



Article

Morphology of Lower Limb Muscles in Mechanical Injury

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Abstract: The results demonstrated significant differences in muscle tissue response depending on the type of anesthesia used. Rats subjected to general anesthesia exhibited more pronounced muscle fiber necrosis, higher levels of inflammatory cell infiltration, greater edema, and delayed regenerative processes compared to those receiving local anesthesia at both 24 and 72 hours post-injury. In contrast, local anesthesia was associated with better preservation of muscle structure, higher vascular density, and more favorable conditions for tissue regeneration. These findings suggest that the choice of anesthesia plays a critical role in the severity of muscle damage and the dynamics of inflammatory and regenerative responses following mechanical injury.

Keywords: morphometry, edema, degenerative changes, muscle tissue regeneration

1. Introduction

Mechanical injury to skeletal muscle is a common pathological condition that may occur due to trauma, surgical procedures, or accidents. Such injuries often result in complex structural and functional changes within muscle tissue, including edema, inflammation, necrosis, and impaired regeneration. The severity of these changes depends on multiple factors, such as the extent of injury, vascular supply, and systemic physiological responses [1], [2]. Experimental animal models have been widely used to investigate muscle damage mechanisms and recovery processes. Understanding early morphological alterations is essential for developing effective therapeutic and anesthetic strategies. Therefore, studies focusing on muscle tissue responses after mechanical injury remain highly relevant in experimental and clinical medicine [3], [4].

Anesthesia is an integral component of experimental and clinical trauma management, as it directly influences physiological and metabolic processes. Both local and general anesthesia are commonly used during surgical interventions and experimental procedures involving muscle injury. However, anesthetic agents may differently affect tissue perfusion, inflammatory reactions, and cellular metabolism. These effects can subsequently alter the extent of muscle damage and regeneration following trauma. Despite widespread use, the impact of anesthesia type on muscle morphology after mechanical injury is not yet fully understood. This knowledge gap highlights the importance of comparative studies assessing tissue-level changes under different anesthetic conditions [5], [6].

Previous studies have reported that general anesthesia may induce systemic hemodynamic changes, leading to altered blood flow and tissue oxygenation. Reduced perfusion can exacerbate ischemic damage and intensify inflammatory responses in injured muscles. In contrast, local anesthesia is thought to preserve systemic stability and

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maintain better regional blood supply [7], [8]. Morphological investigations have demonstrated variations in necrosis, inflammatory infiltration, and vascular density depending on anesthetic exposure. However, many earlier studies focused on short observation periods or limited morphometric parameters. As a result, comprehensive temporal analyses of muscle tissue changes remain limited.

Morphometric analysis provides an objective method for assessing structural alterations in muscle tissue following injury. Parameters such as muscle fiber edema, inflammatory cell infiltration, degenerative changes, and regeneration are critical indicators of tissue response and healing potential [9]. Evaluating these parameters at different time points allows for a better understanding of injury progression and recovery dynamics. Early post-injury periods, particularly within the first 72 hours, are crucial for inflammatory regulation and tissue repair initiation. Differences observed during this phase may significantly influence long-term functional outcomes. Therefore, morphometric studies at 24 and 72 hours after injury are particularly informative [10].

Based on these considerations, this study focuses on comparing morphological and morphometric changes in lower limb muscles after mechanical injury under local and general anesthesia. The research addresses an important empirical gap regarding the role of anesthesia type in muscle tissue damage and regeneration. Using a controlled experimental rat model allows for standardized injury induction and reliable histological assessment. The findings are expected to contribute to a better understanding of anesthesia-related tissue responses. Moreover, the results may have implications for optimizing anesthetic choices in trauma and surgical settings. Ultimately, this study aims to support evidence-based strategies that minimize muscle damage and promote effective tissue regeneration.

2. Materials and Methods

This experimental study was conducted using an animal model to evaluate morphological changes in skeletal muscle following mechanical injury. A total of 48 adult, sexually mature rats aged 5–6 months and weighing 200–220 g were included in the study. The animals were maintained under standard laboratory conditions in accordance with established sanitary and ethical guidelines. All rats were clinically healthy and showed no behavioral abnormalities prior to the experiment. Individual identification was ensured using special body markings. The study design allowed controlled comparison of muscle tissue responses under different anesthetic conditions.

The animals were randomly divided into two experimental groups, each consisting of 24 rats. Group I received local anesthesia, while Group II was subjected to general anesthesia during the experimental procedures. Anesthesia was administered via intraperitoneal injection of bupivacaine at a dose of 150 mg/kg. Mechanical injury was induced in the lower limb muscles under sterile conditions. The injury model was standardized to ensure consistency across all animals. This grouping enabled evaluation of the influence of anesthesia type on post-traumatic muscle morphology [11], [12].

Muscle tissue samples were collected at two predetermined time points, 24 hours and 72 hours after injury. At each time point, animals were sacrificed in compliance with humane euthanasia principles. Euthanasia was performed under chloroform anesthesia by puncture of the left ventricle until complete exsanguination. Biopsies were obtained from the injured lower limb muscles during autopsy. The collected muscle specimens were immediately fixed in a 10% formalin solution. Proper fixation ensured preservation of tissue architecture for subsequent histological analysis.

Following fixation, muscle tissues were processed using standard histological techniques. The samples were dehydrated, cleared, and embedded in paraffin blocks. Serial sections with a thickness of 5–7 μm were prepared using a microtome. The sections were stained with hematoxylin and eosin for morphological evaluation. Light microscopy was used to examine structural changes in muscle fibers. All histological assessments were conducted under identical conditions to maintain consistency.

Morphometric analysis was performed to quantify structural alterations in muscle tissue [13], [14]. The evaluated parameters included muscle fiber edema, inflammatory

cell infiltration, degenerative changes, vascular density, and signs of muscle tissue regeneration. Measurements were obtained using standardized morphometric methods and recorded for each group and time point. The collected data were organized into tables for comparative analysis. Results were expressed as mean values with standard deviations. This methodological approach enabled objective assessment of anesthesia-related differences in muscle injury and recovery.

3. Results

The aim of the study was to compare morphometric changes in lower limb muscles after mechanical injury in rats receiving local and general anesthesia. Materials and methods. The experiment was conducted on 48 sexually mature rats, 5-6 months old (m=200-220 g). The animals were kept in standard conditions that meet sanitary regulations. Special marks on the body were used to identify the rats. During the experiment, the animals were healthy, with no changes in behavior. The rats in the experiment were anesthetized by intraperitoneal administration of bupivacaine (150 mg/kg). In two groups, on the 1st and 3rd days after, the muscles of the lower limb were taken during autopsy in compliance with the principles of humane treatment of animals, some of the animals were removed from the experiment by euthanasia, under chloroform anesthesia, by puncture of the left ventricle until complete exsanguination. The obtained biomaterial (muscle tissue) was fixed in 10% formalin solution. After fixation, the muscle tissue was excised and embedded in paraffin using the standard technique. Next, sections 5-7 μm thick were made and stained with hematoxylin and eosin. Results and discussion. These data allow a visual comparison of the degree of muscle tissue damage and the inflammatory response depending on the type of anesthesia in the two groups over time. Average area of necrotic fibers (mm^2): reflects the degree of muscle tissue damage. In the general anesthesia group, there is a larger area of necrotic fibers, which indicates more serious damage. The second indicator indicates the intensity of the inflammatory response. In the general anesthesia group, the percentage of inflammatory cells is higher, which indicates a more active inflammatory reaction. The next indicator is the state of the blood supply to the muscle tissue. In the local anesthesia group, the density of the vascular network is higher, which may indicate better blood supply. The last morphometric parameter measures the volume of edema in the muscle tissue. In the general anesthesia group, the level of edema is higher, which indicates more pronounced tissue damage. Based on the data obtained, the results were entered in tables 1 and 2.

Table 1. Morphometric data 24 hours after injury

Parameter Group I (local anesthesia)	Group II (general anesthesia)	Group II I (local anesthesia)
Average area of necrotic fibers (mm^2)	0,15 \pm 0,02	0,30 \pm 0,05
Percentage of inflammatory cells (%)	20 \pm 3	35 \pm 5
Vascular density (vessels/ mm^2)	8 \pm 1	6 \pm 1
Edema level (mm^3)	1,2 \pm 0,2	2,5 \pm 0,3

Table 2. Morphometric data 72 hours after injury

Parameter	Group I (local anesthesia)	Group II (general anesthesia)	Group II I (local anesthesia)
Average area of necrotic fibers (mm ²)		0,10 ± 0,01	0,25 ± 0,04
Percentage of inflammatory cells (%)		15 ± 2	30 ± 4
Vascular density (vessels/mm ²)		10 ± 1	7 ± 1
Edema level (mm ³)		0,8 ± 0,1	2,0 ± 0,2

4. Discussion

The present study demonstrates that the type of anesthesia significantly influences morphological changes in lower limb muscles following mechanical injury. Comparative morphometric analysis revealed more severe structural damage in animals receiving general anesthesia than in those subjected to local anesthesia. These differences were evident at both 24 and 72 hours after injury, indicating a sustained effect of anesthesia on tissue response. Increased necrosis, edema, and inflammatory infiltration were consistently observed in the general anesthesia group. In contrast, the local anesthesia group showed milder tissue alterations and earlier signs of recovery. These findings highlight the role of anesthetic modality in modulating post-traumatic muscle pathology.

One of the key observations of this study was the significantly larger area of necrotic muscle fibers in the general anesthesia group [15], [16], [17]. This suggests that general anesthesia may exacerbate ischemic and metabolic stress within injured muscle tissue. Systemic hemodynamic changes induced by general anesthetic agents can impair microcirculation and oxygen delivery [18]. Reduced tissue perfusion may increase susceptibility to secondary muscle damage following trauma. Previous experimental studies have similarly reported enhanced muscle degeneration under conditions of systemic anesthesia. Therefore, the observed increase in necrosis is consistent with existing morphological evidence.

Inflammatory response intensity was another important parameter affected by anesthesia type. The general anesthesia group demonstrated a higher percentage of inflammatory cell infiltration at both observation periods. Excessive inflammation can delay tissue repair and contribute to prolonged muscle dysfunction. In contrast, local anesthesia was associated with a more moderate inflammatory response. This controlled inflammation may create a more favorable environment for muscle regeneration. The findings support the hypothesis that limiting systemic anesthetic effects can reduce post-traumatic inflammatory burden [19].

Vascular density analysis revealed notable differences between the two experimental groups. Muscles exposed to local anesthesia showed higher vascular density, suggesting better preservation of microcirculatory function. Adequate blood supply is essential for nutrient delivery, waste removal, and activation of regenerative pathways. Conversely, reduced vascular density observed under general anesthesia may hinder effective tissue repair. These results align with the observed differences in edema severity between the groups. Enhanced edema in the general anesthesia group further indicates compromised vascular and lymphatic function [20].

Taken together, the results of this study emphasize the importance of anesthesia choice in experimental and clinical settings involving muscle injury. Local anesthesia appears to mitigate tissue damage, limit inflammation, and support regenerative

processes more effectively than general anesthesia. These findings may have practical implications for trauma surgery and postoperative management. Selecting anesthetic strategies that preserve local tissue homeostasis could improve healing outcomes. However, the study is limited by its experimental design and animal model. Future research should explore molecular mechanisms and long-term functional recovery to further clarify anesthesia-related effects on muscle regeneration.

5. Conclusion

Thus, the analysis of the evaluated morphometric indicators allows us to conclude that general anesthesia leads to more severe muscle tissue damage and a more pronounced inflammatory response compared to local anesthesia following mechanical injury. This effect was consistently observed at both 24 and 72 hours after trauma, indicating a sustained influence of anesthesia type on muscle morphology. Increased necrosis, edema, and inflammatory cell infiltration under general anesthesia suggest impaired microcirculation and delayed regenerative processes. In contrast, local anesthesia demonstrated a protective effect by preserving vascular density and supporting early tissue recovery. These findings highlight the importance of anesthetic selection in minimizing secondary muscle damage after injury. From a clinical perspective, the results suggest that local anesthesia may be preferable in procedures involving muscle trauma when feasible. **It is therefore recommended that clinicians and researchers consider the potential tissue-level effects of general anesthesia when planning surgical or experimental interventions.** Future studies are recommended to investigate molecular mechanisms and long-term functional outcomes to further optimize anesthesia strategies for muscle injury management.

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