

### Quantitative study of muscle spindles in suboccipital muscles of human foetuses.

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(Inf obl=242, sup obl=190, rcp major=98, lat pterygoid=20.3, opponens pollicus=17.3, trap=2.2, lat=1.4)

The proprioceptive inputs from the cervical musculature play an important role in head-eye co-ordination and postural processes. Deep cervical muscles in humans are shown to have high spindle content. The density, distribution and morphology of muscle spindles were studied in superior oblique capitis, inferior oblique capitis and rectus capitis posterior major and minor three small suboccipital muscles. The muscles were obtained, post-mortem from stillborn human foetus. The spindle density was calculated as the ratio of mean spindle content to the mean wet weight of that muscle in grams. The distribution and arrangement of spindles within the muscle and their arrangement was studied. The spindle density of superior oblique muscle was found to be 190, that of inferior oblique was 242 and the rectus capitis posterior contained 98 spindles per gram of muscle. No tendon organs were seen. The serial transverse sections of inferior oblique muscle revealed muscle spindles of varying sizes, length varying between 100-650 microns and, diameter 50-250 microns. A complex parallel arrangements of group of large spindles were seen in the belly of the inferior oblique muscle, while the polar regions contain few small isolated spindles. The relevance of such high spindle receptor content in these tiny muscles is discussed.

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>> [Abstract](#)

The proprioceptive inputs from the cervical musculature play an important role in head-eye co-ordination and postural processes. Deep cervical muscles in humans are shown to have high spindle content. The density, distribution and morphology of muscle spindles were studied in superior oblique capitis, inferior oblique capitis and rectus capitis posterior major and minor three small suboccipital muscles. The muscles were obtained, post-mortem from stillborn human foetus. The spindle density was calculated as the ratio of mean spindle content to the mean wet weight of that muscle in grams. The distribution and arrangement of spindles within the muscle and their arrangement was studied. The spindle density of superior oblique muscle was found to be 190, that of inferior oblique was 242 and the rectus capitis posterior contained 98 spindles per gram of muscle. No tendon organs were seen. The serial transverse sections of inferior oblique muscle revealed muscle spindles of varying sizes, length varying between 100-650 microns and, diameter 50-250 microns. A complex parallel arrangements of group of large spindles were seen in the belly of the inferior oblique muscle, while the polar regions contain few small isolated spindles. The relevance of such high spindle receptor content in these tiny muscles is discussed.

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The neck is regarded as an important proprioceptive organ for postural processes.<sup>[1]</sup> Disturbances of gait are demonstrated in experimental animals by interfering with sensory inputs from upper cervical region, either by damaging or anaesthetising, excision or sectioning of neck muscles, or cutting upper cervical dorsal roots.<sup>[1], [2], [3]</sup> Ipsilateral denervation of cervical musculature is found to have similar effect as that of unilateral labyrinthectomy. In man, dizziness and ataxia are seen to follow damage or anaesthesia of neck muscles, whip lash injuries, altered cervical vascularity or disturbances of cervical sympathetic tone.<sup>[1]</sup> In animals, convergence of vestibular and neck proprioceptive inputs are demonstrated at the level of vestibular nuclei,<sup>[4]</sup> thalamic neurons,<sup>[5]</sup> cerebral cortex<sup>[6]</sup> and also with the afferent inputs from the extraocular muscles, at superior colliculus.<sup>[7]</sup> The importance of proprioceptor inputs from dorsal cervical muscles, in the complex mechanisms of postural control and head-eye co-ordination is now well recognised.

It has been shown that the muscle spindle receptors of the perivertebral muscles of the cat are the major proprioceptors of the neck and not the joint capsules.<sup>[1]</sup> Indeed, high spindle content and their complex organisation is demonstrated in dorsal neck muscles of the cat<sup>[8], [9]</sup> In humans also, small deep cervical muscle of the neck, are known to have high spindle content.<sup>[10], [11]</sup> This work was aimed at studying the density, morphology and distribution of muscle spindles in three small suboccipital muscles in humans.

» Material and methods



The muscles were obtained, at periods varying from 24 to 48 hours postmortem, from the suboccipital region of four human stillborn foetuses, where the written consent was given by the parents, for the hospital disposal of the body. The foetuses with anencephaly, craniovertebral anomaly or other congenital CNS defects were not included.

Dissection : The suboccipital region was dissected to expose the suboccipital triangle, bounded by the muscles-rectus capitis posterior major, rectus capitis posterior minor (medially), superior oblique (laterally), and inferior oblique (inferiorly).<sup>[12]</sup> The posterior arch of atlas, the spine and the transverse process of axis vertebra, vertebral artery and first primary ramus were identified. Each muscle was identified from its origin to insertion, cleared from surrounding fat, fibrous tissue, and was removed as completely as possible. The muscle, rectus capitis posterior minor, placed medial to and overlapped by rectus capitis posterior major was removed along with the latter and the 2 were processed as a single muscle. 12 muscles were processed for teased preparations, one muscle (inferior oblique) was subjected to serial paraffin sections. The wet weight of each muscle was measured in saline before transferring it into the fixative for teased preparation. A linen suture was placed at the end of the muscle and muscle was kept under stretch over a wooden spatula, to maintain its orientation and to avoid shrinkage of the muscle. Teased silver preparations were used to observe spindle morphology, and quantification. The distribution and arrangement of the spindles within the muscle belly was studied by serial axial section of the muscle. The muscles were washed in Ringer's solution and

processed by Barker and Ip's modification of the deCastro's technique for the teased preparation. [13]

The muscles were fixed for 4 to 6 days in a freshly prepared solution containing chloral hydrate (1 gm), 90 % alcohol (45 ml), distilled water (50 ml) and concentrated nitric acid ( 1 ml). Following fixation, the muscles were washed for 24 hours in running tap water. Muscles were then placed in a solution containing 95 % alcohol (25 ml) and ammonia solution (0.1 ml) for 48 hours. This was followed by incubation at 37°C in 1.5 % aqueous solution of silver nitrate (w/v) for 5 days. Following silver impregnation, the tissues were reduced for two days in a freshly prepared solution of hydroquinone (2 gm) and 25 % formic acid (100 ml). Glycerine was then used to store the stained muscles. A minimum period of 4 to 6 weeks was allowed for softening of the tissues.

Once the muscles softened, the strips of fascicles were dissected along their length with a pair of fine needles under dissecting microscope at low magnification, and teased preparation of spindles were made. A muscle spindle was identified as an encapsulated spindle shaped structure with an innervating nerve at the equator (in most cases). The spindles were counted under the grid and the total counts were recorded for individual muscle. The mean weight and mean spindle content for each muscle was calculated by averaging the data from four muscles in each group. The spindle density was estimated for each muscle as a ratio of the mean number of spindles in the muscle (mean spindle content) to the mean wet weight of that muscle in grams. The size of muscle spindles, their arrangement, nerve fiber diameter, proximity to major blood vessel or innervating nerve, were noted. For making serial sections, the muscle was washed in Ringer's solution, processed through graded alcohol, and was embedded in paraffin wax. Serial sections were cut at 25 micron thickness, mounted on slides, and were stained with haematoxylin and eosin. Many projected drawings of spindles within the cross section of the muscle were made and put together, to understand the distribution of spindles within the muscle.

#### » Results



Teased preparation : The teased silver preparation of muscle fibers revealed muscle receptors in longitudinal section. A muscle spindle was recognised as a darkly stained, encapsulated, elegant spindle shaped structure with innervating nerve fiber at its equatorial region. [Figure.1] Most spindles were seen in the region of major blood vessel or nerve fiber. The diameter of the nerve fiber innervating the equatorial region was around 15 microns, near the spindle. The arrangement of spindles like tandem spindles, sharing common intrafusal fibers with fusion of the capsule at the ends, or, overlapping stacks of 3-4 spindles were also seen, commonly in the inferior oblique muscle. Many spindles were seen in circumscribed regions within the muscle, while other regions did not contain any spindles. Interestingly, no tendon organs were found in any of the three muscles. Since the muscles could not be freshly harvested, (within 24 hours), due to the process of autolysis, finer morphological details of the spindle (end plates, nerve endings, nuclei etc.) could not be well demonstrated. [Table I] shows the mean wet weight, mean spindle content and spindle density of three suboccipital muscles. The spindle density of other muscles in humans (as recorded by other workers) [11] is given in table II for comparison.

Serial paraffin section : Muscle spindles were seen in the interfascicular regions, commonly adjacent to the blood vessels or major nerve trunk. Very few, small spindles were seen at the polar regions of the muscle, while sections at the belly of the muscle revealed as much as 15 spindles in transverse section. The size of the muscle spindle varied significantly. The small spindles extended for around 150 microns, and had diameter of 50 microns at the equator. The large spindles were as long as 650 microns and the diameter of spindle capsule at the equator was around 200 microns. They were found to have around 4-5 bag fibers and 7-10 chain fibers. These large spindles formed clusters of 4 to 5, and were arranged in parallel, often sharing common capsule, over variable length [Figure. 2]. Such complex arrangement of large spindles was seen at the periphery of the muscle only in a certain region in the belly of the muscle, extending over a length of around 2.5 mm [Figure. 3]. Transverse sections through rest of the muscle belly, showed isolated medium sized spindles (length 250 microns, diameter 100 microns), scattered in the interfascicular region [Figure. 4]. Groups of 2 or 3 spindles were seen only at the periphery of the muscle. Arrangement of muscle spindles with few extrafusar fibers enclosed within the common capsule was also seen [Figure. 4]

» Discussion



The studies in humans have shown that the spindle density is highest in hand, foot and neck muscles, lowest in shoulder and thigh muscles and medium in the more distal muscles of the arm and leg. [10], [11], [14] In general, high spindle densities have been associated with small muscles subserving fine motor tasks, as in lumbricals. High spindle density (50-100/gm) and their complex arrangements (chain like complexes, 26 spindles linked in tandem, paired and parallel groupings) have been demonstrated in dorsal neck muscles of cat. [8], [9], [10] In humans also, deep muscles of the neck are known to have high spindle content. [10] However, their spindle density, distribution and morphology has not yet been studied. As indicated in table II, our study shows that the small suboccipital muscles have a very high spindle content and spindle density, when compared with other human muscles. Interestingly, the oblique muscles are found to have significantly higher spindle density than the recti. This is consistent with the findings of earlier workers, that rotating muscles like pronator quadratus and popliteus, have relatively high spindle densities as compared to the other distal limb muscles. [14] The disparity of the spindle density between the two obliqui, can be explained on the basis of the range of movement each muscle is subjected to. Superior oblique muscle is inserted from atlas to the inferior nuchal line. Only a limited rotational movement occurs at the atlantooccipital joint, and thus this muscle is subjected to lesser range of movement. On the other hand, majority of rotational movements of head on the neck occur at the atlanto-axial joint. The inferior oblique muscle which is diagonally inserted from spine of the axis to the lateral tubercle of atlas, would be subjected to stretch, during almost the entire range of rotational movement at atlanto-axial joint, and thus handles more proprioceptive input.

Significance of small muscles with high spindle density : The presence of high spindle densities in neck muscles concerned with head movement is reported in such disparate species such as man, rat and

cat, and must relate some way to the motor control of these muscles.<sup>[8], [9], [10], [11]</sup> The convergence of proprioceptive afferents from these muscles with vestibular and ocular inputs at various levels of neuroaxis is well recognized. The complex integrative mechanisms involved in head-eye coordination probably demands complex proprioceptive inputs from the neck muscles which probably is the reason for their high spindle content. Though, in humans large dorsal neck muscles like splenius and trapezius are reported to have high total spindle content, (which was attributed to their relatively large size) these muscles have low spindle density.

On the other hand the sub-occipital muscles studied here, are very small (mean weight 0.2-0.5 gm, in human foetuses), and seem incapable of bringing about any significant head rotation. Moreover, they are inserted very close to the craniovertebral joints and are at obvious mechanical disadvantage, as compared to the large powerful rotators of the head, like trapezius and splenius muscles, which are multisegmental and inserted laterally. Thus, the role of these muscles as the rotators or extensors of the head seems doubtful. Their closeness and diagonal arrangement around the joints (as in inferior oblique muscle) and their very high proprioceptive content make them ideal candidates as sensors of joint position and movements of craniovertebral joints. Indeed, physiological studies have shown that small flexion of upper cervical joints cause major changes in firing rate of spindle afferents from perivertebral muscles.<sup>[1]</sup> Also, the presence of such a high spindle content but paucity of tendon organs in these muscles, suggests that these muscles are functionally incapable of sensing contractile tensions but sense length changes and thus the movement. An analogy can be drawn between the lumbrical muscles of the hand, sensing and monitoring the joint movement, rather than causing the movement itself.<sup>[15], [16], [17], [18]</sup> However, the processing of spindle input may be different in lumbricals and sub-occipital muscles. While motoneurons of primate hand may be receiving monosynaptic excitatory input from intrinsic muscles,<sup>[16]</sup> kinesthetic information from the suboccipital muscles may be handled in more complex ways, as evidenced by convergence of vestibular, oculomotor, visual and neck proprioceptive inputs at various levels of neuroaxis.

#### » Conclusion



The present study indicates that the three small muscles of the suboccipital triangle, in humans have very high spindle content and spindle density, and show paucity of tendon organs. The possible role of these small muscles as movement sensors of craniovertebral joints needs to be evaluated further by electrophysiological and biomechanical studies.

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