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Nerve supply of extra ocular muscles

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ABSTRACT

Background: The extra ocular muscles are supplied by oculomotor, trochlear and abdicants nerves. *Aim of our work:* Is to study the pattern of motor innervations of the extra ocular muscles in goat. *Material and Methods:* This study was carried on nerves supplying the oblique and rectus muscles extra ocular muscles of goat. 13 orbits were obtained from 4 goat's right after killing. The distribution of fiber sizes and the sensory pathway in the stem of oculomotor nerves, oculomotor, trochlear and abdicants were investigated. The technique of silver block impregnation was employed to research muscle innervations. Toludine blue was used to block and stain semi thin slices of the nerves Oculomotor, Trochlear and Abdicant as well as muscle specimens. *Results:* The nerve supply of extra ocular muscle is relatively rich. The global zone of the muscles was supplied by thin nerve fibers. Different types of pro prospective organs found in extra ocular muscles. *Conclusion:* The intricate design and operation of the extra ocular muscles control eye movements.

Keywords: Nerve Supply, Extra ocular Muscles, oculomotor, trochlear, abducent.

1. INTRODUCTION

The four extra ocular rectus muscles arise from the common tend nous ring. They extend forward to attach to the eye ball by tend nous expansions that attach to sclera (Turvey & Golden et al., 2012). A large anatomical approach called Silver's stain enables the detailed course and pattern of nerves to be traced (Mu & Sanders et al., 2010; Won et al., 2011). According to animal tissues (Gözil et al., 2002) and (Gülekon et al., 2002) hypo the sized that Silver's stain approach may be helpful for in depth research on the motor control of extra ocular muscles and may enhance our comprehension of the anatomy and function of those muscles. A thorough understanding of the distribution patterns of the nerves and any potential deviations in their course may also improve the security and effectiveness of surgical procedures carried out inside the orbit and on the extra ocular muscles themselves (Shin et al., 2018; Shin et al., 2019) as maintaining the innervations preserves the muscle's features and functions. (Bohlen et al., 2019) looked into the extra ocular muscles structure and their motor nuclei in the mouse. The primary neuroanatomical organization of the motoneurons that innervate the muscles around the eye was also discovered by Bohlen along with the fundamental organization of the orbital musculature. Both eye movements and variations in orbital organization between rodents and primates must be taken into account when analyzing the neuroanatomical organization of the extra ocular motor nuclei in relation to function. Primates have eyes with foveae that are situated in the front. They have wide binocular fields and excellent vision thanks to their specializations, respectively. Investigating if connections across motoneuron pools like this one have functional correlations will be fascinating.

Now new anatomical data on the innervations pattern of the eye muscles have been developed. The intramuscular distribution of the soma to motor nerve fibers is particularly highlighted in the most recent anatomical works on extra ocular innervations, which is significant for understanding the complicated function of those muscles (Haładaj et al., 2019). Extra ocular muscle innervations appear to have several unique characteristics, like a high ratio of nerve branches compared to other skeletal muscles (Abdelhady et al., 2021).

2. MATERIALS AND METHODS

Our study adhered to the PSA University Ethical Committee approved Animal Research Guideline for the Use and Care of Animals in Research, Al-Kharj (PSAU-2022 ANT 18 /43PI). It is a research project that was experimental and ran from December 2021 to September 2022. This research focused on the goat's rectus and oblique extra ocular muscles' nerves.4 goats were killed and immediately after 13 orbits were collected. The eye balls and orbital muscles were excised as a single mass once the orbits were made visible. The entire collection of specimens was fixed for 10 days in 10% formalin. The four rectus, two obliques and supplying nerves were quickly located and separated for further analysis. Each muscle was cleaned of fat and connective tissue and separated into three portions, the proximal, middle and distal thirds for the Ranson block technique of silver impregnation. The latter was removed using the tendon's distal end. The muscle fragments were split into longitudinal and transverse serial slices each 7-u thick, treated with Ranson silver and blocked in paraffin. A few muscle fragments were cut up, fixed in gluteraldehyde and osmic acid, and then blocked in epon for Toludine blue-stained semi thin slices.

From the fresh specimens, the stems of the ten orbits' cranial nerves 3, 4, and 6 were removed, fixed in gluteraldehyde and osmic acid, blocked for semi thin slices, and stained with toludine blue. We fixed the muscle fragments in distilled water and 100% alcohol with 1% ammonia for 48 hours as part of a modified silver block impregnation process. Then, during the following twenty four hours, we transferred it to pyridine. Then, we cleaned it several times in distilled water until the pyridine odor was eliminated (at least twenty four hours). The muscle fragments were placed in 2% AgNo3 for one to three days at 35 degrees in the dark and then washed with distilled water. Then it was cleaned, dehydrated, clarified, and paraffin embedded. Slash at 10u. Remove wax and mount in plastic resin (Femi-Akinlosotu et al., 2019). The semi thin sections of the Nerves supplying extra ocular muscles were stained with Toludine blue to show the myelin sheathes of their component fibers. These sections were photographed at magnification of 400 and then those photos were further magnified and half times. The total magnification 1000 times makes the myelin ring of 2 u diameter equals 2mm using a Perspex quaticule (ruler) with rings from 2mm to 30mm in diameter, the 1000 magnified myelin rings were measured and counted in groups at 2mm intervals. The number of fibers of each group was counted and the total numbers of fibers in each nerve were estimated.

The percentage of each group from the total number of fibers in each nerve was calculated and put in tables. The histograms were constructed to show the percentage of each up and their fiber diameters the fiber size distribution. Histogram of a nerve supplying a muscle with 2 peaks (Bimodal): One at 6-8u in diameter and the other at 10-12u in diameter is a strong evidence of pro prospective innervations of this muscle. 6-8u cover alpha motor efferent’s, 10-12u or more cover the size of afferent fibers from pro prospective endings (i.e., muscle spindles and tendon organs of Golgi).

3. RESULTS

In silver impregnated preparations, the two oblique extra ocular muscles and each of the four rectus muscles were divided into two zones. The muscle fibers in the orbital zone extend toward the bony orbit; nearly all of the muscle fibers on the orbital surface are stained black by Ranson silver and vary in thickness. The global zone is the second zone. It includes the fibers that extend from the muscle's surface toward the globe (Figure 1 A, B). The fibers used in the manufacture of silver were categorized according to their stain from light to dark red. They exhibit various thicknesses as well each muscle's nerve supply is comparatively abundant. The muscle fibers (dark fibers) in the orbital zone were supplied by thick nerve fibers (4-8u) (Figure 1 C, D). All types of muscle fibers (pale, intermediate and red) were supplied by thin nerve fibers in the global zone (2-4u). At the neuromuscular junction, diffuse motor terminals of the en-grappe type that were quite long and rich in arborizations were provided by thick nerve fibers in the orbital fibers (Figure 2A). A more localized motor ending of the en-plate (en-plaque) type with essentially minimal terminal arborization at the neuromuscular junction was supplied by the thin nerve fiber in the global fibers (Figure 2B). The central section of the muscle contained the wide motor band. We found many forms of pro prospective endings in extra ocular muscles of goat. Typical muscle spindles frequently found especially in the middle third of the muscles (Figure 3 A, B).

The structure and innervations simulate typical spindles in skeletal muscles as described by previous studies. Additionally Golgi tendon organs were numerous in the distal tendons of extra ocular muscles. Each has a capsule of collagen fibers, supplied by large mylinated nerve fiber from its distal aspect. The capsule is frequently invaded by few extrafusal muscle fibers (1-5 fibers). These tendon organs were not found in human. Palisade endings (innervated myotendinous cylinders) were frequent in the goat and found at the distal myotendinous junction. Each has a thin capsule, innervated by relatively thin nerve fiber and it usually receives the insertion of a single muscle fiber. These palisade endings are unique to extra ocular muscles especially in man. The localization of these endings at the distal myotendinous line makes them susceptible to damage during surgery for strabismus which affects the result of the operation. Finally, few spiral endings were found in the middle third of extra ocular muscles. They wrapped around an extrafusal muscle fiber and were innervated by thick nerve fiber. Mylinated fibers with a diameter ranging from 2 u to 22 u were found in the segments extracted from the trunks of the nerve's oculomotor, trochlear and abdicant near to the brain stem. This region extends from the intrafusal muscle fibers to the y- efferent motor fibers (2-4u). The alpha motor efferent fibers to extrafusal fibers of extra ocular muscles (4–8u), the group II afferents from the muscle spindles (8–12u) and the group I afferents from the muscle spindles and tendon organs (12-22u) (Figure 4, 5). The histograms of the fiber size distribution of the oculomotor and abdicant nerves were bimodal with the peaks at (6-8u) and (1012u). More than 15% of the fibers are 2-6u, because the global fibers are supplied mainly by relatively thin fibers of this range, the orbital fibers supplied by thicker nerve fibers (Tables 1, 2) (Figure 6, 7).

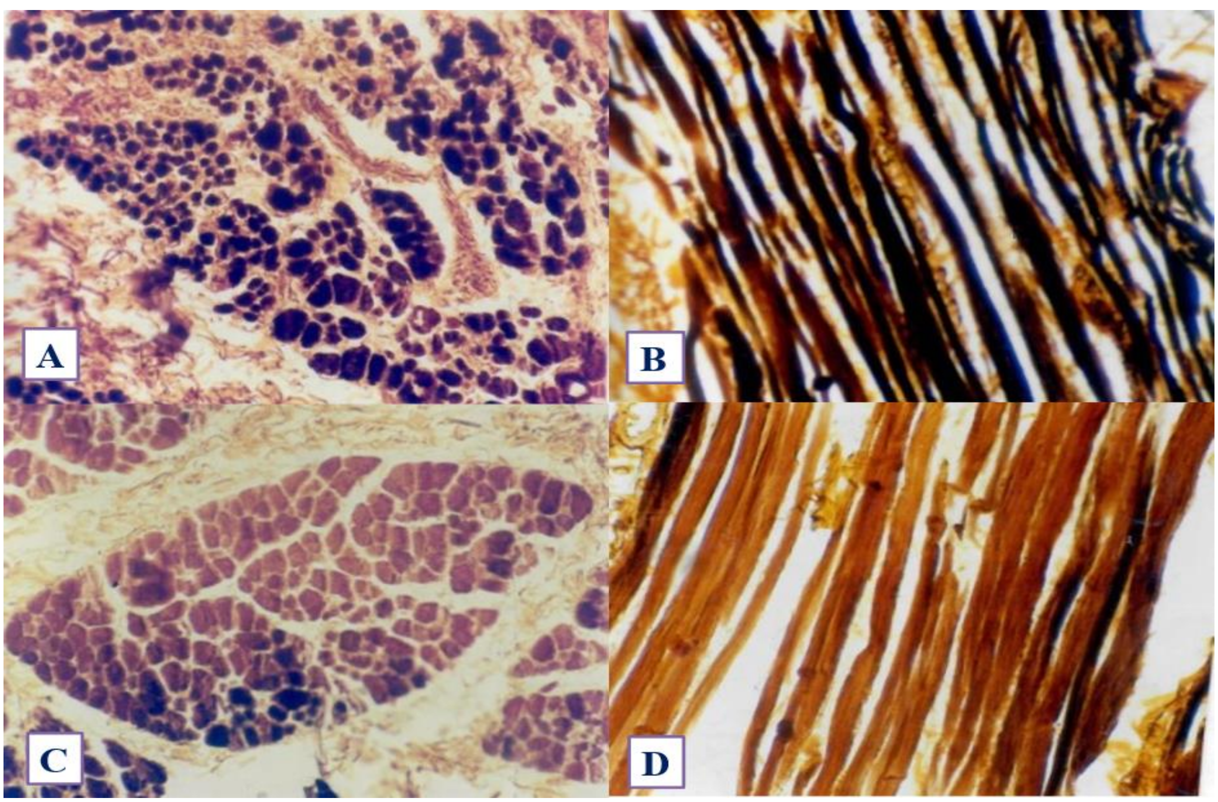

Figure 1 (A, B) Transverse and longitudinal sections of the superior rectus muscle: Orbital zone the fibers are stained black and of different thickness (C, D) Transverse and longitudinal sections of the lateral rectus muscle global zone. The fibers showing grading of color from pale, intermediate to red with difference in thickness (Ranson silver x 100).

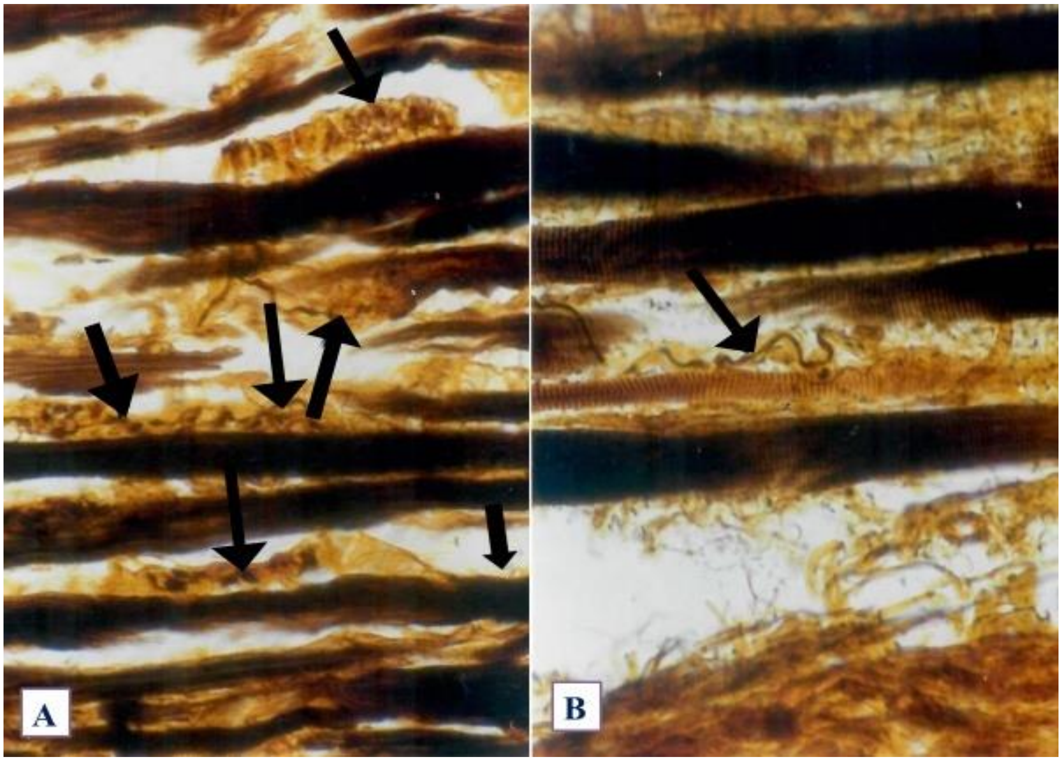

Figure 2 A Longitudinal section showing orbital type 2 muscle fibers with many motor endings. B Global red muscular fiber type 3 in longitudinal section, with a single en plate motor terminal supplied by a thin nerve fiber (→) (Ranson silver x 400).

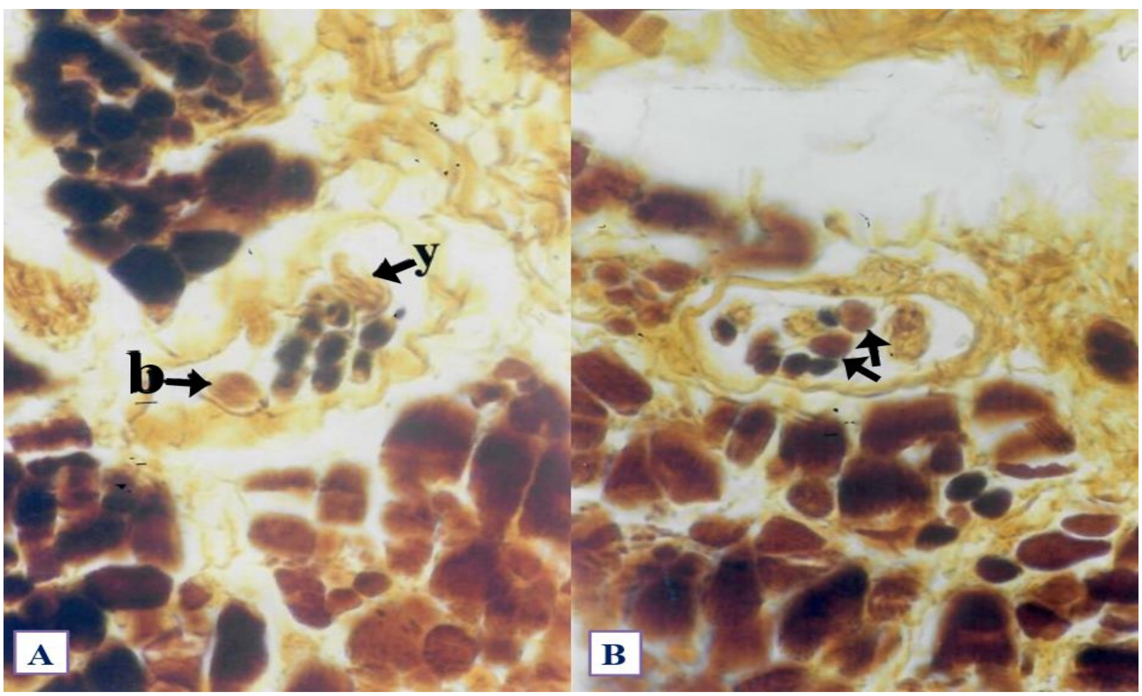

Figure 3 A Transverse section of muscle spindle in superior rectus showing one nuclear bag (b→) and intafusal fibers (y→B) there are two nuclear bag fibers (→) (Ranson silver x 400).

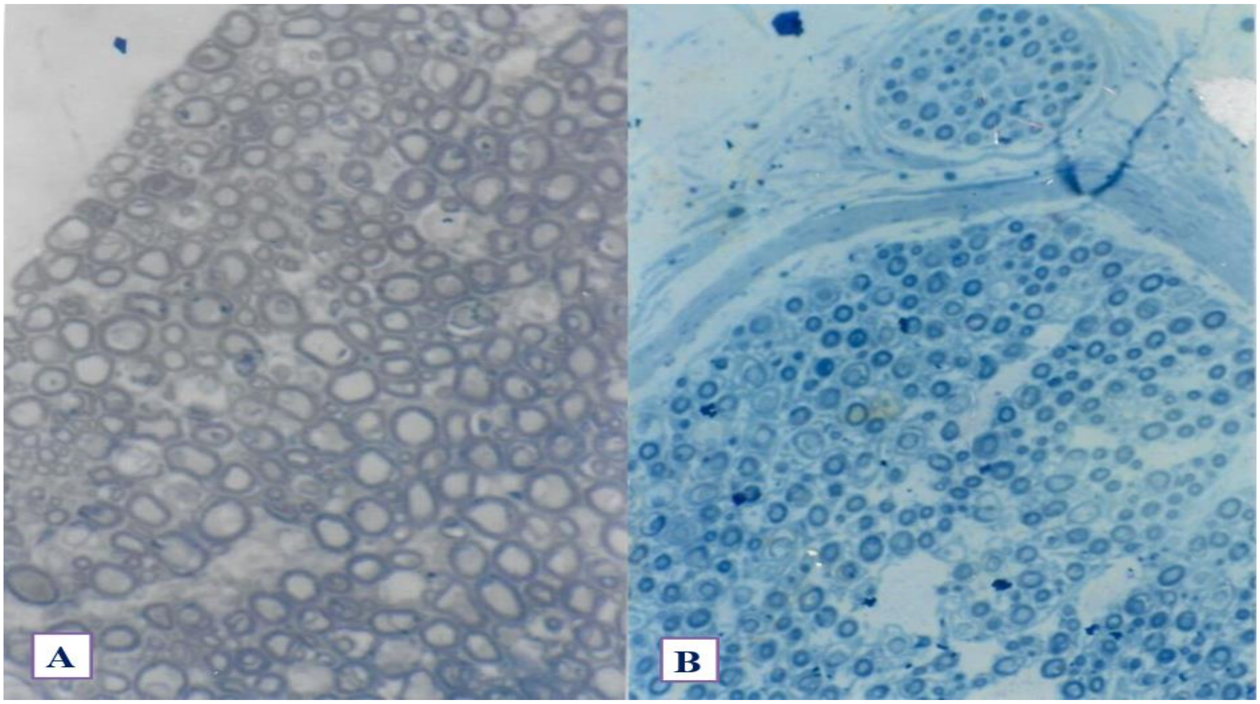

Figure 4 A: Near the brain stem, a semi-thin transverse segment of the oculomotor nerve's trunk reveals fiber-size components. (Toludine blue x 400) B: Semi-thin transverse sections in the trunk of abdicant nerve showing large main branch and small branch, the fibers of different sizes are mixed (Toludine blue x 200).

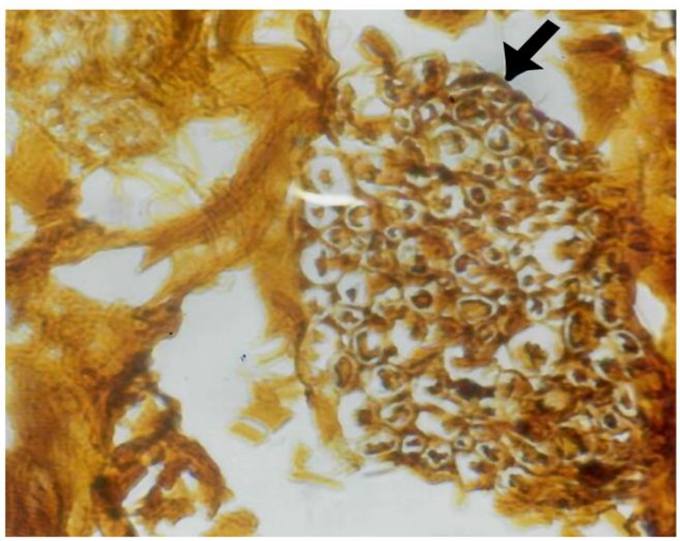

Figure 5 Transverse section of one branch of trochlear nerve. It contains fibers of different sizes (→) (Ranson silver x 400).

Table 1 showing the fiber size distribution in the trunk of oculomotor nerve

	2-4	4-6	6-8	8-10	10-12	12-14	14-16	16-18	18-20	20-22	Total
No of fibers	68	114	155	85	135	88	71	54	44	39	853
Percentage of fibers	7.9	13.3	18	9.9	15	10.5	8.3	6.3	5	4.5	

Table 2 showing the fiber size distribution in the trunk of abdicant nerve

	2-4	4-6	6-8	8-10	10-12	12-14	14-16	16-18	18-20	20-22	Total
No of fibers	27	39	66	40	50	33	22	20	18	16	331
Percentage of fibers	8	11.7	19.9	12	15	9.9	6.6	6	4.5	4.8	

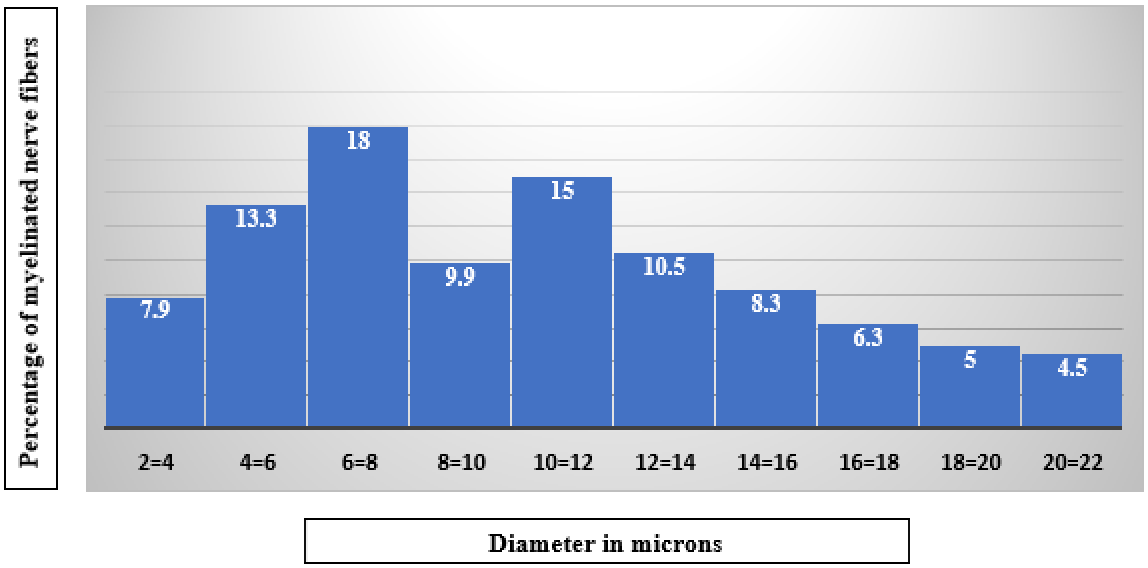

Figure 6 Histogram showing the fiber size distribution in oculomotor nerve. The range of fiber diameters is 2-22 microns. The fiber size distribution is bimodal with two peaks at (6-8) and (10-12) microns.

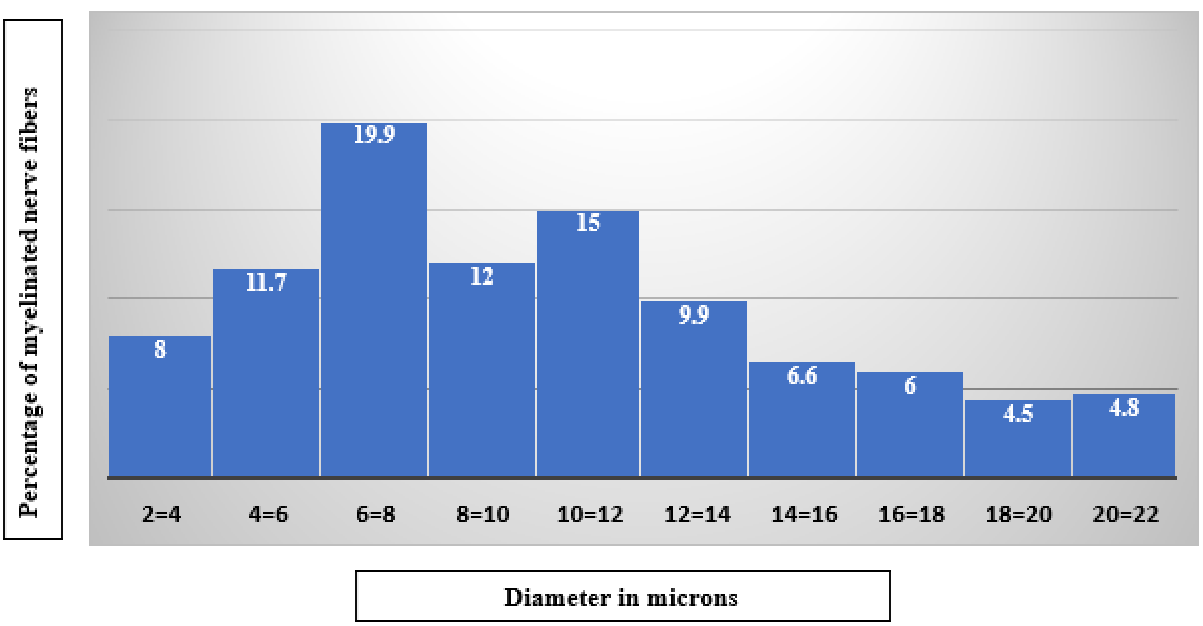

Figure 7 Histogram showing the fiber size distribution in abducent nerve. The range of fiber diameters is 2-22 microns. The fiber size distribution is bimodal with two peaks at (6-8) and (10-12) microns.

4. DISCUSSION

Six different types of fibers were found in the goat's extra ocular muscles. Orbital fibers with single innervations are the first type. This is the most common type of fiber found in the rectus and oblique muscles' orbital layer. It leaves a single point of dark neuromuscular contact with en-grappe edgings. The ending is located in the middle of the muscle fibers and is nourished by moderately thick nerve fiber. Orbital doubly innervated fiber was the second type. The remaining 20 percent of the orbital layer is made up of this fiber. This fiber type has several en-grappe nerve terminals spaced out throughout its length, which are fed by moderately thick orbital type I nerve fibers. Despite having several innervations, these fibers have a limited ability to twitch towards their center and have a sluggish contraction both near and far from it (Jacoby et al., 1989; Büttner-Ennever et al., 2007). It was associated with structural variation of myosin is forms along their length. The longitudinal changes in fiber type may be caused by innervations by motoneurons with various functional characteristics, according to (Jacoby et al., 1990). However (Anderson et al., 2011) found that the many plates on muscle fibers were supplied by the same nerve fiber, refuting this idea of multi neuronal innervations.

Extra ocular muscle has a relatively rich nerve supply, indicating that there are few motor units (number of muscle fibers per nerve) (up to 10 muscle fibers) (Lienbacher et al., 2011). The precision with which muscular force can be raised or decreased depends on the size of the motor unit. Small motor units, which often have fine control and enable gradual incrimination of force, are most suited for adjusting the force needed for eyeball fixation and movement big motor units, which are often attached to muscles that support posture and body weight, can only modify force in large steps (Eberhorn et al., 2005). Due to the distinct innervations system of the muscles, some muscle regions may be selectively activated during various movements or phases of the eyeball's particular movement (Demer et al., 2011). (Shin et al., 2019) state that new knowledge regarding the pattern of nerve distribution in the rectus muscles as determined by Silver's stain may be useful in comprehending the function of the muscles and the varied path physiology of strabismus. The precise location of these nerves may also shed light on the intricate mechanisms underlying physiological reflexes and the synchronization of eye movements.

5. CONCLUSION

We looked at various muscle fiber types that are present in the extra ocular muscles. The intricate design and operation of the extra ocular muscles control eye movements. It is still debatable how proprioception affects the regulation of eye movement particularly throughout the maturation of the visual sensory system, proprioception may determine visual orientation, regulate visual processing, and contribute to binocular function.

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Authors' Contributions

All authors contributed to the research and/or preparation of the manuscript. Ali Hassan A Ali, Abdulrahman S Binsaleh, Mohammed Obaid Alanaziand Mohammed Salman Alkhaldi participated in the study design and wrote the first draft of the manuscript. Bander Hather Alharthi, Saad Abdullah Alshnnanand Ahmed Othman Alghamdi collected and processed the samples. Saad Fahad Aljoidiand Faisal Rakan Alanazi participated in the study design and performed the statistical analyses. All of the authors read and approved the final manuscript.

Availability of Data and Materials

The data are available upon request from the authors.

Ethics Approval

All series of steps that were implemented in this study that included animal models were in compliance with Ethics Committee of Prince Sattam bin Abdulaziz University Institutional Review Board (PSAU-2022 ANT 18 /43PI).

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Conflict of interest

The authors declare that there is no conflict of interests

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