Morphometric Evaluation of Posterior Funiculus Nerve Fibers in Relation to Aging

By

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Summary: We morphologically evaluated the size of axons in the posterior funiculus in different age groups and examined the changes due to aging. In the past, such studies have been conducted at the cervical spinal cord (C6) level, and a decrease in the size and number of axons due to aging has been noted. The current study was conducted at the lower lumbar spinal cord (L2) level.

Change in nerve fibers with aging is one of the major concerns in recent morphological and physiological research. A decrease in the number and area of axons in the posterior funiculus of the cervical spinal cord with aging has been proven¹⁾. However, no such study has yet been conducted at lumbar level. The difference between the two is that at the cervical cord level, the posterior funiculus is divided into two by the posterior median septum. It is further divided into fasciculus gracilis and fasciculus cuneatus by the posterior intermedial sulcus. But there is no such division at lumbar cord level. The aim of this study is to find if there are changes in morphometric data related to the aging process in the axons of the lumbar posterior funiculus.

Material and Methods

Research was conducted on spinal cords removed from 11 dissection cadavers (5 men and 6 women) ranging in age from 47 to 90 years (average age: 71 years). The causes of death were unrelated to diseases of the central nervous system. The same method as reported previously by the authors^{2–8)} was used for the preparation and mea-

surement of the spinal cord from dissections (fixation, washing, dehydration, embedding, staining, and morphometry). Spinal cord L2 level was identified according to the criteria published by Goto²⁾. The spinal segment were sliced to make transverse sections at 20 μ m thickness. This was followed by staining with the Luxol fast blue-periodic acid-Schiff-hematoxylin (LPH) method. As shown in Fig. 1, we divided both sides of the spinal cord at L2 into medial and lateral parts, and 3 sites from each, i.e. a total of 6 areas, were selected for measurement. The right and left sides of the posterior funiculus of the same specimens were used as sampling sites. Measurement was performed on a total of 22 samples. Using an eyepiece with a square grid and a high-magnification microscope ($\times 3,500$) equipped with an oil-immersion lens, we measured the number and cross-sectional area of myelinated axons in 6 sampling areas of 0.0030 mm² each. The image-analyzer consisted of (a) a microscope (BH2, Olympus, Tokyo, Japan) equipped with a drawing tube (BH2-DA, Olympus, Tokyo, Japan); (b) a digitizer (Crystizer α KC 3300, Graphic, Yokohama, Japan); and (c) a computer (NEC, Tokyo, Japan) with analyzing system software (original programming) for storage of data on-line, calculation and statistical analysis. A regression analysis

Fig. 1. Sampling area in human posterior funiculus at L2, bar = 1 mm.

Table 1. Morphometric data

Age group	N	Number of axons per section (/0.018 mm ²)	Average axonal area (μm ²)	Total axonal areas per section (μm ² /0.018 mm ²)	Average perimeter (μm)
A (40–49)	2	134.0 ± 1.00	3.20 ± 0.006	429.1 ± 2.33	6.25 ± 0.06
B (50–59)	4	129.2 ± 0.62	3.13 ± 0.032	404.9 ± 3.08	6.04 ± 0.06
C (60–69)	4	107.7 ± 5.69	2.72 ± 0.140	295.4 ± 30.5	5.71 ± 0.10
D (70–79)	6	91.33 ± 4.83	2.65 ± 0.092	244.2 ± 21.5	5.52 ± 0.05
E (80–89)	4	93.25 ± 8.75	2.63 ± 0.110	247.8 ± 32.5	5.59 ± 0.03
F (90–99)	2	77.50 ± 3.50	2.63 ± 0.085	204.4 ± 15.8	5.46 ± 0.04

was performed between age and each of the number of fibers, axonal area and axonal perimeter.

Results

Quantitative measurements for the number, cross-sectional area and perimeter of myelinated axons from respective age groups are listed in Table 1. Most of the nerve fibers in the posterior funiculus of the spinal cord were consisted of myelinated fibers. A small number of unmyelinated fibers can also be seen as shown in Fig. 2.

Number of axons

The number of myelinated axons in an area of 0.018 mm² (total of the 6 sampling areas) in the posterior funiculus was 104.1 (ranging from 74 to

135) on average. A statistical analysis was performed for each age group using the one-way ANOVA. The number of axons for groups D (70–79 years), E (80–90 years), and F (90–100 years) was significantly smaller than that for groups A (40–49 years) and B (50–59 years) ($p < 0.001$). A scatter diagram and regression analysis between number and age is shown in Fig. 3. It indicates a decrease in number with aging ($r = -0.86$, $p < 0.001$, Fig. 3).

Size of axons

The average cross-sectional area of myelinated axons in the posterior funiculus was 2.79 μm² (ranging from 2.40 to 3.23 μm²). The average total cross-sectional area for the six sampling areas was 296.6 μm² (ranging from 183.9 to 431.4 μm²) per unit area of 0.0030-mm². One-way ANOVA and

Fig. 2. Photograph of myelinated nerve fiber in human posterior funiculus, bar = 20 μm .

Fig. 3. Linear decrease in the number of spinal nerves in a posterior funiculus region of 0.018 mm^2 with aging ($p < 0.001$).

Fig. 4. Scatter diagram and regression line for the correlation between aging and average axonal area at the L2 level of the posterior funiculus ($p < 0.001$).

regression analysis were performed. The decrease in the area size was larger in groups D (70–79), E (80–90), and F (90–100) than in groups A (40–49) and B (50–59) (significant difference at $p < 0.001$), as in the case of the number of axons. A scatter diagram and regression analysis between cross-sectional areas (average and total) and age are shown in Figs. 4 and 5. These figures indicate a de-

crease in cross-sectional area with aging ($r = -0.74$, $p < 0.001$, Fig. 4; $r = -0.85$, $p < 0.001$, Fig. 5).

Perimeter of axons

The average axonal perimeter in the spinal cord was 5.72 μm (ranging from 5.35 to 6.32 μm). A significant decrease with age was noted for this as well ($r = -0.87$, $p < 0.001$, Fig. 6).

Fig. 5. Scatter diagram and regression line for the correlation between aging and total axonal area at the L2 level of the posterior funiculus ($p < 0.001$).

Fig. 6. Scatter diagram and regression line for the correlation between aging and the axonal perimeter at the L2 level of the posterior funiculus ($p < 0.001$).

Discussion

The posterior funiculus is known to be a conducting pathway for discriminative tactile, vibratory, joint position and motion sensation. Poviton⁹⁾ clinically examined various neurological changes at ages from the 20s to the 80s, and found that most of them were connected with the aging process. Those affecting vibratory sensation, in particular, were markedly apparent.

Up to now, there have been few reports on the morphological changes in human spinal nerve fibers

caused by aging. A morphological comparison of individual age groups at the cervical cord level conducted on myelinated axonal fibers in the posterior funiculus has shown a decrease in number, area, and perimeter of myelinated neuronal axons duaring the aging process¹⁾. The posterior funiculus is divided into two by posterior median septum at cervical and thoracic cord levels, and is further devided into the fasciculus gracilis and fasciculus cuneatus. This is not seen at the lumbar cord level. This study adopted a similar method to the one used in our previous reports¹⁾ at the lumbar cord level. A significant age-related linear correlation was observed for all parameters: number, average area, total area, and average perimeter of myelinated axons within the sampling area. In other words, this proves that the aging process affects not only the cervical level but the lumbar cord level as well. We may presume that the rest of the spinal cord (thoracic and sacral level) is affected in same way.

A similar method to the one used in this study could be applied to research on degeneration of the posterior funiculus, especially the funiculus gracilis, which affects fairly large proportion of elderly people.

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