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Running head: Arterial supply of the trigeminal ganglion

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ABSTRACT

Background: In this study, we explored the specific microanatomical properties of the trigeminal ganglion (TG) blood supply and its close neurovascular relationships with the surrounding vessels. Possible clinical implications have been discussed.

Materials and methods: The internal carotid and maxillary arteries of 25 adult and 4 fetal heads were injected with a 10% mixture of India ink and gelatin, and their TGs subsequently underwent microdissection, observation and morphometry under a stereoscopic microscope.

Results: The number of trigeminal arteries varied between 3 and 5 (mean 3.34), originating from two or three of the following sources: the inferolateral trunk (ILT) (100%), the meningo-hypophyseal trunk (MHT) (100%), and from the middle meningeal artery (MMA) (92%). In total, the mean diameter of the trigeminal branches was 0.222 mm. The trigeminal

branch of the ILT supplied medial and middle parts of the TG, branch of the MHT supplied the medial part of the TG, and the branch of the MMA supplied the lateral part of the TG. Additional arteries for the TG emerged from the dural vascular plexus and the vascular network of the plexal segment of the trigeminal nerve. Uniform and specific intraganglionic dense capillary network was observed for each sensory trigeminal neuron.

Conclusions: The reported features of the TG vasculature could be implied in a safer setting for surgical approach to the skull base, in relation to the surrounding structures. The morphometric data on TG vasculature provide anatomical basis for better understanding the complex TG blood supply from the internal and external carotid arteries.

Key words: trigeminal ganglion, trigeminal arteries, inferolateral trunk, meningo-hypophyseal trunk, middle meningeal artery

INTRODUCTION

The trigeminal ganglion (TG) is located in the middle cranial fossa i.e. in the central part of the skull base, within the dural Meckel's trigeminal cave (Fig. 1A). The meningeal dural roof of the cave is firm and thick in comparison to the thinner periosteal layer separating the TG from the underlying trigeminal impression of the petrous part of temporal bone (Figs. 1B, C) [2]. The TG lies below the temporal lobe of the brain, above the horizontal petrous segment of the internal carotid artery (ICA), and lateral and below to its cavernous segment, from which the TG receives its principal arterial supply. Lateral to the TG, vessels emerge from the middle meningeal artery (branch of the maxillary division of external carotid artery, ECA), and participate in the ganglionic blood supply. Both carotid sources, ICA and ECA, send branches for the TG, as well as for the surrounding dura mater [10]. Small arteries from the periganglionic arterial network penetrate into the ganglionic tissue and form the intraganglionic dense capillary network, feeding all the trigeminal sensory neurons [18]. It has been shown that the distribution of intraganglionic blood vessels in the TG is in close contact with trigeminal ganglionic cells, but separated from them by the intervening layer of the satellite glial cells [8]. Further, the dura mater, which is in close contact with the TG surface, is well innervated by the trigeminal sensory dural branches, and is supplied by the same vessels as the TG [12].

The surgical approach to the posteromedial part of the cavernous sinus, invaded by a pathologic lesion, implies a skull base route with splitting through a free vascular zone between the maxillary and mandibular trigeminal divisions, and followed by mobilization of the TG upward [21]. Direct embolization of small arteries arising from the cavernous segment of ICA is

applied in treating meningeoma, brain or dural arteriovenous malformations (AVM). A detailed knowledge of the vascular morphology of the cavernous segment of ICA is indispensable in accomplishing safe endovascular techniques, in order to minimize the risk of stroke, cranial nerve palsies and blindness [17].

MATERIALS AND METHODS

This micromorphological study on the trigeminal nerves and ganglions included 25 properly injected cadaveric heads of adult persons with a mean age of 65.9 years (range 46-74 y.) from the collection of the Laboratory for Vascular Anatomy of Institute of Anatomy. The internal carotid and maxillary arteries were beforehand perfused with isotonic saline solution, followed by injecting the trigeminal arterial system with a 10% warm mixture of India ink and gelatin. Then, the specimens were fixed in a 4% formaldehyde solution for 3 weeks. Additionally, we used two heads of fetuses (32 weeks gestational age), injected with India ink and gelatin and then fixed in 4% formalin solution, in order to perform the finest microdissection of the Meckel's trigeminal cave elements *in situ*. We accessed the middle cranial fossa after removing most of the calvaria and the overlying brain. Trigeminal blood vessels were carefully microdissected and examined under the stereoscopic microscope (Leica MZ6). Further, we accomplished precise drawings and measurements of the vessels by means of Leica DFC295 digital camera and Leica Interactive Measurements software. The study protocol was approved by the Ethics Committee of the Faculty of Medicine.

Four India ink & gelatin injected TGs underwent longitudinal and transverse section in 1 mm thick slabs. These specimens were cleared according to the technique of Spalteholtz, by complete dehydration, and impregnation with benzyl benzoate and methyl salicylate. In that manner becoming transparent, these TGs were analyzed through translumination, i.e. diaphanoscopy. In addition, one TG with a complete dural envelope was dehydrated, embedded in paraplast, sectioned transversely, serially in 4 µm thick slices, and stained both with hematoxilin & eosin and Masson trichrome dye. We microscopically analyzed the cleared specimens and histological slides for better understanding of a TG structure and the position of visible blood vessels filled with India ink or erythrocytes.

Statistical analysis was accomplished using the Statistica 64-bit v. 13 software (TIBCO Software Inc., 2017, Tulsa, USA). The statistical analysis comprised the mean

values and standard deviations of the measured data, and the T-test for independent samples. The probability level of $p < 0.05$ was considered to be significant.

RESULTS

There were two main sources providing arterial branches to the trigeminal ganglion (TG): the internal carotid artery (ICA) and the middle meningeal artery (MMA).

The inferolateral trunk (ILT) arose in all cases (100%) from the lateral surface of the horizontal part of the C3 (intracavernous) segment of ICA. It coursed laterally through the cavernous sinus, above and over the middle segment of the abducens nerve (Fig. 2A). It gave rise to the tentorial artery, ophthalmic, and trigeminal branches. There was a singular trigeminal branch in 60% of the cases and two branches in 40% of the cases (average 1.4 per ganglion). Its diameter ranged from 0.140 to 0.430 mm (mean, 0.260 mm) (Table 1). It extended laterally, along the external convex border of the TG, under the initial segments of V1 and V2 nerves, irrigating the medial and middle parts of the TG.

The meningohypophyseal trunk (MHT) originated from the convexity of the posterior bend of the horizontal portion of the ICA C3 (intracavernous) segment, in all the specimens (100%). It gave off the tentorial artery (the artery of Bernasconi-Cassinari), the inferior hypophyseal, and the dorsal meningeal arteries. The trigeminal branch arose from the tentorial artery, always singular, branching below the roof of the Meckel's trigeminal cave supplying the ophthalmic part of the TG (Fig. 2B). Its average diameter was 0.178 mm, ranging from 0.090 to 0.260 mm (Table 1).

The middle meningeal artery (MMA), branch of the maxillary artery, is an extension of the external carotid artery into the cranial cavity. It entered the skull through the foramen spinosum, postero-laterally to the TG. Our study confirmed that the TG was supplied by the trigeminal branch which originated from the MMA (Fig. 2C). It was present in 23 (92%) cases, singular in all the specimens but one (4%), which had an additional vessel from the same origin. After taking its origin from the MMA, the trigeminal branch coursed medially and slightly anteriorly, and its diameter ranged between 0.11 and 0.34 mm (mean 0.214 mm) (Table 1). The trigeminal branch entered the Meckel's cave to supply the lateral, mandibular part of the TG, and the intracranial part of V3, giving off a small branch for the surrounding dura mater.

We have compared the diameters of trigeminal branches coming from the ILT and MMA by means of Student's T-test for independent samples, as there were cases with two arteries of

a kind, in both groups. The values of ILT trigeminal branches diameters were much larger than the ones of trigeminal branches originating from MMA, the difference being proven as highly significant ($259.7 \pm 55.09 \mu\text{m}$ vs. $213.8 \pm 50.8 \mu\text{m}$; t-value 3.37, df = 60, p = 0.0013).

Additional arterial supply to the TG originated from the dural vascular plexus and the vascular network of the plexal segment of the trigeminal nerve.

Dural arterial network over the surface of the TG gave rise to slender perforating vessels, which entered the tissue of the plexal segment of the trigeminal nerve and the ganglion itself (Fig. 3A). Small vessels, originating from parent arteries that supply the trigeminal nerve, coursed longitudinally along the nerve fascicles, following them into the Meckel's cave. There they formed an arterial network of the plexal trigeminal segment, and entered into the concave surface of the TG. Within the ganglion, the arteries branched into capillary loops surrounding clusters of ganglionic cells (Figs. 3B, 3C).

The whole specimens of the TG injected with India ink & gelatin and cleared by the Spalteholtz technique showed different density of microvessels, mainly capillaries. We found longitudinally oriented vessels in the cisternal segment of the trigeminal nerve, a network of vessels in the plexal segment, and very dense and rich network of the intraganglionic vessels interconnected with anastomotic channels (Fig. 4A). The higher magnification of the intraganglionic capillary plexus exposed vessels coursing along and around the small groups of neurons, and the details showed individual capillaries curving around each of the ganglionic cells (Figs. 4B, 3C).

DISCUSSION

The ganglionic cells have high metabolic demands and are therefore very sensitive to hypoxia. Ischemia from a capillary closure or hypoperfusion is a well-recognized event in diabetic injuries to neurons, resulting in neuronal cells death. The role of nitric oxide has been implied in the pathogenesis of brain injury from hypoxia-ischemia [13]. The literature reports on morphological analyses of the blood supply of the TG and TN are very rare and incomplete, and we still lack a precise picture of the trigeminal vascular pattern [3, 5, 18]. The researchers were mainly focused on the position and syntopy of the TG itself since that knowledge could be implied in a safer surgical approach to the skull base [21]. On the other hand, the interventional neuroradiology requires a precise description of the branching pattern of small arteries arising from the cavernous segment of ICA. This is indispensable for direct embolization in treating meningioma, brain and dural arteriovenous malformations (AVM),

thus minimizing the risk of complications (stroke, cranial nerve palsies, and blindness) [17, 20].

The trigeminal nerve emerges from the lateral portion of middle surface of pons. Its cisternal segment courses through the pontocerebellar cistern, and then it enters the Meckel's cave, now becoming the plexal segment, where the sensory fibers originated from the trigeminal ganglion. Further, the TN divides into three main branches of the TG, known as ophthalmic, maxillary and mandibular divisions [5, 10, 21]. The trigeminal ganglion is typically supplied by the trigeminal branches from two sources: a) from ILT and MHT originating from the ICA, and b) from MMA, originating from the ECA. Trigeminal branches from both carotid systems ramify into small arteries, which enter the ganglion from its medial and lateral borders, from above and below, as well as from the cisternal segment of trigeminal nerve and the branching convex border of the TG [3, 6, 10, 11, 14, 19].

The ILT, as a constant branch of the intracavernous segment of ICA, gave rise to the trigeminal branches in all cases (100%). There was one branch in 60% of the cases, and two in 40%, with an average 1.4 per ganglion. Harris and Rhoton [9] have reported an incidence of 80% of the cases analyzed. In our study, its mean diameter was 0.260 mm, ranging from 0.140 to 0.430 mm. During exploration of the cavernous sinus, we noted the constant trajectory of the ILT crossing over the middle part of the abducens nerve. This topographic feature could be of significance in neurosurgery during operation on the cavernous sinus, in order to facilitate identifying and preserving the ILT, a crucial source of blood supply for intracavernous cranial nerves and TG [10]. Bergmann [3] described a Gasserian twig of the ICA arching along the distal border of the TG. This arch gave off branches to the dura mater, to the three trigeminal divisions, and to the TG. In general, our findings are in line with this description. On the other hand, our results confirmed that the trigeminal branch of the ILT did not reach the lateral part of the TG, except in 2 (8%) cases.

In all the specimens (100%) the MHT participated in the supply of TG's ophthalmic part. Its trigeminal branch had an average diameter of 0.178 mm, ranging from 0.090 to 0.260 mm. The role of this trunk in the TG supply was reported by only a handful of authors [1, 10, 14].

The MMA in 92% of ganglions gave rise to the trigeminal branch for the supply of the lateral, mandibular part of the TG, and the intracranial part of V3, the results similar to literature data from other studies [1, 6, 11, 14]. In our study, the trigeminal branch of the MMA had an average diameter of 0.214 mm, ranging from 0.11 to 0.34 mm. A group of

authors [6] stated that trigeminal branches arose from the petrosal artery in almost 50% of cases, in 34% being doubled. However, according to our previous findings [7], and also this present study, we found a MMA origin of trigeminal branches in all case except for two, with dominantly one artery per ganglion. Two little branches, petrosal and trigeminal, always took their origin in close mutual vicinity, emerging from a large (mean 1.3 mm) MMA [7]. The precise dissection of injected specimens, the one we applied, is necessary for better visualization of slender vessels. There was a short common stem for trigeminal branches, immediately after its origin from the MMA, splitting usually into two twigs for the TG. One can easily misinterpret this finding as two independent arteries from their very beginning.

Our morphometric analysis was the first one to present the diameters of the trigeminal branches. We have measured the calibers of trigeminal branches arising from two different sources, the ILT and the MMA. The statistical comparison between them gave a highly significant difference (t-value 3.37, df = 60, p = 0.0013). Trigeminal branches deriving from the ILT were larger, and destined to supply the medial two thirds of the TG. The lateral third of the TG was vascularized by smaller trigeminal arteries originating from the MMA.

Splitting the maxillary and mandibular divisions of the TG from each other in a transtrigeminal approach (TTA) in order to reach the cavernous sinus still poses a challenge for neurosurgeons [21]. The present anatomic and histologic study confirmed the usefulness of the TTA. Both the mandibular part of the TG and the intracranial part of V3 received supply from the MMA, in contrast to the ophthalmic and maxillary portions of the TG, supplied by the ILT branches. Our results are in accordance with the findings from other studies [6, 11, 14, 19].

The rich dural vascular plexus over the TG was composed of vessels coming from dural twigs of ILT, MHT and MMA. Fine perforating arteries left the dural arterial network and supplied the plexal segment of the TN and the TG itself (and vice versa). There are two neuropeptides from the activated dural sensory fibers of the TG neurons: substance P (SP), neurotransmitter of nociceptive and maybe of the other sensory modalities impulses, and the calcitonin gene-related peptide (CGRP), both promoting vasodilatation and plasma extravasations [4, 15]. They induce the release of sensitizing inflammatory mediators (histamine, 5HT, nitric oxide, heparin) from the present mast cells, i.e. the mast cell degranulation. This activation of the trigeminovascular system may play a role during migraine, resulting in headache [4]. The vasculature of the TG and surrounding dura likely

contributes to the migraine symptoms, but further research is required in order to explore the microcirculatory density in parts of the TG and the distribution of mastocytes.

The plexal segment of the TN occupies the Meckel's dural cave. It received longitudinal twigs which followed the fascicules of the cisternal segment of TN, inside the arachnoid sleeve. The trigeminal arteries, which may originate from the basilar artery (BA) and its side branches, supply the TN: the superolateral and inferolateral pontine arteries (i.e., the two long circumferential pontine arteries), the anterior inferior cerebellar artery (AICA), and the superior cerebellar artery (SCA) [5, 21]. The smaller vessels showed a general course parallel to the fascicules, as already described [3]. Plexiform constitution of the crossing rootlets of trigeminal fascicules' plexal segment changed the appearance of the arterial plexus, from parallel to mesh-like.

Very dense and rich capillary network of the intraganglionic vessels, interconnected with anastomotic channels, showed a uniform density of microvessels (a so-called microcirculatory bed) throughout the TG. According to another study, the mandibular division of the fetal TG had the highest, and maxillary division showed the lowest presence of microcirculatory bed components [1]. It should be noted that Bergmann [3] observed frequent ampulliform dilatations on capillaries and small vessels. Our careful examination of the whole TG specimen injected with India ink & gelatin, and cleared by the Spalteholtz technique, presented a compact area composed of highly tortuous small vessels, coiled around the ganglionic cells. These vessels gave the appearance of spherical or pear-shaped ampullae at the point of their course deflection. It was obvious that no real capillary dilatations existed in the straight parts, but depending on the angle of view, the tortuosities of capillaries appeared enlarged just because of the superposition of their portions.

Limitations of the study and perspectives

The main drawback of the present study is that only a part of the arterial network of TG can be identified using Spalteholtz technique. Further, the limitation of our study could be the sample size, i.e. the number of the available heads, especially fetal, in order to establish precise definitions on the variations of arterial supply of the trigeminal ganglion. On the other hand, to our knowledge, there are no such studies on the TG microcirculatory component quantification in the adult ganglions. The goal of our future study should be to quantify the elements of TG microcirculation using the immunohistochemical staining method against an endothelial marker, with making particular effort to compare the distribution of microcirculatory elements between three parts of the TG.

CONCLUSIONS

The present study provides evidence for variations in the arterial supply of the TG, with the dominance of ILT from the ICA, and, to a smaller degree, the participation of the MMA from the ECA. The arterial contribution from the dural covering of the TG, and from the TN itself are additional components which form the circulatory bed of the TG. The observed characteristics of the TG vasculature could be implied in a safer surgical approach to the skull base, with consideration for the surrounding structures, and for better understanding of close relationships between the trigeminal ganglionic cells, dura mater and corresponding blood vessels.

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Table 1. Parent arteries and their trigeminal branches for the supply of TG

Artery	Incidence (%)	Number of trigeminal branches (M)	Diameter (μm); Range (Mean \pm SD)
Inferolateral trunk	25 (100)	1 – 2 (1.4)	140 - 430 (260 \pm 55.09)
Meningohypophyseal trunk	25 (100)	1	90 - 260 (178 \pm 40.42)
Middle meningeal artery	23 (92)	1 – 2 (1.04)	110 - 340 (214 \pm 50.84)
Total	25 (100)	3 – 5 (3.34)	90 - 430 (222 \pm 60.17)

FIGURE LEGENDS

Figure 1. Micromorphological characteristics of the trigeminal ganglion (TG). A. Dissection of the central area of the skull base showing parts of the trigeminal nerv after removal of the brain: left and right cisternal (1, 1'), plexal (2), trigeminal ganglion (3), ophthalmic nerve (4), maxillary nerve (5), mandibular neve (6); (7) dura mater (removed on the left side); (8) middle meningeal artery; (9) basilar artery; (10, 10') left and right internal carotid arteries; (11, 11') left and right anterior cerebral arteries; (12, 12') left and right middle cerebral arteries. B. Coronal section of the TG demonstrates meningeal dural roof (1), and periosteal floor (2) of Meckel's trigeminal cave, containg in a plate arranged clusters of ganglionic cells (3), and numerous nerv fascicules

(arrows) (Trichrome Masson). C. The higher magnification of coronal section of the TG with the dural covering (1), plate of ganglionic cells (2), and a nerv fascicle (arrows). Note the larger blood vessels with red cells (Trichrome Masson staining).

Figure 2. The main trigeminal arteries. A. The ILT (1) courses laterally crossing over the abducens nerve (2), and gives off the ophthalmic (3) and the trigeminal branch (4) running bellow the right TG (5); (6) oculomotor nerve (the dura of the lateral wall of the cavernous sinus has been removed). B. The MHT (1) branches into the tentorial artery and the trigeminal branch (2) for the vascularization of TG and dura (removed) (3) over the right TG (4); (5) right trigeminal nerve (the lateral dural wall of the cavernous sinus has been partially removed). C. Trigeminal branch (1) from the middle meningeal artery (2) approaches to the mandibular part of TG (3); (4) right trigeminal nerve.

Figure 3. Contributing trigeminal arteries for the TG. A. Perforating branches (arrows) coming from the dural covering (1) (elevated) enter the right TG (2); (3) right trigeminal nerve. B. Branches from the arterial network (arrows) of the terminal, plexal segment of trigeminal nerve (1); (2) right trigeminal ganglion. C. Arterial network (arrows) of the TG (arrows) with capillary loops surrounding ganglionic cells; (1) plexal segment of trigeminal nerve.

Figure 4. The whole cleared specimen injected with India ink and gelatin. A. Capillary network of the plexal segment (2) of the trigeminal nerve (1), and the TG (3). B. Capillary plexus of the TG (detail). C. The higher magnification of capillary plexus (detail).







