

Basic Science

## Histological analysis of the rectus capitis posterior major's myodural bridge

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### Abstract

**BACKGROUND CONTEXT:** In recent literature, a soft-tissue communication between the rectus capitis posterior major (RCPma) muscle and the cervical dura mater has been identified. To the best of our knowledge, this communication has yet to be validated from a histological perspective nor has it been examined for neural tissue.

**PURPOSE:** The purpose of this study was to examine the composition and true continuity of the communication between the RCPma and the dura mater at a microscopic level. The communication was also inspected for the presence of proprioceptive neurons.

**STUDY DESIGN:** An anatomical and histological analysis of a novel structure in the atlantoaxial interspace.

**METHODS:** Gross dissection was performed on 11 cadavers to remove the RCPma, the soft-tissue communication, and a section of posterior cervical dura mater as one continuous unit. Paraffin embedding and sectioning followed by hematoxylin and eosin staining was conducted to validate the connection. Staining with antineurofilament protein fluorescent antibodies was performed to identify proprioceptive neural tissue on one specimen, and all findings were recorded via photographic documentation.

**RESULTS:** Histological investigation revealed a tendinous matrix inserting into both the RCPma and the posterior aspect of the cervical dura mater in all 11 specimens. In the one specimen examined for neural tissue, antineurofilament protein fluorescence revealed proprioceptive neurons within the communication. Immunoperoxidase staining demonstrated the insertion of these neurons into both the dura mater and the belly of the RCPma.

**CONCLUSIONS:** The existence of a true connection between the RCPma and the cervical dura mater provides new insight in understanding the complex anatomy of the atlantoaxial interspace. The presence of a neural component within this connection suggests that it may serve another function aside from simply anchoring this muscle to the dura mater. Such a connection may be involved in monitoring dural tension and may also play a role in certain cervicogenic pathologies. This study also supports previous reports that no true membrane joins the posterior arch of the atlas to the laminae of the axis and contradicts the conventional belief that the ligamentum flavum joins these two structures. © 2013 Elsevier Inc. All rights reserved.

### Keywords:

Rectus capitis posterior major; Cervical dura mater; Atlantoaxial interspace; Cervical spine; Myodural bridge

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## Introduction

Studies dating back to the 1920s describe soft-tissue attachments between the cervical dura mater and the posterior aspects of the atlas and axis [1]. Disregarded for some time, studies of these dural attachments have resurfaced and are the subject of increasing interest in the current literature [2]. Further analysis has revealed fibrous communications linking both the rectus capitis posterior muscles and the obliquus capitis inferior (OCI) muscle to the cervical dura mater [3–10] (Fig. 1).

In 1992, Kahn et al. [3] conducted an investigation focusing on the posterior intervertebral spaces of the craniocervical joints. In this study, the atlantooccipital interspace was found to contain a tissue bridge connecting the dura mater to the rectus capitis posterior minor (RCPmi) muscle. Examination of the atlantoaxial interspace revealed similar bridges between both the rectus capitis posterior major (RCPma) muscle and the OCI.

In 1995, Hack et al. [4] published a study that further examined the soft-tissue bridge between the RCPmi and the dura mater. This study focused specifically on the myodural communication and, in doing so, brought the suboccipital region under careful examination once again. It is now widely accepted that a true anatomical connection exists

between the RCPmi and the cervical dura mater in the atlantooccipital interspace [4,11,12]. Histological interpretation revealed that the RCPmi fascial connection fuses with the posterior atlantooccipital membrane, which ultimately coalesces with the cervical spinal dura [8,9,11]. It has been suggested that this myodural bridge plays a role in a variety of clinical manifestations, most notably cervicogenic headaches [4,13]. In one case, excision of the connection led to chronic headache relief [14].

In 2011, the first anatomical study was published on the soft-tissue bridge between the RCPma and the dura mater [10]. From a gross anatomical perspective, this soft-tissue communication appeared to be similar in nature to that of the RCPmi (Fig. 2). Evidence now supports that the contents of the atlantoaxial interspace include fibrous tracts originating from both the RCPma and the OCI, which attach to the cervical dura mater [3,10]. The atlantooccipital interspace contains similar soft-tissue components, which link the RCPmi to the dura mater [4,8,9,11,12]. It should also be noted that the ligamentum nuchae was thought to have a similar communication with the cervical dura mater through the atlantoaxial interspace [12,15], but this hypothesis was rejected in 2005 and is still under investigation [9].

Although the anatomical existence of these communications has been previously demonstrated, their origin



Fig. 1. Artist's depiction of the deep cervical spinal muscles. The rectus capitis posterior major (a), obliquus capitis inferior (b), and the obliquus capitis superior (c) are the three muscles that make up the suboccipital triangle. Original illustration produced by Frank Scali, DC.

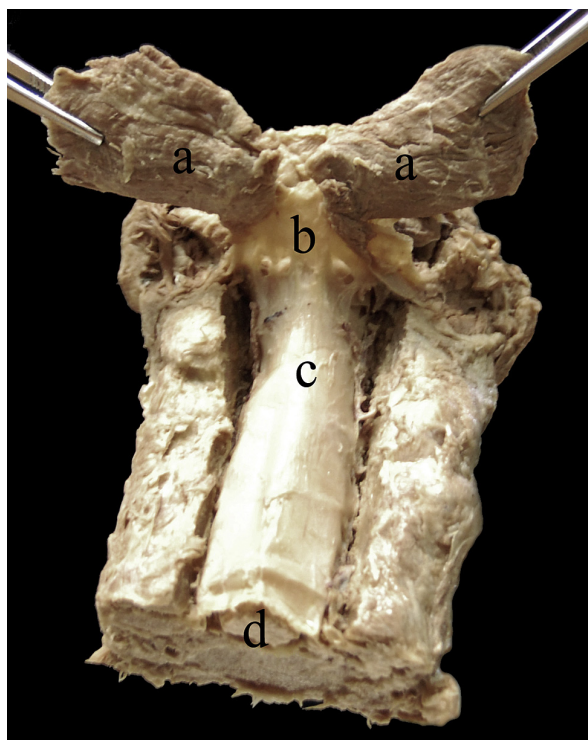


Fig. 2. Image of a cervical spine laminectomy revealing the dural attachment between the rectus capitis posterior major (RCPma) muscles and the cervical dura mater. Rectus capitis posterior major (a), attachment between the cervical dura mater and the RCPma (b), dura mater of the spinal cord (c), and spinal cord (d) at the level of the fifth cervical vertebrae.

and functions remain a matter of debate. It has been proposed that these soft-tissue fibers serve to monitor cervical dural tension during movements of the head and neck, but there is no direct evidence to support this [4,10,12,13]. The purpose of our study was to histologically evaluate the continuity of the myodural communication between the RCPma and the cervical dura mater and to examine the myodural bridge for proprioceptive neurons.

## Methods

A total of 11 human cadavers (6 males and 5 females) were examined in this study. Four cadavers (one male and three females) were selected at random from the Department of Anatomical Sciences at St. George's University, School of Medicine. Another seven specimens (five males and two females) were selected at random from the Department of Anatomical Sciences at Logan College of Chiropractic.

The objective of these dissections was to isolate the RCPma, a small section of cervical dura mater, and the fibrous communication as one continuous segment. Ten of the 11 cadavers were fixed using a formalin-alcohol-phenol solution and one was dissected 12 hours postmortem without being embalmed. None of the 11 specimens revealed any signs of anatomical variants, surgery, or trauma in the cervical area of interest. This study was conducted in accordance with all protocols for cadaveric research.

Using a hacksaw and a Stryker model 810 oscillating saw, the specimens were divided along the midsagittal plane from the most caudal region of the skull to the level of the seventh cervical vertebrae. A surgical scalpel separated the soft tissue from the suboccipital region so that each muscle could be identified in its midsagittal orientation. The fibrous communication between the RCPma and the dura mater was identified from a midsagittal perspective. The left RCPma was removed from its attachment at the inferior nuchal line and spinous process of the second cervical vertebrae from a medial approach. Once the RCPma was isolated from its osseous attachments, the fibrous communication was located in its distal attachment at the anteroinferior pole of the belly of the RCPma and was traced along to the posterior aspect of the cervical dura mater in the atlantoaxial interspace. To maintain the connection with both the RCPma and the dura mater, a 2×2 cm section of cervical dura was excised. The samples were fixed in neutral buffered formalin to be sent to the Department of Pathology at St. Louis University for analysis and interpretation of results.

All samples were dehydrated with 100% ethanol and xylene and infiltrated with melted paraffin wax. The samples were embedded in sectioning cassettes, and the wax was allowed to harden as it cooled to room temperature.

The paraffin block was then trimmed and faced for sectioning. Using a rotary microtome, 6- $\mu$ m-thick sections were cut and placed on glass microscope slides for rehydration and staining with hematoxylin and eosin. They were dehydrated once more and mounted under glass coverslips. Hematoxylin and eosin-stained sections were photographed with a Zeiss research light microscope using a  $\times 2.5$  objective lens. The images were recorded with a digital camera.

Several sections of each paraffin block were used for immunohistochemical analysis of one male specimen. Sections were rehydrated with xylene, graded ethanol, and distilled water for 3 minutes each at room temperature. Primary antibody (antineurofilament protein, 1:50, 1:250) was diluted in 1/10th blocking solution for 2 hours at room temperature (5% normal goat serum, 1% bovine serum albumin, and 0.25% Triton X-100 in phosphate-buffered saline [PBS]). Samples were then rinsed in PBS for 10 seconds and washed in PBS three times for 5 minutes each. A secondary antibody (goat anti-mouse rhodamine, 1:400) was diluted in 1/10th block solution at room temperature in the dark, for 1 hour. Samples were then washed in PBS three times, 5 minutes per wash. Finally, samples were rinsed using distilled water and mounted in FlouoroGel II with 4',6-diamidino-2-phenylindole using a #1.5 glass coverslip. In the control experiment, the primary antibody was replaced with normal mouse immunoglobulin G (1:125).

The remaining cut paraffin sections were used for immunoperoxidase staining. Samples were deparaffinized and rehydrated using xylene and graded ethanol for 3 minutes each. After antigen retrieval, samples were washed in PBS twice, 3 minutes per wash. Endogenous peroxidase was blocked in H<sub>2</sub>O<sub>2</sub>/methanol (100  $\mu$ L of 30% H<sub>2</sub>O<sub>2</sub> per 40 mL MeOH) for 30 minutes at room temperature. After washing in PBS twice (3 minutes per wash), wax wells were added and samples were blocked for 1 hour at room temperature in a humidity sealed container (1% bovine serum albumin, 5% normal goat serum, and 0.05% Triton X-100 in PBS). Samples were washed in PBS for 5 minutes. Primary antibody (neuropeptide protein 1:50, 150  $\mu$ L per slide) was diluted in antibody buffer overnight at 4°C in a sealed humid chamber. Controls were run with normal mouse immunoglobulin G (1:100, 150  $\mu$ L per slide). Samples were washed in PBS three times for 15 minutes each, and secondary antibody (goat anti-mouse peroxidase) was added. After three washes in PBS for 15 minutes each, the peroxidase substrate reaction mix was prepared, and sections were incubated in drops of reaction mixture for 15 minutes. The reaction was stopped by washing with distilled water, and hematoxylin diluted to 1:5 in distilled water was used as the counter stain (30 minutes). Samples were dehydrated through graded ethanol and xylene and mounted using a coverslip with Permount. Results from the antineurofilament protein and immunoperoxidase staining procedures were visualized with an Olympus 41BX

research light/epifluorescent microscope equipped with a DP72 digital camera system.

## Results

Histological analysis revealed that in all 11 samples, the communication between the RCPma and the dura mater was tendinous in nature. The tissue in all samples inserted directly into the RCPma and also attached to the posterior surface of the cervical dura mater (Figs. 3 and 4). In the one specimen analyzed for neurons, binding of antineurofilament protein indicated the presence of proprioceptive neurons throughout the length of the tissue (Fig. 5). Further staining with immunoperoxidase demonstrated the pattern of nerve distribution throughout the tissue (Fig. 6). The proprioceptive fibers were found to insert into the muscle (Fig. 6A), span the length of the fibrous connection (Fig. 6B and C), and insert directly into the cervical dura mater (Fig. 6D).

In all 11 cadavers, gross dissection revealed a firm attachment of the RCPma to the spinous process of the atlas. A dense conjunctive tract firmly adhered to the anterior surface of the RCPma and OCI, extended through the atlantoaxial interspace, continued anteriorward inside the epidural space of the spinal canal, and ended on the posterior surface of the dura mater. On excision of the RCPma, myodural bridge, and section of dura mater, it was apparent that the three anatomical structures were connected as one unit. Neither the ligamentum flavum nor any other true membrane was noted to join the posterior arch of the atlas to the lamina of the axis.

## Discussion

The myodural communication between the RCPma and the dura mater provides new insight in understanding

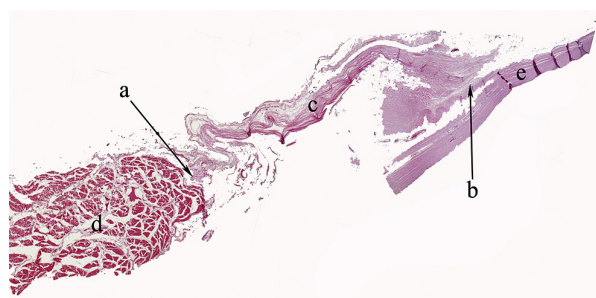


Fig. 3. Hematoxylin and eosin stain; right side sagittal section of the connection between the rectus capitis posterior major (RCPma) muscle and the cervical dura mater in a female cadaveric specimen. Histological analysis depicts the soft-tissue connection inserting into the belly of the RCPma (a) and the posterior aspect of the cervical dura mater (b). Also labeled is the soft-tissue communication between the dura and the RCPma (c), the muscle fibers of the RCPma (d), and the posterior cervical dura mater (e).

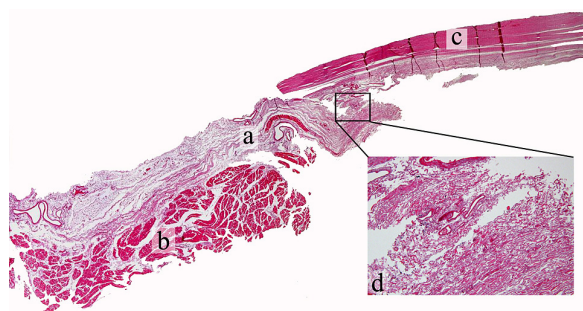


Fig. 4. Hematoxylin and eosin stain; left side sagittal section of the connection between the rectus capitis posterior major (RCPma) muscle and the cervical dura mater in a male cadaveric specimen. Histological analysis depicts the soft-tissue communication (a) between the RCPma (b) and the cervical dura mater (c). Provided is a higher magnification of the soft-tissue communication at the point of contact with the dura mater (d).

the complex anatomy of the atlantoaxial interspace. This study supports the suggestion by Kahn et al. [3] that no true membrane joins the posterior arch of the atlas to the laminae of the axis, which contradicts the conventional belief that the ligamentum flavum joins these two structures [11].

The contents of the atlantoaxial interspace were previously described as dense conjunctive tracts originating from the RCPma and the OCI. Analysis of these tracts revealed the presence of anterior fascia from both the RCPma and the OCI along with the elements of periosteal tissue [3]. Our study reiterates that there is a connection between the RCPma and the dura mater and also validates its continuity with both structures from a histological perspective. This study supports the evidence that the atlantoaxial interspace contains a fascioperiosteal connection that inserts directly into the RCPma and the cervical dura mater. The presence of proprioceptive tissue within this structure suggests that it may serve a purpose other than simply anchoring the RCPma to the dura mater.

Effective dural monitoring would require a system to relay information concerning alterations in tension to



Fig. 5. Sagittal section of the connection between the rectus capitis posterior major and the cervical dura mater depicting positive fluorescence after staining with antineurofilament protein antibodies.

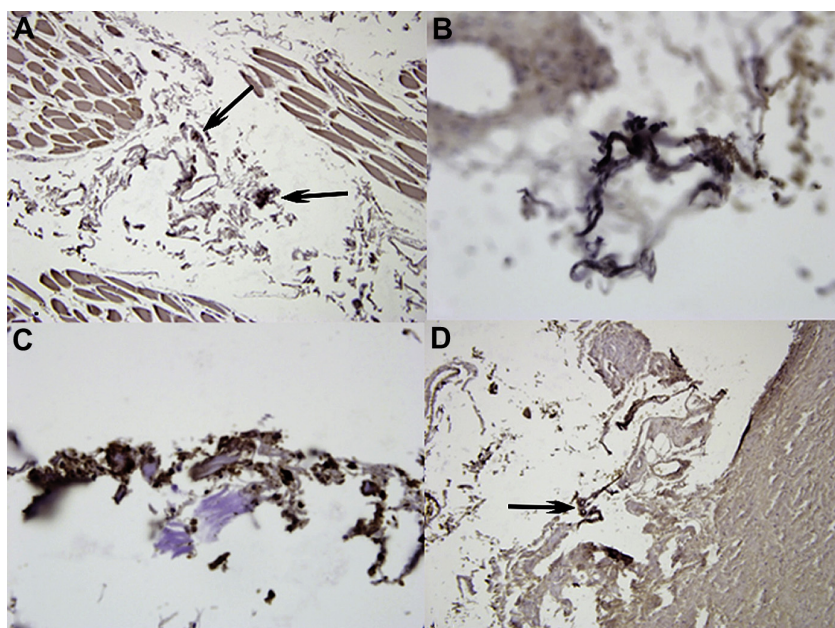


Fig. 6. Sagittal section of the connection between the rectus capitis posterior major (RCPma) muscle and the cervical dura mater. Immunoperoxidase staining indicating neural tissue (represented by arrows) within the belly of the RCPma (A), in the perimuscular soft tissue (B), in the peridural soft tissue (C), and inserting into posterior aspect of the cervical dura mater (D).

higher centers of the brain. Tension monitoring systems, such as the myotatic reflex, project information to integrating centers in the central nervous system via proprioceptive afferents [16]. It is reasonable to suggest that proprioceptive neurons may aid in transmitting information for compensatory realignment of the atlantooccipital and atlantoaxial vertebral joints.

We speculate that the existence of proprioceptive fibers within this communication establishes biofeedback of dural tension during ranges of motion of the head and neck. During these movements, adaptation of dural tension may rely on forces produced by the suboccipital muscles that communicate with the cervical dura. Because of the close proximity of the leptomeninges, it is possible that myodural biofeedback may play a role in maintaining the integrity of the subarachnoid space and, subsequently, cerebrospinal fluid pressure. If this mechanism does exist, its failure may result in a variety of clinical manifestations including those arising from increasing dural tension, namely cervicogenic headaches [13,17–19]. In one neurosurgical case report, excision of the analogous RCPmi myodural bridge significantly reduced chronic headaches and is consistent with the proposed etiology aforementioned [14].

In our study, immunohistochemical analysis was performed specifically on the myodural communication of the atlantoaxial interspace. Neurohistological studies should also be conducted on the myodural tissue connecting the RCPmi to the dura mater to gain a better understanding of the cervicooccipital region. Further investigation is also encouraged to clarify the function of

the neural component of this communication found in the atlantoaxial interspace. It should also be examined in a larger sample population, as this study only examined neuronal tissue in one specimen. The unusual nature of this attachment between a muscle and dura mater warrants further attention as it may provide insight to clinical manifestations involved in dural tension and other pathological conditions related to the integrity of the meninges.

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