The tibial nerve	4	1
	5	2
	7	2

Electrophysiological outcomes when stimulating each nerve branch of femoral nerve and tibial nerve

Paired recording electrodes were placed sequentially from the lateral to the medial. The recording electrodes in the quadriceps and gastrocnemius were placed as is shown in **Figure 1A** and **B**. When stimulating each nerve branch respectively, relative independent muscle subgroup contraction was induced and different EMGs were observed in each muscle subgroup.

Muscle contraction

When each branch of the main muscle branch of the femoral nerve was electrically stimulated from the lateral to the medial respectively, it was found that contraction of the lateral muscle (LM), intermediate muscle and medial muscle (MM) of the quadriceps could be induced. For the femoral nerve, which consisted of six nerve branches, electrical stimulation of individual nerve branches 2–5 from the lateral to the medial, induced contractions of LM, intermediate muscle, and MM. However, when electrically stimulating the nerve branch 1 and nerve branch 6, no contraction of the muscle subgroup was observed (**Table 2**). For the tibial nerve, which consisted of four nerve branches, when lateral nerve branches 1–3 and the medial nerve branch were electrically stimulated from the lateral to the medial respectively, the individual contractions of LM1–3, and MM of the gastrocnemius were observed (**Table 3**).

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Table 2 | Contractions of the subgroups of the quadriceps when electrically stimulating each branch of the muscle branch of the femoral nerve individually

	Nerve trunk	NB1	NB2	NB3	NB4	NB5	NB6
LM	+		+				
IM	+			+	+		
MM	+					+	

All separated branches of the muscle branch of the femoral nerve were named, for example NB1, from the lateral to the medial, respectively. IM: Intermediate muscle of quadriceps; LM: lateral muscle of quadriceps; MM: medial muscle of quadriceps; NB: nerve branch. + represents the contraction of the corresponding muscle subgroup when the femoral nerve trunk or each nerve branch is stimulated.

Table 3 | Contractions of the subgroups of the gastrocnemius when electrically stimulating each branch of the tibial nerve

	Nerve trunk	LNB1	LNB2	LNB3	MNB
LM1	+	+			
LM2	+		+		
LM3	+			+	
MM	+				+

The branches of the tibial nerve were named, for example, LNB1–3 from the lateral to the medial, respectively, whereas the most medial branch was named the MNB. LM1: Lateral muscle 1 of the gastrocnemius (lateral muscle1); LM2: lateral muscle 2 of the gastrocnemius (lateral muscle2); LM3: lateral muscle 3 of the gastrocnemius (lateral muscle3); LNB: lateral nerve branch; MM: medial muscle of the gastrocnemius (medial muscle); MNB: medial nerve branch. + Represents the contraction of the corresponding muscle subgroup when the tibial nerve trunk or each nerve branch is stimulated.

EMG

The EMG waves recorded in different muscle subgroups were different when each nerve branch was electrically stimulated (**Figure 2A–F**). For the femoral nerve, which consisted of six nerve branches, EMGs were recorded in the quadriceps when each of nerve branches 2–5 were electrically stimulated individually but EMGs were not recorded when either nerve branch 1 (**Figure 2A**) or nerve branch 6 (**Figure 2F**) was stimulated. When electrically stimulating nerve branch 2, EMGs can be recorded in channels 1–5, with the major waves in channel 1 and 3 upward and the major waves in channel 2, 4, and 5 downward. The waves with the largest amplitude occurred in channels 3 and 5 (**Figure 2B**). When stimulating nerve branch 3, EMGs were recorded in channels 1–5, with the major waves in channels 1, 2, 4 and 5 upward and the major waves in channel 3 downward. The waves with the largest amplitude were in channel 5 (**Figure 2C**). When stimulating nerve branch 4, EMGs were recorded in channels 1–5, with the major waves in channels 2, 4, and 5 downward, and the major waves in channels 1 and 3 upward. The waves with the largest amplitude were obtained in channels 3 and 5 (**Figure 2D**). When stimulating nerve branch 5, EMGs can be recorded in channels 1–5, with the major waves in channels 2, 4 and 5 downward and the major waves in channels 1 and 3 upward. The wave with the largest amplitude was in channel 3 (**Figure 2E**). The tibial nerve consists of four nerve branches. EMGs could be recorded in the gastrocnemius when any one of the lateral nerve branches 1–3, or the medial nerve branch was electrically stimulated. When stimulating lateral nerve branch 1, EMGs can be recorded in channel 1 and channel 3, with the major wave in channel 1 downward, and the major wave in channel 3 upward. The wave with the largest amplitude was associated with channel 1 (**Figure 3A**). When stimulating lateral nerve branch 2, EMGs can be recorded in channel 2, with the major wave downward (**Figure 3B**). When stimulating lateral nerve branch 3, EMGs can be recorded in channels 3 and 4, both major waves. The largest amplitude wave was seen in channel 3 (**Figure 3C**). When stimulating the medial nerve branch, EMGs were

recorded in channel 4, as a major downward wave (**Figure 3D**).

Tissue morphology of the nerve branches around the neuromuscular junction into the muscle

HE staining of the tissues showed that around the nerve's entrance into the muscle most of the nerve branches consisted of individual neural tracts (**Figure 4A**), but a few nerve branches consisted of more than two neural tracts after the nerves entrance into the muscle (**Figure 4B**).

Discussion

Traditionally, the movement of a limb resulted from the coordinated activities of multiple muscle groups under the fine control of the nervous system. The functional activity of a single muscle was considered to be simple, i.e., either the whole muscle contracted or the whole muscle relaxed. The traditional model has significant limitations in explaining the motion of a joint. Corresponding to the complex joint's movements, the muscles around the joints are usually cross-joint distributed. Some muscles (e.g., supraspinatus and infraspinatus) were distributed from multiple anatomical points rather than an individual point (Fabrizio and Clemente, 2014; Rudolf et al., 2019). Accordingly, we speculate that these muscles are likely to perform much more complex activities than simple whole-block contractions or relaxations.

Previous anatomical studies and EMG studies have indicated that even a single belly muscle has several compartments (MacFadden and Brown, 2010; Wickham and Brown, 2012; Méndez et al., 2013; Larsen et al., 2014; Watanabe et al., 2016). Our earlier studies also revealed the presence of fascia-like tissues within the gastrocnemius, which suggested that there might be an intermediate structure in the muscle (i.e., muscle subgroups) before its division into microscopic muscle fibers. Our later experimental study in mice confirmed this intermediate structure (i.e. the lamellar cluster of MEPs, the innervating nerve branches and the corresponding muscle fibers) that can be considered an independent structural and functional unit (Yin et al., 2019).

In the present study, we performed further observations in rabbits. Local anatomy indicated that peripheral nerves sent out several nerve branches around its entrance into the muscle. Electrophysiological testing of nerve function has been increasingly used in the field of peripheral nerve injury and regeneration (Aminoff, 2004; Uzun et al., 2006; Karşıdağ et al., 2008; Patel et al., 2018; Wang et al., 2019; Taylan and Barishaner, 2020). Our electrophysiological findings revealed that when electrically stimulating each nerve branch individually, relatively independent muscle subgroup contraction was induced and different EMGs was observed in each muscle subgroup. From the histological perspective, HE staining indicated that most of the nerve branches around the neuromuscular junction consisted of one individual neural tract. However, a small number of nerve branches consisted of two neural tracts, which indicated that these nerve branches would develop new fine branches after entering the muscle. This suggested that the lower limb muscles of the rabbit can be further divided into muscle subgroups, which are innervated by different dominant nerve branches and consequently participate in the formation of more complex muscle activities. We concluded that, in rabbits, the nerve branches and their innervated muscle subgroups could be viewed as an independent structural and functional unit, an MCU. The MCUs link the muscles and the motor units to better interpret various complex and elaborate muscle activities.

This study has some limitations. The present study focused on anatomical and functional descriptions without statistical analysis. The sample size of this study was small, making this a preliminary, exploratory study. Additional techniques could be applied that further investigate the structural basis of the

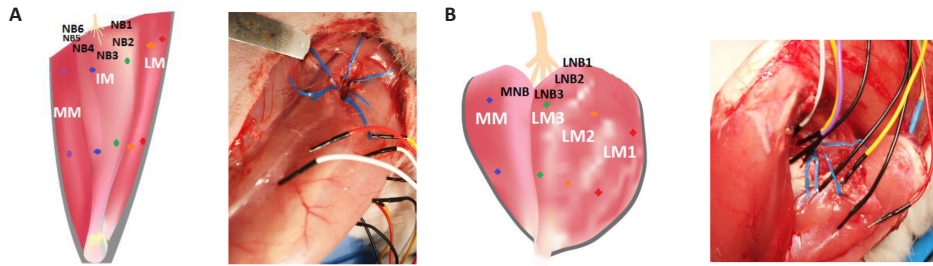


Figure 1 | The placement of paired recording electrodes in the femoral nerve (A) and tibial nerve (B) for electrophysiological study in rabbits.

The muscle branches of the femoral nerve were named, for example, NB1–4 from the lateral to the medial, respectively. The branches of the tibial nerve were named LNB1–3, from the lateral to the medial, whereas the most medial branch was named MNB. IM: Intermediate muscle of quadriceps; LM: lateral muscle of quadriceps; LNB: lateral nerve branch; MM: medial muscle of gastrocnemius; MNB: medial nerve branch; NB: nerve branch. Red dots represent the placement of the paired recording electrodes in channel 1. Yellow dots represent the placement of the paired recording electrodes in channel 2. Green dots represent the placement of the paired recording electrodes in channel 3. Blue dots represent the placement of the paired recording electrodes in channel 4. Purple dots represent the placement of the paired recording electrodes in channel 5.

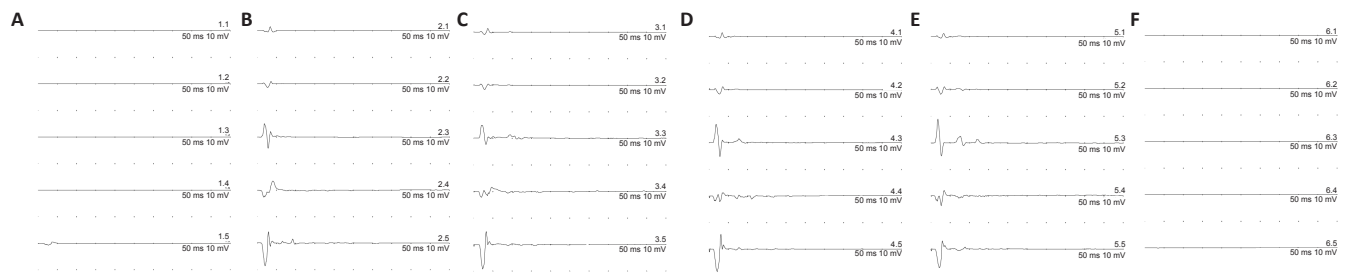


Figure 2 | Electrophysiology when electrically stimulating each branch of muscle branch of the femoral nerve in rabbits.

(A–F) Electrically stimulating lateral nerve branch 1 (A), nerve branch 2 (B), nerve branch 3 (C), nerve branch 4 (D), nerve branch 5 (E) and nerve branch 6 (F), respectively from the lateral to the medial. Electrophysiology parameters: Vertical calibration, 10 mV; horizontal calibration, 50 ms. Each trace was generated by the same strength of electrical stimulus.

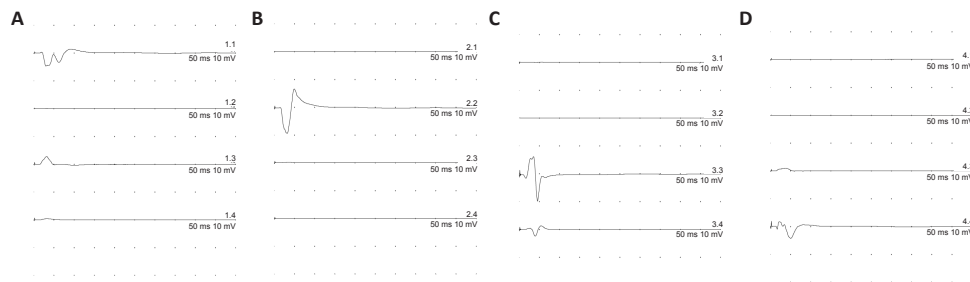


Figure 3 | Electrophysiology when electrically stimulating each branch of the tibial nerve in rabbits.

(A–D) Electrically stimulating lateral nerve branch 1 (A), lateral nerve branch 2 (B), lateral nerve branch 3 (C), and medial nerve branch (D). Electrophysiology parameters: Vertical calibration, 10 mV; horizontal calibration, 50 ms. Each trace was generated by the same strength of electrical stimulus.

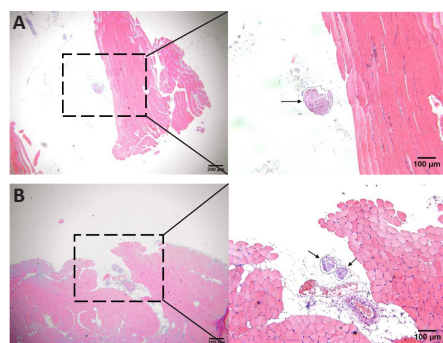


Figure 4 | Typical micrograph of the hematoxylin and eosin staining of the tissues around the neuromuscular junction in the lower limb in rabbits.

(A) Nerve branch around the neural entrance into the muscle consisting of an individual neural tract (MNB to MM). (B) Nerve branch around the neural entrance into the muscle consisting of two neural tracts (NB2 to LM). LM: lateral muscle of quadriceps; MM: medial muscle of gastrocnemius; MNB: medial nerve branch; NB2: nerve branch 2. Arrows represent neural tract. Scale bars: 200 μ m in original images, and 100 μ m in enlarged images.

MCUs in rabbits, such as the three-dimensional distributions of the innervating nerve branches and the MEPs in the muscles. Different techniques may be more amenable for larger sampling to give statistical validity. The identification

and characterization MCUs in other muscles, such as the upper limb muscles, would support a general theory. We speculate that the number of MCUs and MEPs lamella clusters in the upper limb muscles may be less than in the lower limb muscles owing to their more elaborate structural and functional differentiations. A small number of nerve branches consisted of two neural tracts, therefore, the number and boundary of MCUs in each muscle require further animal experiment studies.

The results of this experimental study will be instructive in clinical research. In particular, recording electrodes should be placed based on distributions of the MCUs in EMG, which could indicate neuromuscular pathway disorders in a more precise way. Generally, our results may provide far-reaching implications from a clinical perspective, including patients who need muscle transplantation owing to trauma or other factors (Chuang, 2008; Barrera-Ochoa et al., 2017; Yi Lee et al., 2019). It would be important for clinicians to identify and use intact MCUs when performing nerve–muscle transplantation to preserve the structural and functional integrity of the MCUs.

In conclusion, the muscles of the lower limb in the rabbit can be subdivided into different muscle subgroups, each innervated by different nerve branches, thereby allowing much more complex muscle activities than traditionally stated. Together, the nerve branches and the innervated muscle

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subgroups can be viewed as an independent structural and functional unit.

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Conflicts of interest: The authors declare no competing interests.

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References

- Aminoff MJ (2004) Electrophysiologic testing for the diagnosis of peripheral nerve injuries. *Anesthesiology* 100:1298-1303.
- Barrantes FJ (2021) Possible implications of dysregulated nicotinic acetylcholine receptor diffusion and nanocluster formation in myasthenia gravis. *Neural Regen Res* 16:242-246.
- Barrera-Ochoa S, Collado-Delfa JM, Sallent A, Lluch A, Velez R (2017) Free neurovascular latissimus dorsi muscle transplantation for reconstruction of hip abductors. *Plast Reconstr Surg Glob Open* 5:e1498.
- Buchthal F, Schmalbruch H (1980) Motor unit of mammalian muscle. *Physiol Rev* 60:90-142.
- Chan JP, Clune J, Shah SB, Ward SR, Kocsis JD, Mozaffar T, Steward O, Gupta R (2020) Examination of the human motor endplate after brachial plexus injury with two-photon microscopy. *Muscle Nerve* 61:390-395.
- Chen XY, Hao F, Gao YD, Zhao W, Duan HM, Yang CY, Li XG (2021) Rehabilitation training combined with neurotrophin 3-chitosan scaffolds enhanced skeletal muscle morphology and functional recovery in rats with spinal cord injury. *Zhongguo Zuzhi Gongcheng Yanjiu* 25:2514-2520.
- Chuang DC (2008) Nerve transfer with functioning free muscle transplantation. *Hand Clin* 24:377-388, vi.
- Dario P, Carrozza MC, Micera S, Massa B, Zecca M (2001) Design and experiments on a novel biomechatronic hand. In: *Experimental robotics VII* (Rus D, Singh S, eds), pp 159-168. Berlin, Heidelberg: Springer Berlin Heidelberg.
- Fabrizio PA, Clemente FR (2014) Anatomical structure and nerve branching pattern of the human infraspinatus muscle. *J Bodyw Mov Ther* 18:228-232.
- Farina D, Negro F, Muceli S, Enoka RM (2016) Principles of motor unit physiology evolve with advances in technology. *Physiology (Bethesda)* 31:83-94.
- Fischer AH, Jacobson KA, Rose J, Zeller R (2008) Hematoxylin and eosin staining of tissue and cell sections. *CSH Protoc* 2008:pdb.prot4986.
- Heckman CJ, Enoka RM (2012) Motor unit. *Compr Physiol* 2:2629-2682.
- Jang HS, Cho KH, Murakami G, Cho BH (2016) Topographical relationships of intramuscular nerves and vessels of the motor endplates in the thigh and gluteal regions of human fetuses: an immunohistochemical study. *Surg Radiol Anat* 38:587-596.
- Karşıdağ S, Özcan A, Sahin S, Karşıdağ S, Kabukçuoğlu F, Uğurlu K, Baş L (2008) Electrophysiologic and histopathologic evaluation of peripheral nerve regeneration at different nerve segments and with different repair techniques. *Acta Orthop Traumatol Turc* 42:278-283.
- Larsen CM, Juul-Kristensen B, Olsen HB, Holtermann A, Søgaard K (2014) Selective activation of intra-muscular compartments within the trapezius muscle in subjects with subacromial impingement syndrome. A case-control study. *J Electromyogr Kinesiol* 24:58-64.
- Lazaridis K, Tzartos SJ (2020) Myasthenia gravis: autoantibody specificities and their role in MG management. *Front Neurol* 11:596981.
- Lindstedt SL (2016) Skeletal muscle tissue in movement and health: positives and negatives. *J Exp Biol* 219:183-188.
- Liu QG, Huang QM, Liu L, Nguyen TT (2019) Structural and functional abnormalities of motor endplates in rat skeletal model of myofascial trigger spots. *Neurosci Lett* 711:134417.
- MacFadden LN, Brown NA (2010) The influence of modeling separate neuromuscular compartments on the force and moment generating capacities of muscles of the feline hindlimb. *J Biomech Eng* 132:081003.
- Mani D, Almklass AM, Hamilton LD, Vieira TM, Botter A, Enoka RM (2018) Motor unit activity, force steadiness, and perceived fatigability are correlated with mobility in older adults. *J Neurophysiol* 120:1988-1997.
- Méndez GA, Gatica VF, Guzmán EE, Soto AE (2013) Evaluation of the neuromuscular compartments in the peroneus longus muscle through electrical stimulation and accelerometry. *Braz J Phys Ther* 17:427-434.
- Patel NP, Lyon KA, Huang JH (2018) An update-tissue engineered nerve grafts for the repair of peripheral nerve injuries. *Neural Regen Res* 13:764-774.
- Piasecki M, Ireland A, Piasecki J, Stashuk DW, Swiecicka A, Rutter MK, Jones DA, McPhee JS (2018) Failure to expand the motor unit size to compensate for declining motor unit numbers distinguishes sarcopenic from non-sarcopenic older men. *J Physiol* 596:1627-1637.
- Rudolf R, Khan MM, Witzemann V (2019) Motor endplate-anatomical, functional, and molecular concepts in the historical perspective. *Cells* 8:387.
- Taylan SB, Bariskaner H (2020) Effects of dexmedetomidine and dexketoprofen on the conduction block of rat sciatic nerve. *Neural Regen Res* 15:929-935.
- Ujiie N, Nakano T, Shimizu K (2017) Measurement of the action potential of the nerve for the isolation of the nerve in the operative field: An experimental study in rabbit. *Trans Jpn Soc Med Biol Eng* 55Annual:524-525.
- Uzun N, Tanriverdi T, Savrun FK, Kiziltan ME, Sahin R, Hanimoglu H, Hanci M (2006) Traumatic peripheral nerve injuries: demographic and electrophysiologic findings of 802 patients from a developing country. *J Clin Neuromuscul Dis* 7:97-103.
- Wang ZY, Qin LH, Zhang WG, Zhang PX, Jiang BG (2019) Qian-Zheng-San promotes regeneration after sciatic nerve crush injury in rats. *Neural Regen Res* 14:683-691.
- Watanabe K, Otsuki S, Hisa T, Nagaoka M (2016) Functional difference between the proximal and distal compartments of the semitendinosus muscle. *J Phys Ther Sci* 28:1511-1517.
- Wickham JB, Brown JM (2012) The function of neuromuscular compartments in human shoulder muscles. *J Neurophysiol* 107:336-345.
- Yi Lee TM, Sechachalam S, Satkunanatham M (2019) Systematic review on outcome of free functioning muscle transfers for elbow flexion in brachial plexus injuries. *J Hand Surg Eur Vol* 44:620-627.
- Yin X, Yu T, Chen B, Xu J, Chen W, Qi Y, Zhang P, Li Y, Kou Y, Ma Y, Han N, Wan P, Luo Q, Zhu D, Jiang B (2019) Spatial distribution of motor endplates and its adaptive change in skeletal muscle. *Theranostics* 9:734-746.

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