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Injury and Apoptosis in the Palatopharyngeal Muscle in Patients with Obstructive Sleep Apnea-Hypopnea Syndrome

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Study Design A

Data Collection B

Statistical Analysis C

Data Interpretation D

Manuscript Preparation E

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Background: This study aimed to elucidate the possible activity of the mitochondrial-mediated apoptotic pathway (MMAP) in obstructive sleep apnea-hypopnea syndrome (OSAHS).


Material/Methods: A control group, a mild OSAHS group, a moderate OSAHS group, and a severe OSAHS group were included. Masson staining, hematoxylin and eosin staining, and terminal deoxynucleotidyl transferase dUTP nick-end labeling (TUNEL) assay were performed to assess collagen fiber hyperplasia, pathological morphology, and cell apoptosis, respectively, in muscle samples.

Results: In the OSAHS groups, the palatopharyngeal muscle fibers were larger, with apparent hypertrophy and increased elastic fiber content. The proportions of type I fibers were markedly higher in the control group than in the moderate and severe OSAHS groups ($P < 0.05$). Moreover, apoptosis was significantly enhanced in the muscle cells of the OSAHS groups. The Bax expression levels gradually increased across the 4 groups (lowest in the control group and highest in the severe OSAHS group) ($P < 0.05$); conversely, the p38 and p62 expression levels did not significantly differ among groups ($P > 0.05$).

Conclusions: A decrease in the proportion of the different fiber types can result in collapse of the upper airway. The pathogenesis of OSAHS appears to involve muscle cell apoptosis via MMAP.

MeSH Keywords: **Apoptosis • Autophagy • Muscle Fibers, Skeletal • Pharyngeal Muscles • Sleep Apnea, Obstructive**

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Background

Obstructive sleep apnea-hypopnea syndrome (OSAHS) is characterized by repeated collapse and obstruction of the upper respiratory tract during sleep, affecting the sleep quality of more than 10% of adults [1]. Previous studies have shown that upper respiratory muscle obstruction in patients with OSAHS occurs mainly in the postpalatal tissue [1–3], including the velopharyngeal soft tissue and palatopharyngeal arch. However, the mechanisms of OSAHS have been unclear. Most researchers believe that many factors, such as nerve injury, inflammation [4], type 2 diabetes [5], abnormal pharyngeal structure, and distribution pattern of muscle fibers, play an important role in pharyngeal collapse in patients with OSAHS and upper respiratory tract obstruction.

The function of the skeletal muscle in mammals is determined by various types of muscle fibers; the speed of strength generation, fatigue resistance, and energy metabolism depend on the type of muscle fibers (slow-twitch muscle fibers or anaerobic fast-twitch muscle fibers), which constitute the muscle [6,7]. The characteristics and efficiency of skeletal muscle regeneration depend on the injury method and skeletal muscle type (fast- versus slow-twitch type). For example, freezing-, notexin-, or cardiotoxin-induced regeneration is not accompanied by progressive replacement of the muscle by fibrous tissues; however, regeneration proceeds differently in slow- and fast-twitch muscle fibers, as demonstrated in the crush-induced model [8,9]. Previous studies have shown that the ratio of type I slow-twitch muscle fibers (Myh7) decreases in OSAHS palatopharyngeal muscles [10]. Decreasing Myh7 rates can cause fatigue, and affected patients are more prone to experiencing pharyngeal collapse during sleep [11].

Hypoxia results in increased apoptosis and autophagy, which has been confirmed in several studies in animal models and human tissues [11,12]. The main feature of OSAHS is frequent blood oxygen desaturation. Therefore, another objective of this experiment was to determine the expressions of protein factors related to the regulation of apoptosis and autophagy in the palatopharyngeal muscle of patients with OSAHS, and to assess the correlation between protein levels and OSAHS severity.

Herein, we studied the correlation between muscle fiber type distribution and OSAHS by examining myosin heavy chain (MyHC) I and MyHC II expressions in patients with OSAHS. Furthermore, we evaluated the expression levels of apoptosis regulatory factors in the pathogenesis of OSAHS and observed a significant increase in pro-apoptotic Bax expression in the OSAHS palate pharyngeal muscle tissue, which may have made the palatopharyngeal muscle tissues of patients with OSAHS more prone to apoptosis.

Material and Methods

Patients

This prospective study, including OSAHS and control group patients who underwent clinically indicated surgery in Tongji Hospital from October 2012 to June 2014, was approved by the Ethics Committee of Union Medical College Hospital affiliated to Tongji Medical College of Huazhong University of Science and Technology of the People's Republic of China (ethical number: S1001). Written informed consent was obtained from all participants before surgery.

The OSAHS group inclusion criteria were: adult age and male sex, OSAHS diagnosed using overnight polysomnography (PSG), performance of a physical or Müller's maneuver to assess oropharyngeal plane obstruction, and no history of pharyngeal or maxillofacial surgery. The control group inclusion criterion was: simple chronic tonsillitis without a history of snoring not meeting the diagnostic criteria for OSAHS on overnight PSG requiring simple tonsillectomy.

The exclusion criteria were: history of upper airway surgery, history of receiving radiotherapy and chemotherapy, history of mental illness, past or present treatment for OSAHS (e.g., complementary positive-pressure oral appliances) [13], liver or kidney dysfunction or history of heart failure; hypothyroidism, and history of human immunodeficiency virus positivity, amyloidosis, neuromuscular disease, or long-term alcoholism.

PSG

Preoperatively, all participants underwent overnight PSG (Embla S4000 32 System, Medcare Flaga, Iceland) in a sleep laboratory. Manual scoring of the electronic raw data was performed in accordance with the American Academy of Sleep Medicine criteria. Subjects with mild (apnea-hypopnea index [AHI] of 5–15/h), moderate (AHI of 15–30/h), or severe (AHI \geq 30/h) OSAHS were recruited, and those with an oxygen desaturation index (based on a 4% decrease in oxygen saturation) and 10–60 events per hour were assessed during a home-screening night [14].

Sample preparation

In both groups of subjects, muscle samples were obtained from the resected parts of the palatopharyngeal muscles exposed during surgery. All surgeries were performed under general anesthesia without the use of local anesthetics. Care was taken to avoid clamping or pulling on the sample tissues. The muscle tissues were cut into 2 parts using a sterile scalpel blade. One part was fixed in formalin and embedded in paraffin, while the other was rapidly frozen in liquid nitrogen, placed

into a sterile 1.5-mL centrifuge tube, and stored at -75°C until further processing.

Hematoxylin and eosin (HE) staining

The post-fixed palatopharyngeal muscle tissues were embedded in paraffin and cut into $5\text{-}\mu\text{m}$ coronal sections using a microtome. The paraffin-embedded muscle sections were deparaffinized with xylene and rehydrated using an ethanol gradient (100–70% v/v) (Tianjin Sheng Winton Chemical Co., Ltd., Tianjin, China), followed by washing with water. The sections were stained with 0.1% (w/v) HE (Nanjing Aoduofuni Biotechnology Co., Ltd., Nanjing, China) and examined under light microscopy (Olympus BX53; Olympus, Tokyo, Japan).

Masson staining

The tissue sections were fully fixed, dewaxed, and stained with 2 or 3 anionic dyes. The modified Mallory trichrome staining method was better adapted to the molecular size of the anionic dye and the tissue permeability of the sample. According to the permeability of different tissues, different sizes of anionic dyes are used to dye the tissue. The equipment used is the same as the fluorescent dyeing equipment. Collagen fibers, mucus, and cartilage are stained blue; muscle fibers, cellulose, and red blood cells are stained red; nuclei are stained blue-black.

Immunohistochemistry analysis

Immunostaining with antibodies directed against MyHC isoforms was performed using standard techniques. Tissue sections ($5\text{-}\mu\text{m}$ thick) were labeled with primary antibodies against slow-twitch muscle fibers (anti-skeletal myosin NOQ7.5.4D, M8421, 1: 1000; Sigma, Missouri, USA) and fast-twitch muscle fibers (anti-skeletal myosin MY-32, M4276, 1: 200; Sigma) and incubated overnight at 4°C . The second antibody was incubated with biotinylated immunoglobulin (Ig) against mouse IgG at 24°C for 30 minutes. Thereafter, diaminobenzidine was used as a substrate (NEOBIOSCIENCE, China), and horseradish peroxidase reaction labeled with Streptomyces antibiotic protein was used to detect the activity. Finally, counterstaining was performed using HE. Negative controls were prepared by replacing the primary antibodies with phosphate-buffered saline.

The sections were viewed and photographed using a light microscope (Leica, Germany) connected to a computer. Each sample was consecutively photographed at a $10\times$ enlargement objective. The numbers of types I and II fibers were counted and the overall percentages of the 2 types were calculated. Each is expressed as a percentage of the total number of fibers. The percentages of types I and II fibers in the mild OSAHS group (AHI of 5–15/h), moderate OSAHS group (AHI of 15–30/h), and severe OSAHS group (AHI ≥ 30 /h) are expressed as means \pm standard

deviations. The participants with an AHI < 5 /h were classified into the control group.

Western blotting

The protein expression levels of Bax, Bcl-2, and p38 in the palatopharyngeal muscle were determined by Western blotting. The total protein of the muscle tissues was extracted using a RIPA lysis buffer (Beyotime, Shanghai, China) according to the manufacturer's instructions. The protein levels were determined using a BCA protein assay kit (Beyotime).

Fifteen micrograms of each protein lysate was separated via 12% alkyl sulfate-polyacrylamide gel electrophoresis and transferred to a polyvinylidene difluoride membrane (Bio-Rad, Hercules, CA, USA). After incubation in a closed solution containing 5% skimmed milk Tris-buffered saline for 1 hour, the membrane was incubated overnight at 4°C , and the primary antibody was diluted as follows: Bax (1: 1000, Abcam), Bcl-2 (1: 2000, Abcam), P38 (1: 4000), and α -tubulin (1: 5000, Abcam). After being washed, the membranes were incubated with suitable horseradish peroxidase-conjugated secondary antibody (1: 5000; Antgene, Wuhan, China) for 1 hour at room temperature. The membranes were visualized in ECL Solution (Pierce Biotech, Inc., Rockford, IL, USA). Images of the membranes were captured using medical film (Kodak, Rochester, NY, USA). Quantitation of the detected bands was performed using Photoshop 5.0 software with α -tubulin used as an internal control.

Terminal deoxynucleotidyl transferase dUTP nick-end labeling (TUNEL) staining

In situ detection of apoptotic cells of the pharyngeal muscle was performed using the TUNEL kit according to the manufacturer's instructions (Roche Diagnostics, Mannheim, Germany). DAPI staining solution (Beyotime) was used to counterstain the nuclei for 5 minutes at room temperature. The sections were finally examined using a laser scanning confocal microscope (Nikon, Japan).

Statistical analysis

All data are expressed as means \pm standard deviations and were analyzed using SPSS 19.0 software. Comparisons among the 3 groups of the immunohistochemistry mean percentages and among the 4 groups of the Western blotting results were performed using one-way analysis of variance (ANOVA). P values of < 0.05 were considered statistically significant.

Table 1. Demographic features and mainly polysomnographic features of ninety subjects.

| | Mild OSAHS group | Moderate OSAHS group | Severe OSAHS group | Control group |
|--------------------------|------------------|----------------------|--------------------|---------------|
| Sample number | n=13 | n=7 | n=63 | n=7 |
| Age (year) | 31.54±13.45 | 36.29±9.8 | 37.86±7.34 | 22.57±5.83 |
| BMI (kg/m ²) | 23.65±2.5 | 27.12±1.79 | 27.55±3.31 | 23.44±4.0 |
| AHI (/h) | 9.7±2.97 | 24.49±5.09 | 65.96±20.97 | 2.13±0.84 |
| LSaO ₂ (%) | 89.23±2.31 | 74.14±8.09 | 65.17±13.27 | 89.6±4.04 |
| MSaO ₂ (%) | 95.57±1.5 | 94.63±0.5 | 91.65±4.29 | 95.04±2.29 |

* The demographic features and main PSG parameters were reported as mean±standard deviation.

Table 2. The proportion of fibers subtypes of palatopharyngeus muscle in control and severity of OSAHS groups.

| | Mild OSAHS group | Moderate OSAHS group | Severe OSAHS group | Control group | p |
|-------------|------------------|----------------------|--------------------|---------------|-------|
| Type I (%) | 31.55±8.16* | 27.74±9.15 | 22.53±10.6 | 30.93±10.9 | 0.011 |
| Type II (%) | 68.45±8.16 | 72.26±9.15 | 77.48±10.6 | 69.07±10.9 | 0.011 |

* Comparison of the proportion of fibers subtypes among the 4 groups was performed by one-way ANOVA. A p-value <0.05 was considered statistically significant.

Results

Patients' basic clinical characteristics

The enrolled subjects were all men, with a mean age of 36.77±9.074 years and a mean body mass index (BMI) of 26.86±3.3989 kg/m². Control muscle samples were obtained during tonsillectomy in 7 patients with chronic tonsillitis with no history of symptoms suggestive of sleep apnea. They had a mean age of 24.00±6.325 years and a mean BMI of 23.44±3.9797 kg/m² (Table 1). These patients had various degrees of clinical signs associated with OSAHS, including an AHI >5/h on overnight PSG. The patients with OSAHS had undergone uvulopalatopharyngoplasty (with bilateral tonsillectomy) for the treatment of OSAHS. They were divided into mild (AHI of 5–15/h), moderate (AHI of 15–30/h), and severe OSAHS (AHI ≥30/h) groups (Table 2).

Morphological changes in the palatopharyngeal muscle in HE and Masson staining

To examine the morphological changes in the palatopharyngeal muscles in the different OSAHS groups, HE staining and Masson staining were performed. As shown in Figure 1A, the stained collagen fibers are blue and the stained muscle fibers are red under optical microscopy. As the severity of OSAHS increased, the arrangement of the muscle fibers became more disordered, the area of the muscle fibers gradually decreased, and the proliferation of the connective tissues increased.

In contrast, in the control group, the muscle fibers were arranged neatly and tightly, the shape and size of the muscle fibers were regular, there was no obvious atrophy of the muscle fibers, and a small amount of collagen connective tissue could be seen around the muscle cells. Similar results were found in HE staining (Figure 1B). In the OSAHS group, the diameter of the muscle fibers became disordered; in the control group, the cross-section of the muscle fibers was round or polygonal, with a relatively uniform diameter, and a small number of small fibers were observed. Therefore, these data reveal that the morphological changes in the palatopharyngeal muscles become more disordered as the severity of OSAHS increased.

Type I/II fiber expression in the palatopharyngeal muscle

Repeated snoring, hypoxemia, and other factors in OSAHS patients can cause mechanical damage and inflammation of the upper respiratory tract muscle tissue [15]. In our study, immunohistochemistry was performed to determine the changes in types I and II fibers. The sections were viewed and photographed using a light microscope connected to a computer. Each sample was consecutively photographed with a 10× enlargement objective. The 2 fiber types were expressed in the cytoplasm (Figure 2A). In the control group, the proportions of type I and type II muscle fibers were 30.93±10.9% and 69.07±10.9%, respectively. In the mild OSAHS group, the proportions of type I and type II muscle fibers were 31.55±8.16% and 68.45±8.16%, respectively. In the moderate OSAHS group, the proportions of type I and type II muscle fibers were 27.74±9.15% and

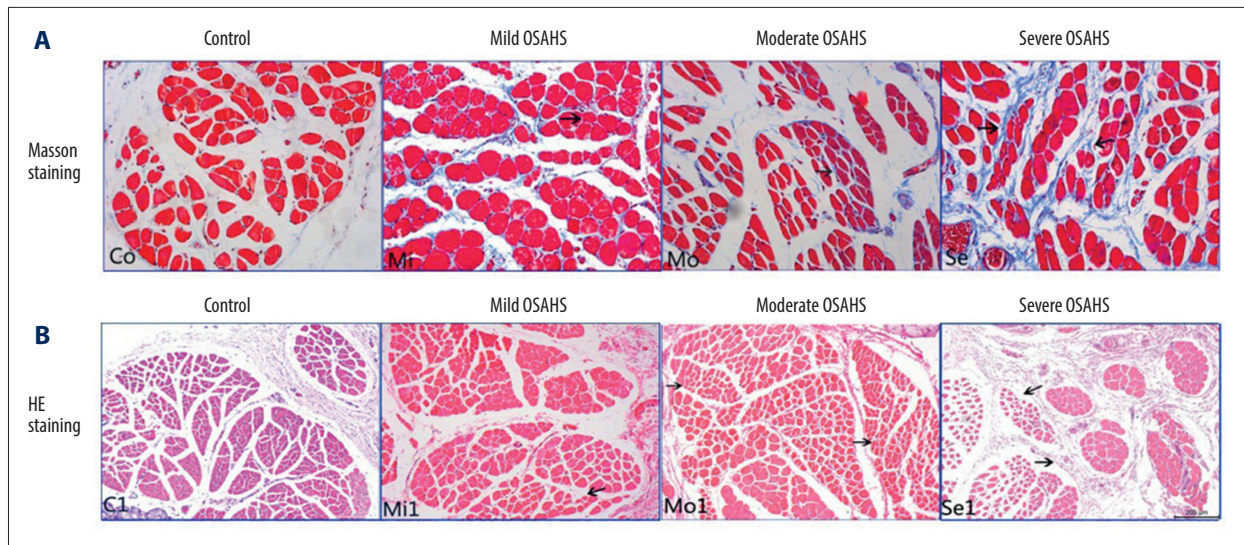


Figure 1. Morphological alterations in the palatopharyngeal muscle in the control group and the different obstructive sleep apnea-hypopnea syndrome (OSAHS) groups. **(A)** Masson staining of palatopharyngeal muscle tissues; scale bar, 50 μ m. **(B)** Representative images of hematoxylin and eosin (HE) staining of palatopharyngeal muscle tissues; scale bar, 50 μ m.

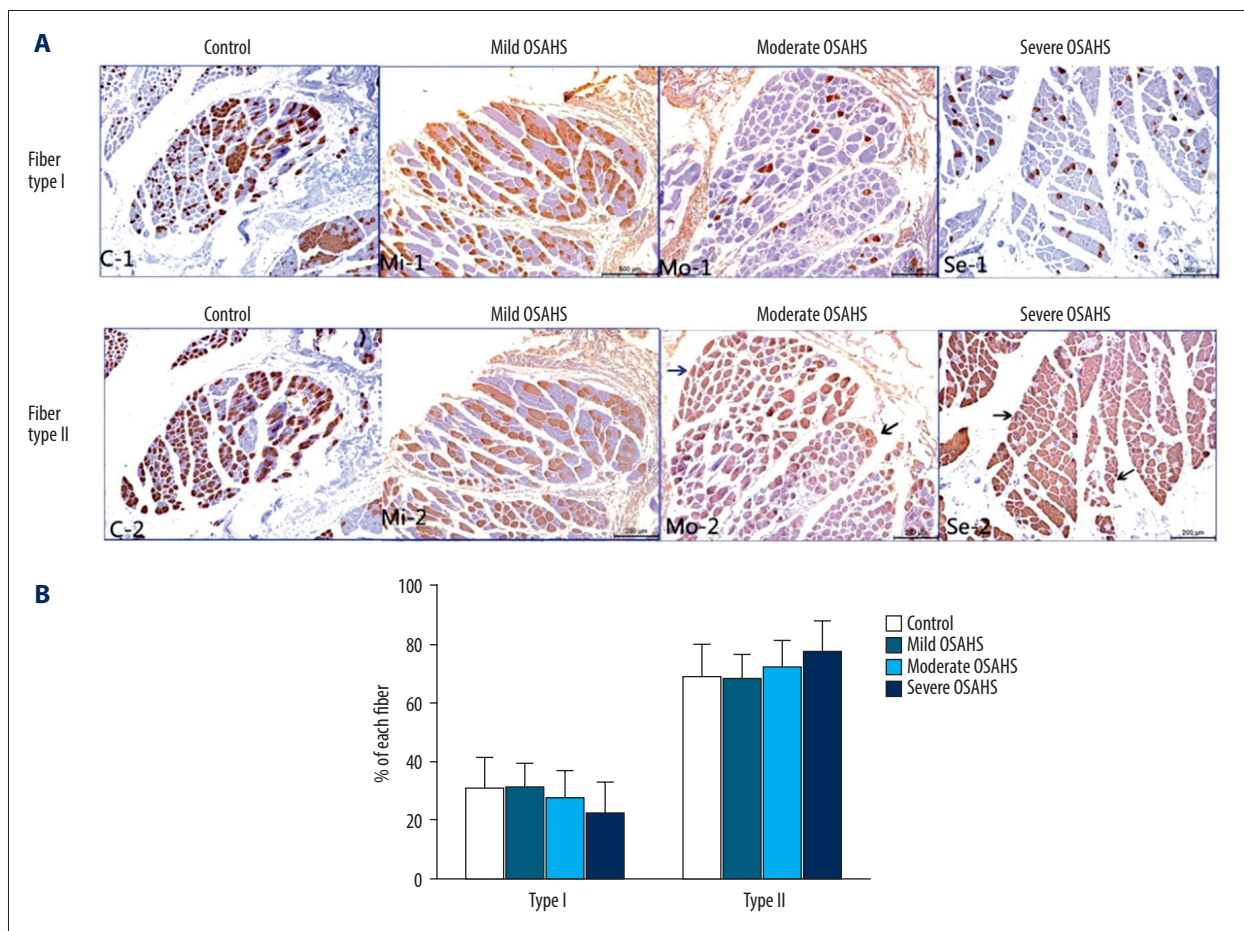


Figure 2. Representative immunohistochemistry of the palatopharyngeal muscle. **(A)** Representative image of type I/II fibers in the palatopharyngeal muscle tissue. **(B)** Proportion analysis of type I/II fibers in the obstructive sleep apnea-hypopnea syndrome (OSAHS) groups.

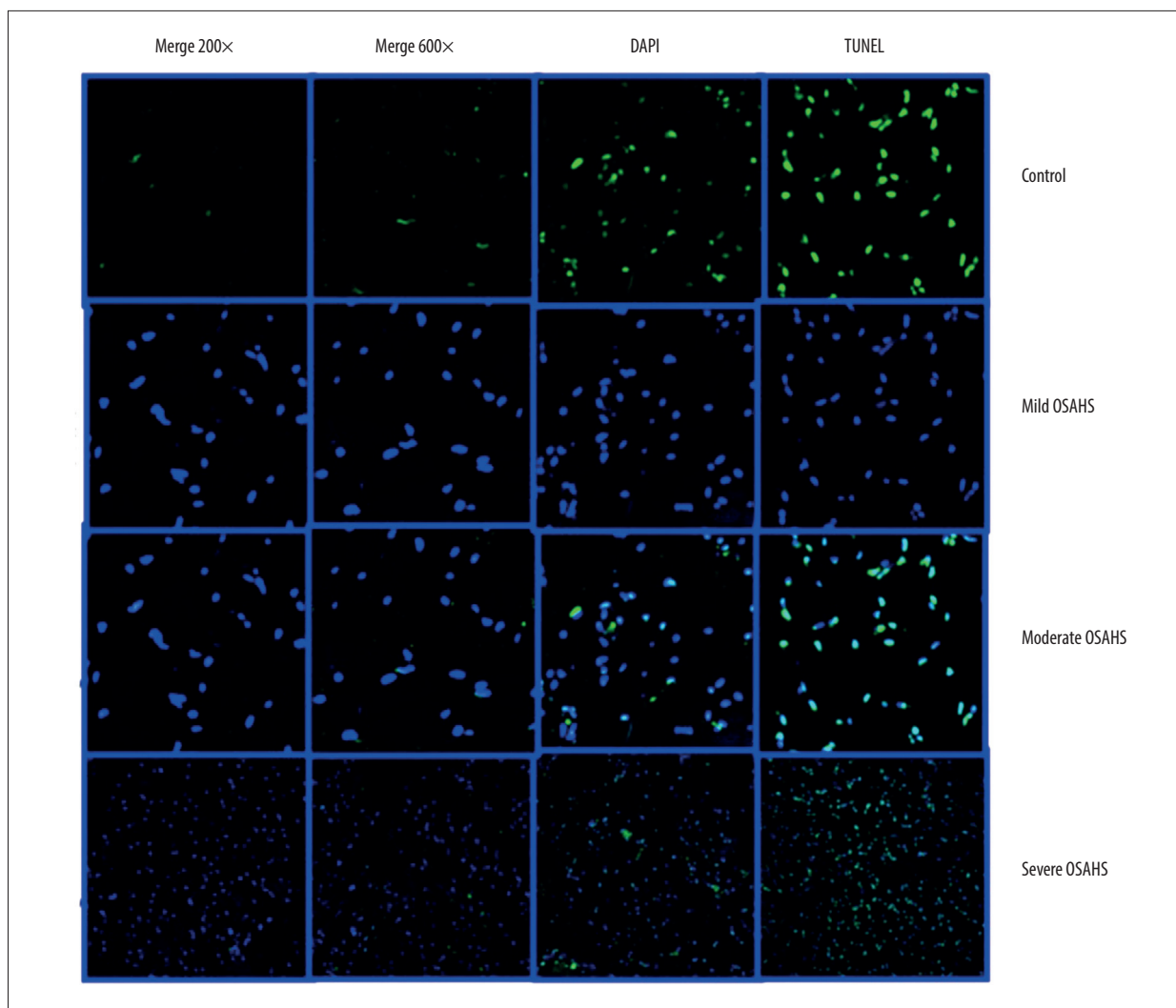


Figure 3. Muscle fiber apoptosis measured using terminal deoxynucleotidyl transferase dUTP nick-end labeling (TUNEL) assay. Representative immunofluorescence staining for TUNEL (green) and DAPI (blue) and merged images from the palatopharyngeal muscle fiber tissues of each group.

72.26±9.15%, respectively. In the severe OSAHS group, the proportions of type I and type II muscle fibers were 22.53±10.6% and 77.48±10.6%, respectively. As the OSAHS severity increased, the ratio of type I fibers gradually decreased, while that of type II fibers gradually increased (Figure 2B).

Palatopharyngeal muscle fiber apoptosis in the OSAHS groups

We performed TUNEL staining to evaluate the degree of muscle cell apoptosis. DAPI was used to visualize the cell nuclei; thus, the combination of TUNEL and DAPI staining depicted the proportion of apoptosis. In the control group, few TUNEL⁺ cells were observed in the palatopharyngeal muscle fibers. In the OSAHS groups, TUNEL⁺ cells were widely noted (Figure 3), and as the OSAHS severity increased, the number of TUNEL⁺ cells increased.

Relevant protein expression in the OSAHS groups

To elucidate the molecular mechanisms in chronic OSA-induced ventricular structural remodeling, we also detected the following apoptosis- and autophagy-related proteins, including Bax and p62. The pro-apoptotic Bax expression was upregulated in the OSAHS group compared with that in the control group, while the p62 expression did not differ significantly among the 4 groups (Figure 4A, 4B). In addition, the mitogen-activated protein kinase (MAPK) pathway was analyzed to determine the mechanism of palatopharyngeal muscle apoptosis. However, the p38 expression did not differ in the OSAHS groups (Figure 4A, 4B). Taken together, these data support that the p38 MAPK signaling pathway and cell autophagy may be unrelated to the apoptosis observed in patients with OSAHS.

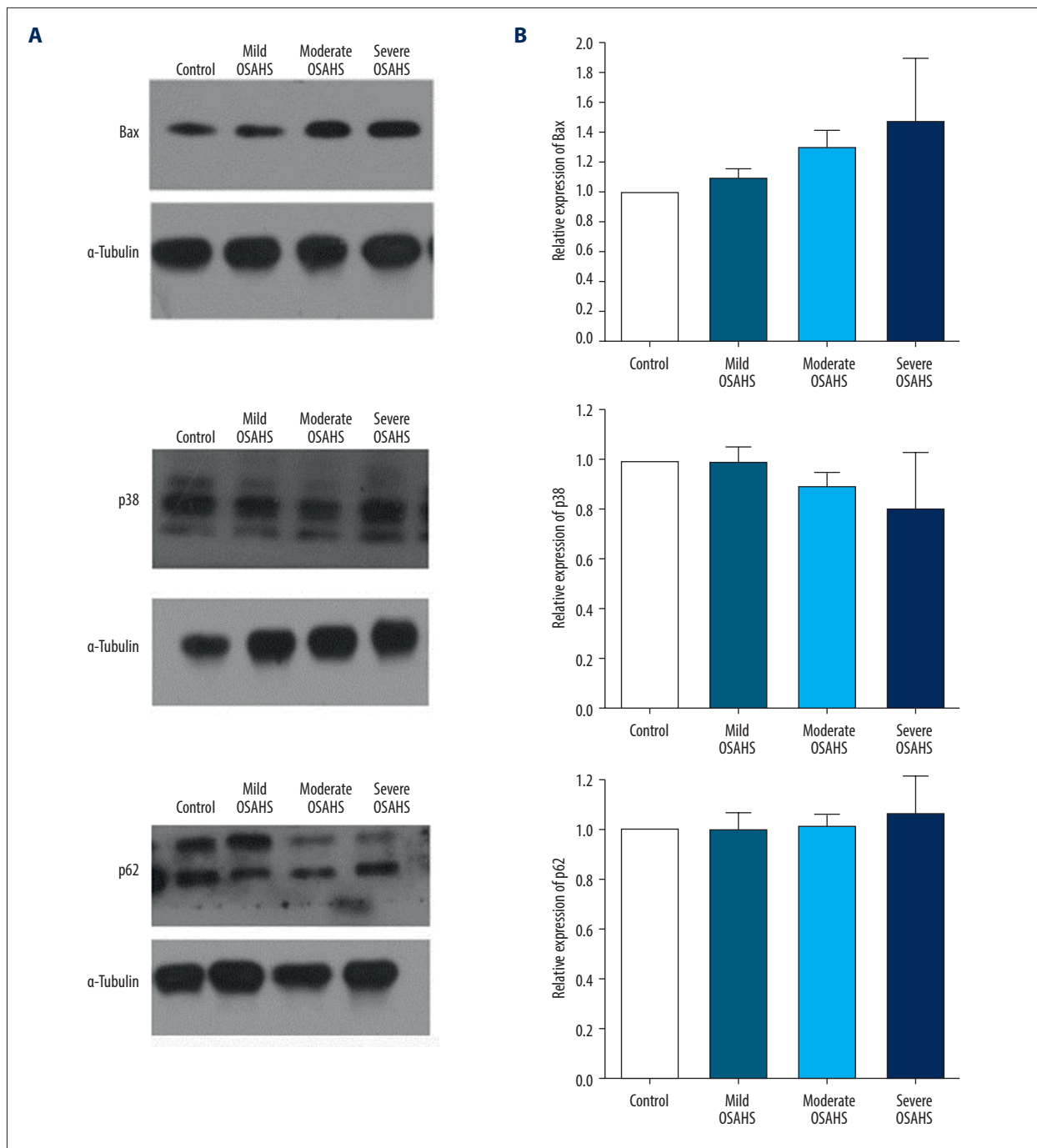


Figure 4. Expression of apoptosis-related protein in the patients with obstructive sleep apnea-hypopnea syndrome (OSAHS). (A) Representative band of Bax, p38, and p62. (B) Relative protein quantification of Bax, p38, and p62.

Discussion

In this study, several methods were used to investigate the differences in pathological morphology, muscle fiber subtypes, and proteins related to apoptosis and autophagy in the palatopharyngeal muscle between patients with OSAHS and controls. We initially drew the following conclusions. 1) Compared with

those of the controls, the muscle fibers of the patients with OSAHS were swollen, the nuclei were ingressed, the muscle fiber diameters were inconsistent, the fiber arrangement was disordered, the muscle fibers were atrophied, the fiber types were grouped together, and small round or angular fibers were observed adjacent to the perimysium. 2) Collagenous fiber hyperplasia was more common in the patients with OSAHS than

in the controls. 3) The proportion of type I muscle fiber (anti-fatigue muscle fibers) decreased in the patients with an AHI $\geq 30/h$ and did not decrease further at higher AHIs. 4) Bax expression gradually increased across the 4 groups (control group < mild OSAHS group < moderate OSAHS group < severe OSAHS group), while p38 and p62 expression showed no significant difference among them. These findings may be important in elucidating the development of potential treatments for OSAHS.

The pharyngeal muscle is one of the main dilatation muscles of the upper airway, and its functional status directly affects its role in maintaining airway patency. This muscle plays an important role in the pathogenesis of OSAHS, especially its pathomorphology [16]. This study investigated the relationship between the histopathological changes of the palatopharyngeal muscle and OSAHS in patients with mild, moderate, and severe OSAHS and the control group.

Under normal circumstances, components of the velopharyngeal structure have a certain proportion and arrangement. In patients with OSAHS, palatopharyngeal muscle atrophy increases with severity and finally transforms into fibroblasts, which causes connective tissue muscle denervation within the tissue and increases the proportion of collagen fibers. A previous study has found that collagen fibers could regenerate nerve fibers in the skeletal muscle [17] and form a physical barrier that separates the nerves from the tapering skeletal muscle fibers, resulting in denervated skeletal muscles. There are severe morphological abnormalities involved, the most significant changes of which are an increase in the amount of connective tissue, an abnormal change in muscle fiber size, an increased proportion of small fibers, and a large number of developmental stages of fiber MyHC isoforms [18–20]. However, some studies found different results. Smirne et al. [11] studied 9 cases and 4 cases of non-snoring and habitual snoring in patients with pharyngeal constrictor muscles, respectively; they found no neurogenic changes in the muscle fibers because intermittent hypoxia only affects specific pharyngeal muscles and nerves of the chin and tongue, while the impact on pharyngeal constrictor muscle innervation is relatively small. Our results are consistent with previously reported findings and indicate the existence of the denervation phenomenon in the palatopharyngeal muscles of patients with OSAHS. The constrictor muscle can lose its innervation from the corresponding nerve and undergo atrophy. Finally, the muscle function was damaged, which may be one of the pathogenic mechanisms of OSAHS.

In our study, we determined the phenotype of palatopharyngeal muscle fibers in each OSAHS group using histochemical staining techniques. The proportion of type II fibers was higher in the moderate and severe OSAHS groups, and the proportion of type I fibers was lower in the OSAHS groups than in

the control group. This change indicates that the phenotype of palatopharyngeal muscle fibers may vary with the severity of OSAHS. The proportion of type II fibers increased in the OSAHS groups as compared with that in the control group, suggesting increased muscle fatigue at night. At the same time, the proportion of type I fibers was lower in the OSAHS groups than in the control group, indicating that the anti-fatigue muscle was reduced. Previous studies have shown that under conditions of endurance training or low-oxygen and strenuous exercise loads, human muscles can be converted from type I to type II fibers, thereby increasing muscle resistance [21]. Further, the proportion of type I fibers in the palatopharyngeal muscles of patients with OSAHS decreases. Lower proportions of type I fibers can lead to fatigue and the muscle is more likely to collapse during sleep, which is similar to our findings. In patients with OSAHS, the palatopharyngeal muscle can adapt to the upper airway opening, causing nighttime stenosis by repeated intense contraction, similar to the effect of endurance training on different types of muscle fibers [22].

Certain factors in the body are stimulated when the apoptosis level changes, resulting in excessive or insufficient apoptosis and damage to the body. Intermittent hypoxia is a common factor that stimulates apoptosis and reportedly promotes apoptosis of the myocardial, liver, nerve, and other tissues. Previous studies have shown that antioxidants are significantly higher in chronic intermittent hypoxia groups than in control groups [23–25]. Although chronic intermittent hypoxia is the main feature of OSAHS, no studies on airway muscle apoptosis have been reported to date. Our study is the first to reveal the expression levels of apoptosis regulatory factors involved in the pathogenesis of OSAHS. Further, we found that pro-apoptotic Bax expression increased significantly in the OSAHS palate pharyngeal muscle tissues, which may explain why the palate pharyngeal muscle tissues in patients with OSAHS are at increased risk of apoptosis.

The Bcl-2 family plays an important role in the mitochondrial-mediated apoptotic pathway. Bax is the first identified homologous Bcl-2 gene. In this study, Bax expression in the pharyngeal muscle increased with the severity of OSAHS, suggesting that the mitochondrial-mediated apoptotic pathway is involved in the pathogenesis of OSAHS. Bax is an important pro-apoptotic regulator in this pathway [26]. The expression levels of Bax increased in the patients with OSAHS as the AHI increased, indicating that the higher the AHI of patients with OSAHS, the greater the degree of apoptosis in the pharyngeal muscle; the TUNEL staining results further confirmed this inference.

The p38 MAPK pathway, which is one of the main members of the MAPK family, is mainly involved in inflammatory cell response under stress and apoptosis, which can induce Bax translocation. In this study, we found that p38 expression in

the OSAHS palate pharyngeal muscle did not differ significantly, suggesting that it may not be involved in the OSAHS muscle apoptotic process.

Autophagy is a stress response that ultimately leads to apoptosis, such as organ dysfunction, metabolic stress, atrophy [27], chemotherapeutic intervention [28], pathogen infection, and starvation. If the relevant pressure is resolved, the cells usually regain their homeostasis and return to their initial state. In contrast, if the pressure persists and autophagy is no longer able to support cell survival, the cells can respond by activating apoptotic processes to ensure that they are controlled and effectively eliminated without causing local inflammation [29]. In fact, p62 plays a role in a variety of signal transduction pathway scaffold proteins [30] related to autophagy; however, no change in the p62 expression levels was observed in our study, which may indicate that autophagy was not involved in the pathophysiology of OSAHS.

The present study examined the expression levels of Bax, p38, and p62 in the palatopharyngeal muscles of patients with OSAHS. We found that the expressions of p38 and p62 did not differ significantly among the 4 groups. The preliminary inference was that apoptosis may be related to the incidence of OSAHS and is involved in its pathogenesis through the mitochondrial-mediated apoptotic pathway. The p38/MAPK pathways regulate apoptosis, while the p62 regulation of apoptosis and autophagy may be unrelated to the pathogenesis of OSAHS.

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Conclusions

Our study found that as OSAHS severity increased, the proportion of type I muscle fibers gradually decreased, while that of type II muscle fibers gradually increased. We also explored the possible mechanism of palatopharyngeal muscle injury through the mitochondrial-mediated apoptotic pathway, p38/MAPK pathway, and p62 autophagy. We found that myocyte apoptosis may be involved in the pathogenesis of OSAHS through the mitochondrial-mediated apoptotic pathway, not through the p38/MAPK pathway or p62 autophagy. All of the above-mentioned findings may serve as a basis for developing potential therapeutics for OSAHS. However, more studies with larger patient cohorts and potential targets of palatopharyngeal muscle injury should be performed in the future.

Conflict of interest

None.

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