ORIGINAL ARTICLE

Features of Structural Organization of The Human Facial Nerve

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ABSTRACT

Objective: The aim of the study was to investigate the intra-trunk structure of the facial nerve at different age periods.

Study Design: Cross-sectional comparative study

Place and Duration of Study: The data was collected from forensic morgues and clinical hospitals, as well as maternity hospitals in Baku, along with specimens from the primary museum collection of the Department of Human Anatomy. The study was conducted from November 2021 to May 2024.

Materials and Methods: Facial nerves were studied in corpses of those who died suddenly or because of various injuries, i.e. practically healthy people. The microscopic anatomy of the facial nerve was examined in 126 samples. To analyze the internal structure of the facial nerve, from trunk of the facial nerve were collected. Age groups – from fetus to old age. Araldite-Epon blocks were created from these samples following standard electron microscopy procedures. Quantitative indicators obtained during the research were conducted in the IBM Statistics SPSS-26 program with the application of variation and dispersion methods.

Results: The results of the research show that the facial nerve at the level of the stylomastoid foramen of an adult person has a diameter that fluctuates on the right within the range of 1697.4±51.1 μm, and on the left 1630.4±56.1 μm. This indicator is quite stable and changes little throughout a person's mature age. In cross-section, the nerve has an oval or irregular shape. The determining factors can be considered manifestations of asymmetry, individual and age-related variability.

Conclusion: The data obtained allow us to consider that the entire complex of morphological and functional restructuring of the facial nerve represents only individual links in the dynamics regarding the structural organization of the human peripheral nervous system.

Key Words: *Facial Nerve, Myeloarchitecture, Myelin Fibers, Nerve Bundles, Perineurium.*

Introduction

Questions of the intra-stem structure of peripheral nerves represent one of the most interesting pages of neuromorphology. In developing these issues, the basic patterns of morphogenesis and functional properties of the nervous system were established. To date, significant material has been accumulated on the intra-trunk structure of peripheral nerves and neural sheaths, the processes of myelination of myelinated nerve fibers, their age-related characteristics and a number of other issues have been shown.[1,2,3] It has been established that various parts of the peripheral nervous system are represented by nerve conductors that differ significantly from each other in light and electron microscopic organization as well as in functional properties. It is no coincidence that in recent decades the attention of many researchers has again been drawn to the examination of the morphology of the facial nerve, taking into account clinical practice and plastic surgery.[4,5,6] At the same time, a number of unresolved issues related to the morphology of the facial nerve still remain: there is no complete information about the age-related features of the intra-trunk structure of the facial nerve. Clinical manifestations and diagnostic accuracy of facial nerve lesions are primarily associated with the characteristics of its anatomical structure, and the prevalence of these lesions can be attributed to the nerve's susceptibility due to its unfavorable anatomical positioning. At the same time, the intra-trunk structure of the facial nerve has not yet been studied fully enough from the standpoint of modern morphology. The available data often require

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confirmation, often contradict each other, and, ultimately, do not allow for a complete, clear idea of the microscopic structure of the facial nerve. The issues of age morphology, considering the functional features of the nerve, have not yet been sufficiently developed, which is of fundamental theoretical and applied importance. At the same time, the solution of these issues can significantly deepen our understanding of the processes of myelination and intra-trunk reorganizations in the facial nerve, which will undoubtedly contribute to the further development of effective surgical methods and therapeutic measures associated with neurological, traumatic and other lesions of the facial nerve. Based on the above, the purpose of this study is to examine the age-related characteristics of the intratrunk structure and myeloarchitectonics of the facial nerve.

Materials and Methods

The microscopic structure of the facial nerve was analyzed in 126 specimens. The material was obtained by mutual agreement in forensic morgues and clinical hospitals, maternity hospitals in Baku, and preparations from the collection of the fundamental museum of the Department of Human Anatomy were also used. During microscopic examination, we pay attention to the features of the intra-trunk structure of the facial nerve: the number of bundles, the thickness of the peri- and epineurium, the content of myelin fibers in the nerves, and their topography. The composition of myelinated and unmyelinated fibers, along with their topography, caliber, and thickness, was also assessed. Samples were fixed in a solution containing 2% paraformaldehyde, 2% glutaraldehyde, and 0.1% picric acid, prepared in 0.1 M phosphate buffer (pH 7.4). Araldite-Epon blocks were created from the specimens according to established protocols used in transmission electron microscopy. Semi-thin sections (1–2 μm) cut from the blocks on a Leica EM UC7 ultramicrotome were stained with methylene blue, Azure II, or toluoid blue, viewed under a Primo Star microscope (Zeiss), and photographs of the relevant areas were captured using a Canon EOS D650 digital camera. Ultrathin sections were examined with a JEM-1400 transmission electron microscope at a voltage of 80–120 kV, and electrograms were obtained.[7,8]

The obtained digital data were statistically processed using medical statistics methods considering modern requirements. The average values of the obtained samples (M), their standard errors (m), minimum (min) and maximum (max) values of the series were calculated, and the frequencies of occurrence of the studied qualitative features in the series were determined. For a preliminary assessment of the difference between the variation series, the parametric Student's t-test and the assessment of the difference between the shares were used. Then, to check and clarify the obtained results, a non-parametric criterion was used - the Wilcoxon (Mann-Whitney) U-test. For comparison and probabilistic assessment of differences between the values of the compared groups with a small number of variants ($n<30$), we used the nonparametric Wilcoxon (Mann-Whitney) rank U-test. For this purpose, the numerical values of the compared samples were arranged in ascending order in one common row and ranked (numbered 1, 2, 3, etc.). When identical indicators were encountered in the ranking process, a lower number was assigned alternately to the indicators from the first and second compared groups. Calculations were performed on a computer using an EXCEL spreadsheet.[9] The results of the study were recorded, and the data were entered into appropriate tables for subsequent statistical processing.

Results

The results of the study showed that all structural components are registered on the cross section of the facial nerve in fetuses: connective tissue stroma, blood vessels and conductive elements. On average, the diameter of the nerve in the fetus reaches 236.4±29.7 μm on the right, 241.1±35.4 μm on the left. Connective tissue sheaths are already formed: the nerve trunk is surrounded by epineurium 29.7±2.0 μm thick, on the right 27.3±1.6 μm on the left. Each nerve bundle is surrounded by a perineural sheath reaching 16.6±0.9 μm on the right, 16.4±0.6 μm on the left. When studying the myeloarchitectonics of the facial nerve, a “nested” distribution of myelinated nerve fibers in the fetus is often noted, the total number of which reaches 305.9±50.5 in the right nerve, 319.9±50.5 in the left. In childhood, the cross-sectional diameter of the

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nerve on the right is 1205.9±53.6 µm, on the left 1097.4±55.5 µm. At the same time, the thickness of the epineural sheath increases. In the right nerve, this indicator was within 74.9±5.9 µm, in the left nerve this growth interval is 69.7±3.6 µm. Along with the increase in the thickness of the epineural sheath, the degree of development of the perineural sheath practically does not change. The ratio of the content of connective tissue stroma and conductive elements in the facial nerve shifts in favor of the latter: thus, the total area occupied on the cross-section by all myelin fibers in childhood is 17.1±1.4% in the right nerve, 16.1±1.1% in the left. The most variable indicator in childhood is the total number of myelin fibers. In childhood, the right nerve contains 3646.0±313.4, and the left 3278.0±299.0 of all types of myelinated fibers. Myelination of nerve fibers in postnatal ontogenesis has not only quantitative but also qualitative manifestations. Myelin sheaths have more pronounced tinctorial properties, they are more chromophilic. The most stable period in the structural and functional organization of the facial nerve is mature age. The facial nerve of a person in mature age has a diameter that fluctuates within 1697.4±51.1 on the right, 1630.4±56.1 µm on the left. This indicator is very stable and changes little throughout mature age. In the trunk of the human facial nerve, the number of nerve bundles is 6.0±0.51 on the right, 6.5±0.53 on the left. The central sections of the trunk are usually occupied by two or three large bundles, and in its peripheral sections there are several small bundles, usually represented by several dozen myelinated fibers. A feature of the bundle structure of the nerve is the regularity that the smaller the size of the bundles, the greater their number, and vice versa. Sometimes the nerve is composed of a small number of large bundles that differ little in shape from each other. The thickness of the perineurium is determined primarily by the diameter of each nerve bundle and fluctuates within the limits of 56.1 ± 5.6 µm on the right, 52.8 ± 5.5 µm on the left. The epineural sheath of the nerve is well developed. The thickness of the epineural sheath is 137.9±2.1 µm on the right and 135.5±6.0 µm on the left. Myelinated nerve fibers are the main structural elements of the nerve trunk in the facial nerve. In absolute figures, the total number of all myelinated nerve fibers in the right facial nerve is 5982.5±413.1, in the left 5277.3±513.5. It should be noted that by the end of mature age, the structure of the facial nerve gradually loses stability and is subject to certain involutional changes. In old and senile age, the indices of the connective tissue stroma of the nerve increase. The diameter of the nerve on the right in old age is within 1671.4±4.2 µm, on the left - 1659.1±4.6 µm, although the number of myelinated nerve fibers not only does not increase but decreases to a certain extent. The increase in the diameter of the cross-section is due to the increase in the thickness of its connective tissue sheaths. This is especially clearly manifested on the part of the epineural sheath, the thickness of which in old age reaches 135.4±1.8 µm on the right and 136.1±2.2 µm on the left, respectively. The previously established ratio between connective tissue and conductive elements acquires a clearly expressed tendency towards an increase in connective tissue stroma. For example, if in the right nerve in old age, the total area occupied by connective tissue is 89.4±1.0%, on the right, then the same indicator in old age reaches 82.3±2.0%. At the same time, there is a decrease in the percentage of the total area of conductive elements.

Table I: Micrometric Indices of the Facial Nerve

Indicators		Age groups			
		Fruit	Childhood	Mature	Old age
Diameter of the cross-section of the nerve trunk (in µm)	L	241,1±35,4	1097,4±55,5	1630,4±56,1	1659,1±4,6
	R	236,4±29,7	1205,9±53,6	1697,4±51,1	1671,4±4,2
p		<0,001	<0,001	<0,001	<0,001
The number of nerve bundles in the nerve trunk	L	2,7	3,7	6,0	4,7
	R	2,4	3,1	6,5	4,9
p					
Epineurium thickness (in µm)	L	27,3±1,6	69,7±3,6	135,5±6,0	136,1±2,2
	R	29,7±2,0	74,9±5,9	137,9±2,1	135,4±1,8
p		<0,001	<0,05	<0,001	<0,005
Perineurium thickness (in µm)	L	16,4±0,6	36,5±2,1	52,8±5,5	89,4±4,9
	R	16,6±0,9	38,3±2,0	56,1±5,6	93,1±9,5
p		<0,001	<0,005	<0,001	<0,001
Total number of myelinated nerve fibers	L	319,9±50,5	3278,0±299,0	5277,3±513,5	1855,7±62,3
	R	305,9±50,5	3646,0±313,4	5982,5±413,1	2360,0±52,6
p		<0,001	<0,001	<0,001	<0,001

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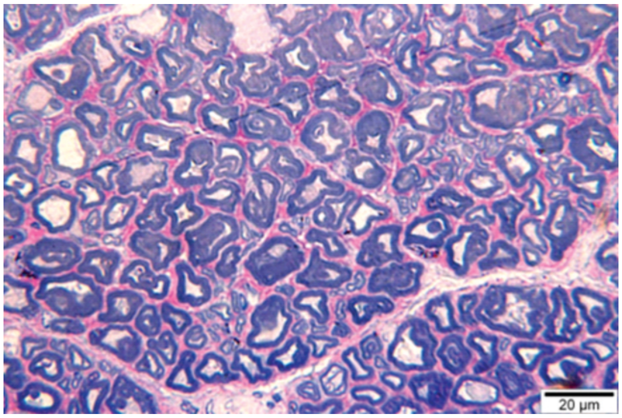

Figure 1. Intra-trunk structure of the facial nerve Age – 21 years Ultrathin sections of 50-70 nm thickness taken from the blocks were stained first with 2% uranyl acetate solution and then with 0.6% pure lead citrate prepared in 0.1 N NaOH solution. Ultrathin sections were examined under a JEM-1400 transmission electron microscope at 80-120 kV and electrograms were recorded

Discussion

A differentiated approach to the treatment of patients of different ages, the allocation of pediatric surgery and gerontology as independent branches of medicine, requires further study of the variability of organs and nerves in individual age groups. In this regard, further study of age-related features of the anatomy of the facial nerve can contribute to the improvement of surgical techniques. In our work, the criteria for morphological maturity of nerves were quantitative estimates of myelinated fibers of individual categories in the nerves of different ages. In the study, the intra-trunk structure of the facial nerve was studied. In this case, the myeloarchitectonics and age dynamics of the intra-trunk structure of the facial nerve were studied for the first time. In this regard, the novelty of the data obtained on the age-related myeloarchitectonics of the nerve is of interest. At the early stage of postnatal ontogenesis, the content of myelinated nerve fibers differs significantly; their total area is more than half the diameter of the nerve, the rest is accounted for by connective tissue structures. In the fetus, the predominant type is small, myelinated fibers. As the body grows the number of fibers in the facial nerve increases, and the ratio of their variety changes. It must be assumed that a significant part of the myelinated nerve fibers in the early stages of ontogenesis is undifferentiated myelinated fibers. In subsequent development, small, myelinated fibers pass into the category of medium and large myelinated nerve fibers. According to the data obtained, the fascicular structure of the nerves is established already during the period of intrauterine development and undergoes further restructuring in postnatal ontogenesis. Regardless of age, they can be either few-fascicular or multi-fascicular. Starting from childhood, the structure of the nerve acquires a more uniform character: the overwhelming majority of nerve fibers have a clearly formed sheath. It should be noted that the cross-sectional diameter of the nerve fibers does not always depend on the thickness of the myelin sheath; small, myelinated fibers may have a pronounced sheath and vice versa, large fibers are often surrounded by a very thin myelin sheath. The results of our studies are consistent with the data[10,11,12] on individual and age-related variability of the facial nerve. In this regard, there is no fundamental difference from similar processes occurring in other structures of the nervous system.[13, 14,15] Considering the morphological changes in the studied nerves in old and senile age, it should be noted that they are manifested both from the side of the conductive elements and the connective tissue stroma of the nerve. The total number of myelinated fibers decreases, the myelin sheath becomes thinner, loses its tinctorial properties. In some areas it is fragmented, unevenly stained with dyes. The results obtained are consistent with the materials presented in the works of[16,17] in relation to other peripheral nerves.

Conclusion

Thus, individual variability of the nerve is expressed in the number and size of nerve bundles, connective tissue elements, changes in the number and spectrum of myelinated fibers in different age periods. In the dynamics of the formation of the intra-trunk structure of the facial nerve and its branches, three main periods of development are noted: Period I intrauterine development up to and including childhood, when active growth and differentiation of myelinated nerve fibers and connective tissue are noted; Period II includes mature age, when the main indicators of nerves acquire relatively stable parameters; Period III occurs in old age, which is accompanied by involutional shifts and destabilization of conductive and stromal elements.

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Recommendations

The results obtained can be recommended and find practical application in the interpretation of the variety of symptoms of inflammatory polyneuritis and neuralgic syndromes and serve as the basis for the development and implementation of effective treatment methods.

Conflict of Interest

Authors declared no Conflicts of Interest

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CONFLICT OF INTEREST

Authors declared no conflicts of Interest.

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DATA SHARING STATMENT

The data that support the findings of this study are available from the corresponding author upon request.