Translational Research in Anatomy 37 (2024) 100321

Contents lists available at ScienceDirect

Translational Research in Anatomy

journal homepage: www.elsevier.com/locate/tria

Brain lymphatic vessels accompanying meningeal arteries in the human

Safiye Çavdar [a,*], Damlasu Altınöz [a], Tevriz Dilan Demir [b], Acar Bayraktaroğlu [a], İlke Ali Gürses [a], Gülnihal Özcan [b,c]

[a] Department of Anatomy, Koç University, School of Medicine, Rumelifener Yolu, Istanbul, Turkey
[b] Koç University Research Center for Translational Medicine (KUTTAM), Rumelifener Yolu, Istanbul, Turkey
[c] Department of Medical Pharmacology, Koç University, School of Medicine, Rumelifener Yolu, Istanbul, Turkey

ARTICLE INFO

Keywords:
Anterior meningeal artery
Middle meningeal artery
Posterior meningeal artery
Dura mater
Meningeal lymphatic vessels

ABSTRACT

Introduction: Recent studies showed the connection between cerebrospinal fluid (CSF) and deep cervical lymph nodes (dCLN) via lymphatic vessels (LV)s. These LVs accompany dural sinuses, cranial nerves, and arteries. The present study aimed to show in addition to the main trunk of the middle meningeal artery (MMA) whether the anterior and posterior branches of MMA; the anterior (AMA) and posterior (PMA) meningeal arteries also have a role in brain lymphatic drainage in humans.

Materials and method: Samples of the middle meningeal artery (MMA) and its two main branches (anterior and posterior), anterior and posterior meningeal arteries (AMA and PMA) were obtained from 3 cadavers and 6 autopsies. Podoplanin (PDPN) (lymphatic endothelial marker) and CD31 (vascular endothelial marker) immunohistochemistry (IHC) were applied to 5 μm thick transverse sections and counterstained with hematoxylin. Furthermore, western blotting techniques were applied to the same tissue samples in 3 autopsy cases to demonstrate the presence of PDPN.

Results: We observed the presence of the LVs along the main trunk of MMA and its two branches, AMA, and PMA. The LVs along the MMA (main trunk) had a larger diameter range than its branches and the AMA and PMA. The diameter of LVs of the PMA was larger than AMA. Furthermore, fluid channels with varying diameters and densities were closely localized to the LVs. The western blot PDPN protein expression confirmed IHC results.

Conclusion: The LVs along the main trunk, anterior and posterior branches of the MMA, AMA, and PMA also provide an extracranial pathway for thought drainage of waste from the brain tissue in humans. Thus, the organization of the extracranial transport of LVs of the brain should be well-defined for understanding the role of lymphatic drainage in various neurological diseases.

1. Introduction

The central nervous system (CNS) was considered to lack lymphatic vasculature and knowledge of macromolecule clearance from the interstitial tissue of the brain was unknown. Recent studies demonstrated that the cerebrospinal fluid (CSF) drains into the deep cervical lymph nodes (dCLN); however, the mechanism of CSF draining into the dCLN is still not described in detail [1–3]. Further, studies have described connections between arachnoid granulations and venous circulation via intradural vascular channels using lymphatic markers in formalin-fixed human cadaver [4]. Recent discoveries showed the presence of glia-lymphatic (glymphatic) and meningeal lymphatic systems [5–7].

Dysfunction of either the glymphatic or meningeal lymphatic system may impair pathological substance clearance and result in various neurological conditions [6,8–10]. Thus, a detailed understanding of how lymphatic clearance in CNS occurs is necessary to reveal the mechanisms of various neurological diseases and for the development of effective therapeutic strategies.

Three major arteries contribute to the arterial supply of the dura: middle meningeal artery (MMA), anterior meningeal (AMA), and posterior meningeal (PMA). The major arterial supply of the dura is via

Abbreviations: AD, Alzheimer's disease; AMA, anterior meningeal artery; BV, blood vessel; CNS, central nervous system; CSF, cerebrospinal fluid; dCLN, deep cervical lymph nodes; LV, lymphatic vessel; mLV, meningeal lymph vessels; MMA, middle meningeal artery; IHC, immunohistochemistry; PBS, phosphate-buffered saline; PDPN, podoplanin; PMA, posterior meningeal artery.

* Corresponding author. Koç University, School of Medicine, Department of Anatomy, Rumelifeneri, 34450, Sarıyer, Istanbul, Turkey.
E-mail address: scavdar@ku.edu.tr (S. Çavdar).

https://doi.org/10.1016/j.tria.2024.100321
Received 3 May 2024; Received in revised form 4 July 2024; Accepted 7 July 2024
Available online 9 July 2024
2214-854X/© 2024 The Authors. Published by Elsevier GmbH. This is an open access article under the CC BY-NC license (http://creativecommons.org/licenses/by-nc/4.0/).

MMA. It supplies the superolateral portion of the dura. The MMA is a branch of the maxillary artery and enters the skull through the foramen spinosum or in some rare cases through the petrosphenoid fissure [11]. The MMA apart from oxygenation, of the dura also has a function of thermoregulation of the brain. The PMA, also termed "the artery of the falx cerebelli", usually arises from the ascending pharyngeal or the occipital arteries [12,13] and provides blood supply to the paramedial and medial portions of the dura covering the cerebellar convexity and the falx cerebelli [14,15]. The PMA can enter the skull through the jugular foramen, foramen magnum, or the hypoglossal canal. The AMA is a branch from the anterior and/or posterior ethmoidal arteries which are branches of the ophthalmic artery. They pass through the anterior and posterior ethmoidal foramen, reach the anterior cranial fossa, and supply the anterior and inferior aspects of the dura.

It has been shown that lymphatic vessels (LV)s accompanying dural sinuses have a role in the extracranial transport of brain lymphatics [2,16,17]. In addition to these LVs, studies have shown the presence of LVs accompanying the main trunk of the MMA using lymphatic vessel endothelial markers [18,19]. The current study aims to show, whether the LVs are also present along the anterior and posterior branches of the MMA, the AMA, and the PMA using lymphatic endothelial marker podoplanin (PDPN) and vascular endothelial marker CD31 immunohistochemistry (IHC). The diameter of the LVs accompanying the meningeal arteries was measured to show the efficiency of each vessel in the extracranial transport of the vessels in the human brain. Furthermore, the western blotting technique will be used to support the PDPN expression in the same tissue sample.

2. Materials and Method

A total of 3 cadavers (1 male and 2 females; age range was 81–83) and 6 autopsy (5 males, and 1 female; age range was 30–58) cases were used in the present study. The tissue samples from autopsy cases were obtained within less than 24 h and none of the cases had a neurological disease. The calvarium was removed by routine autopsy dissection. The dura was detached from its bony attachments and was elevated with care. From the base of the skull, the 0.5 cm^2 samples were obtained from the main trunk and branches (anterior and posterior) of MMA, AMA, and PMA for PDPN and CD31 IHC and western blotting techniques. The western blotting and IHC techniques support anatomical research, they enable the identification/localization of specific proteins within tissues [20]. The samples from the main trunk of the MMA were obtained as the arteries emerged from the foramen spinosum. The anterior and posterior branches of the MMA were harvested 1 cm distal to the bifurcation of the MMA. The PMA samples were obtained from their intracranial segments as they ascended lateral to the internal occipital crest, 2 cm posterior to the edge of the foramen magnum. The AMA samples were obtained at the anterior cranial fossa as the arteries emerged from the anterior ethmoidal foramen (Fig. 1).

2.1. The PDPN and CD31 immunohistochemistry

The tissues from 3 cadavers (age range 81–83, one male and two female) and 3 autopsies (age range 42–58, all male) were fixed with immersion in 10 % neutral buffered formalin for 12h. The consecutive sections were incubated overnight at 4 °C with the monoclonal primary antibodies; either rabbit anti-podoplanin (Sigma Aldrich, P5374, Germany, 1:1000) or rabbit anti-CD31 (NB100-2284, Novus Biologicals, San Diego, CA, USA 1:100) in 2 % bovine serum albumin in phosphate-buffered saline. The methodology used in this study was adapted from our previous publication [21] where a similar approach was applied. Lymphatic vessels and fluid channels were evaluated from each sample and dimension measurements were made using ZEN software. The largest and the smallest diameters and areas of lymphatic vessels and fluid channels were measured from each sample using ZEN Blue software, and the average was calculated for each case.

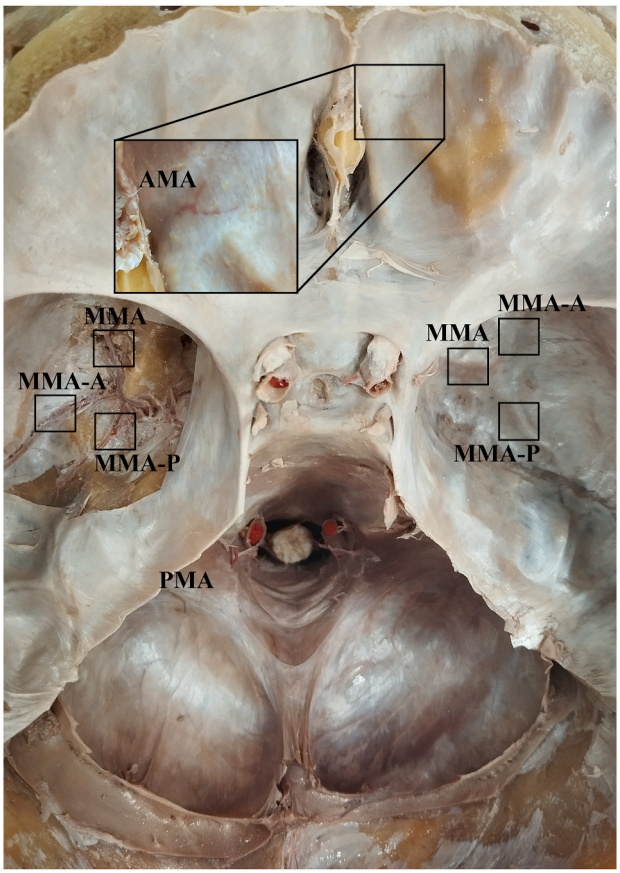


Fig. 1. A representative photograph of the skull base of a formalin-fixed cadaver, indicating the regions where samples of each meningeal artery were collected for immunohistochemistry and western blotting. To enhance the visibility of arteries in the photograph, silicon injection with red dye was done via the internal carotid and vertebral arteries. On the left side of the skull, the meningeal layer of the dura covering the middle meningeal artery and its branches was removed to reveal the course of the arteries. (AMA: anterior meningeal artery, MMA-A: anterior branch of the middle meningeal artery, MMA-P: posterior branch of the middle meningeal artery, PDPN: podoplanin, PMA: posterior meningeal artery). (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

2.2. The PDPN and CD31 western blotting technique

The dura specimens from meningeal arteries of 3 autopsies (age ranged between 30 and 58, two male and one female) were obtained for western blotting. The membranes were incubated with the primary antibodies, including anti-podoplanin (Sigma Aldrich, P5374, Germany, 1:1000) and anti-CD31 (NB100-2284, Novus Biologicals, San Diego, CA, USA 1:1000). The following steps and details of the methodology can be found in Ref. [21]. Furthermore, the quantification of bands was performed by densitometry analysis using Image J, and Podoplanin expression in samples was normalized to CD31 intensity.

3. Results

3.1. The immunohistochemistry findings

Consecutive meningeal artery sections were immune stained with CD31 and PDPN to distinguish blood vessels (BV)s from LVs (Fig. 2A and B). The presence of PDPN-positive LVs along the main trunk of MMA and its anterior and posterior branches, AMA, and PMA were observed (Fig. 3A, B, C, D, E). The diameter range of the LVs accompanying the

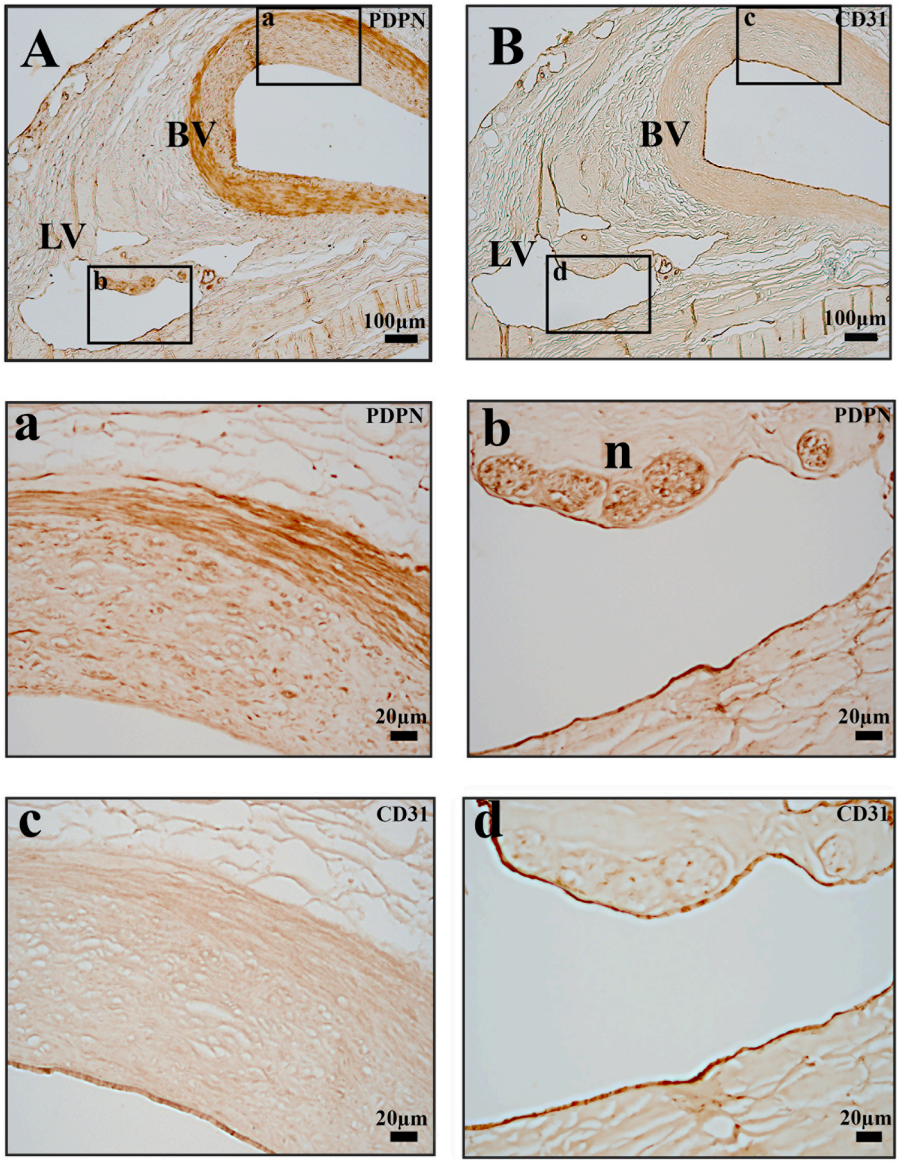


Fig. 2A, B. 2A. A representative photomicrograph of PDPN stained samples obtained from the anterior branch of the MMA. **2B.** Representative photomicrograph of CD31 stained samples obtained from the anterior branch of the MMA. **a.** High magnification of Fig. 2A showing PDPN negative endothelium of BV **b.** High magnification of Fig. 2A showing PDPN positive endothelium of LV **c.** High magnification of Fig. 2B showing CD31 positive endothelium of BV. **d.** High magnification of Fig. 2B showing CD31 positive endothelium of LV. (BV: blood vessel, LV: lymphatic vessel, n: nerve).

main trunk, and the anterior and posterior branches of the MMA were measured as 46.1–1166 µm, 43–572.3 µm, and 33.3–170 µm respectively. Furthermore, the diameter range of the LVs along the AMA was 22.4–188.7 µm and the diameter range of the LVs accompanying the PMA was 43.2–726.9µmn. The average diameter of the LVs accompanying the main trunk of MMA (530.2 ± 359.4) was the largest. The average diameter of the LVs along the anterior branch (272.5 ± 222.2) of the MMA was larger than the posterior branch (87.9 ± 57.9). Furthermore, the PMA (263.2 ± 241.1) had a larger diameter range than the AMA (110.8 ± 60.9) (Table 1). Moreover, the area range of the LVs accompanying the main trunk, and the anterior and posterior branches of the MMA were measured as 1,3–24062 mm^2; 3,7–8,0 mm^2; and 0,9-6,1 mm^2 respectively. Furthermore, the area range of the LVs along the AMA was 0,4–33,8 mm^2 and the range of area of the LVs accompanying the PMA was 1,9–8862,2 mm^2. The average area of the LVs accompanying the main trunk of MMA (6108,9 ± 9310 mm^2) was the largest. The average area of the LVs along the anterior branch (5,9 ± 3,0 mm^2) of the MMA was larger than the posterior branch (3,0 ± 1,8 mm^2). Furthermore, the PMA (2143,5 ± 3814,8 mm^2) had a larger area range than the AMA (7,3 ± 0,8 mm^2) (Table 1).

Fluid channels of varying diameters and densities were closely localized to the lymphatic and vascular structures. While the fluid channels did not show PDPN immunoreactivity (Fig. 4A), they were found to lack an endothelium lining (Fig. 4B). The diameter range of the fluid channels closely localized to the main trunk and its anterior and posterior branches, AMA, and PMA was 22.9–80 µm, 15.8–54 µm, 15.2–59.4 µm, 10.7–67 µm and 16.7–77.6 µm respectively. Furthermore, the average diameter of fluid channels closely located to the main trunk and its anterior and posterior branches, AMA and PMA was measured as 54.6 ± 20.1, 31.7 ± 16.2, 49.2 ± 19, 26.8 ± 16.7, 51.5 ± 27.9 respectively. Furthermore, the area range of the fluid channels closely localized to the main trunk and its anterior and posterior branches, AMA, and PMA was 0,9–130,3 mm^2; 0,2-0,9 mm^2; 0,4-1,9 mm^2; 0,2-0,5 mm^2 and 0,4-1,5 mm^2 respectively. Also, the average area of fluid channels closely located to the main trunk and its anterior and posterior branches, AMA and PMA was measured as 39,3 ± 55,5 mm^2;

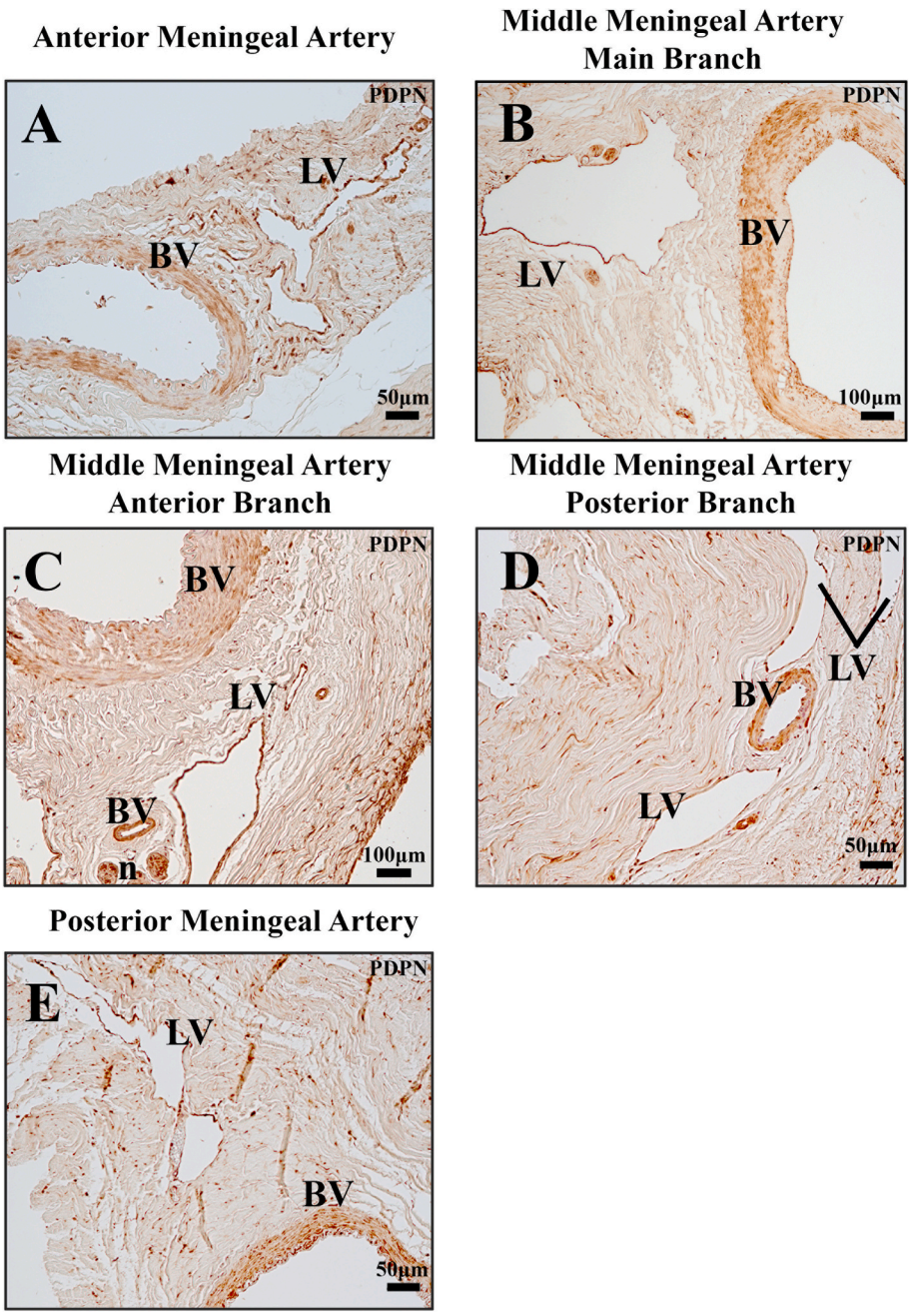


Fig. 3A, B, C, D, E. Photomicrographs of all meningeal LV samples (AMA, MMA and its anterior and posterior branches, and PMA respectively) demonstrate positive endothelial staining with PDPN. (BV: blood vessel, LV: lymphatic vessel, n: nerve).

Table 1
Range of diameters and areas of the LVs and fluid channels accompanying the main trunk, anterior and posterior branches of the middle meningeal artery; anterior meningeal artery, and posterior meningeal artery.

	Diameter lymphatic vessels (µm)		Area of lymphatic vessels (mm^2)		Diameter range of fluid channels (µm)		Area of fluid channels (mm^2)	
	Range	Mean (SD)	Range	Mean (SD)	Range	Mean (SD)	Range	Mean (SD)
MMA Main Branch	46.1–1166	530.2 ± 359.4	1.3–24062	6108.9 ± 9310	22.9–80	54.6 ± 20.1	0.9–130.3	39.3 ± 55.5
MMA Anterior Branch	43–572.3	272.5 ± 222.2	3.7–8.0	5.9 ± 3.0	15.8–54	31.7 ± 16.2	0.2 - 0.9	0.5 ± 0.2
MMA Posterior Branch	33.3–170	87.9 ± 57.9	0.9 - 6.1	3.0 ± 1.8	15.2–59.4	49.2 ± 19	0.4 - 1.9	1.2 ± 0.8
AMA	22.4–188.7	110.8 ± 60.9	0.4–33.8	7.3 ± 0.8	10.7–67	26.8 ± 16.7	0.2 - 0.5	0.6 ± 0.2
PMA	43.2–726.9	263.2 ± 241.1	1.9–8862.2	2143.5 ± 3814.8	16.7–77.6	51.5 ± 27.9	0.4 - 1.5	1.8 ± 0.5

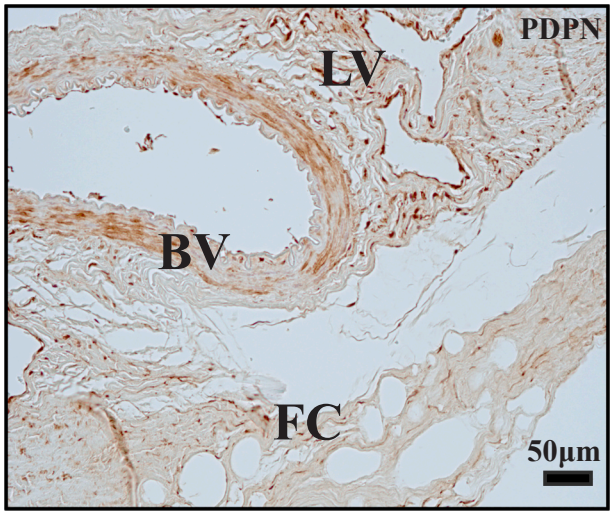


Fig. 4A, B. **4A** A representative PDPN staining section showing fluid channels closely localized to lymphatic and vascular structures with varying diameters and density. **4B:** Consecutive hematoxylin and eosin stained section showing fluid channels with no endothelial cell lining (BV: blood vessel, FC: fluid channels, LV: lymphatic vessel).

0,5 ± 0,2 mm^2; 1,2 ± 0,8 mm^2; 0,6 ± 0,2 mm^2; and 1,8 ± 0 mm^2 respectively.

3.2. *The western blotting findings*

The PDPN western blotting results were verified by vascular endothelial marker CD31. The protein expression of PDPN in the anterior branch of the MMA was greater than in the main trunk and posterior branch of the MMA. The PDPN protein expression was higher in the PMA compared to the AMA. Thus, IHC results were supported by the western blotting technique (Fig. 5A and B).

4. Discussion

The current IHC results of PDPN and CD31 demonstrated the presence of LVs accompanying the main trunk, anterior and posterior branches of the MMA, AMA, and PMA. The average diameter of the LVs along the main trunk of MMA was larger than AMA and PMA. The average diameter of the LVs along the anterior branch was larger than the posterior branch of the MMA. The average diameter of the LVs along the PMA was larger than the AMA. The western blotting highly supported our IHC results. Furthermore, fluid channels were present and closely localized to the lymphatic and vascular structure along the main trunk, and anterior and posterior branches of the MMA, AMA, and PMA.

The removal of waste products from the brain's interstitial spaces was a mystery until recent discoveries. Iliff et al. (2012), were pioneers who injected fluorescent tracers into the subarachnoid space of the rat and using vivo two-photon imaging, they showed that CSF enters the parenchyma along paravascular spaces surrounding the penetrating arteries and the brain interstitial fluid is cleared along paravenous drainage pathways. Furthermore, Louveau et al. (2015) discovered LVs accompanying the dural sinuses termed "meningeal lymph vessels" (mLV) which carry both fluid, immune cells, and macromolecules from the CSF to the dCLN. Studies mainly focused on the mLV that accompanied the superior and transverse dural sinus. However, recent studies have shown the presence of LVs not only accompanying the dorsal but also basal dural sinuses [17,22,23]. Some studies described LVs along

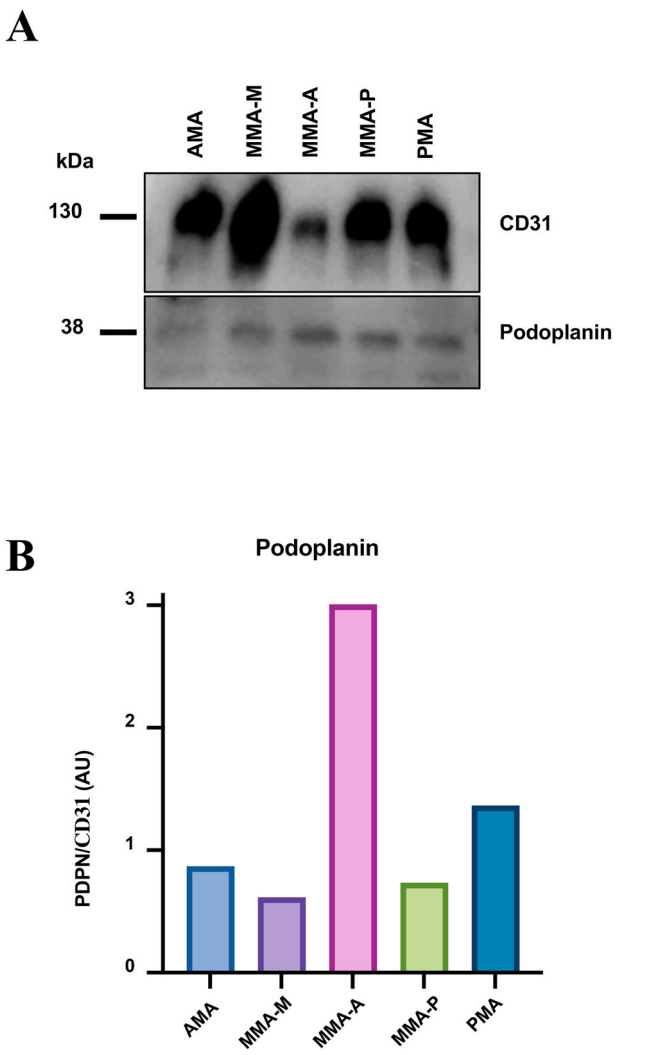


Fig. 5A, B. **5A** Investigation of PDPN expression by western blot in meningeal arteries.
5B, densitometric analysis of PDPN expression in meningeal arteries (AMA: anterior meningeal artery, MMA-A: anterior branch of the middle meningeal artery, MMA-P: posterior branch of the middle meningeal artery, PDPN: podoplanin, PMA: posterior meningeal artery).

cranial nerves and BVs which provided a pathway for the drainage of waste from the cranium [24–27].

Aspelund et al. (2015a), studied the brain lymphatic system on a transgenic model (complete aplasia of the dural lymphatic vessels) and control mice and showed that the LVs were present along the main trunk of MMA of control mice; however, were absent in the transgenic mice. Ahn et al. (2019) infused the macromolecular tracer Quantum Dot 705 into the cisterna magna or brain parenchyma of Prospero homeobox protein 1 (lymphatic endothelial marker) - Green fluorescent protein mice and traced the signal under a high-resolution fluorescence stereomicroscope showed the filling of the main trunk of the MMA. The present study showed that in addition to the well-known LV accompanying the dural sinuses, LVs along the meningeal arteries may also provide an extracranial pathway for the drainage of waste from the brain tissue in humans. The results of the current study showed that in addition to the presence of LVs along the main trunk of the MMA, LVs were also present along the anterior and posterior branches of the MMA, AMA, and PMA. Thus all 3 meningeal arteries have a role in the extracranial transport of waste from the brain. The larger diameter of the LVs along the main trunk of the MMA and the anterior branch of the MMA may indicate more effective drainage contributions compared to the

AMA and PMA. Furthermore, the diameter of the PMA is greater than AMA thus, the PMA can be more effective compared to the AMA.

It has been shown that the solutes in the LVs travel in a direction opposite to blood flow toward larger and more proximal vessels [26,28]. The present study confirms the results of the former studies; thus, we measured the large diameter range of the main trunk of the MMA compared to its anterior and posterior branches. The LVs along the MMA, AMA, and PMA can exit the skull through the foramen spinosum, anterior ethmoidal foramen, and foramen lacerum respectively, and reach the dCLN.

The amount of contribution of the LVs accompanying dural sinuses, cranial nerve, and arteries in the clearance of the lymphatic solutes is not yet well known. Louveau et al. (2018) investigated the potential entry routes of the CSF into the mLVs, labeled anti-lymphatic vessel endothelial hyaluronan receptor 1 antibodies were injected into the CSF and followed through mLV. The LVs along the MMA were not labeled by the injected antibodies. They suggested that these vessels may be draining the dura but not the CSF. Thus, there are numerous questions yet to be answered for example, which LVs are more efficient compared to the others, whether the disfunction of one of the routes may result in specific neurological diseases, and does the efficiency of each route varies between species, etc.

Fluid channels were referred to as "juice channels" in early anatomical research which also hypothesized that they would be involved in fluid transport and/or resorption [29]. Human parasagittal dura was the subject of a recent 3D micro-CT investigation, which revealed linkages between the fluid channels and suggested that the dural channels might act as a reservoir for CSF drainage [30]. Furthermore, a study demonstrated a larger fluid channel in the elderly compared to younger humans [31]. The large fluid channels observed in the elderly can be related to less turnover and drainage of CSF. Mack et al. (2009) also suggested that these structures may participate in CSF movement and/or clearance. Furthermore, Johnson (2021) evaluated fluid channels in 113 dura (peri-superior sagittal sinus dura) of Alzheimer's (AD) and non-AD patients and found that non-AD dura had numerous fluid channels and dura with AD had significantly fewer channels. The current study shows the presence of fluid channels of varying diameter and density, closely localized to the LVs along the dura accompanying the meningeal arteries, which may participate in the CSF circulation and the clearance of waste. These fluid channels can be depositing areas before the drainage. The different sizes of the fluid channels may be related to age and/or functional differences [32].

The specificity of the BV endothelial marker CD31 antibody can be regarded as the limitation of the present study. Thus, CD31 labeled not only LV but also BV endothelial cells (Fig. 2A and B). However, PDPN labeled only LV endothelial cells Thus vessels both labeled with CD31 and PDPN were classified as LV, and vessels labeled only with CD31 were classified as BV (Fig. 2A and B). Similar studies using CD31 in mice [33–35] and in humans [16] also showed the overlapping labeling of BV and LV. Furthermore, in addition to the LV endothelium PDPN also stained fibroblasts, perineurium, and Schwann cells. Therefore, careful observation is necessary to distinguish the two-vessel type. Thus, this issue should be kept in mind during the interpretation of the western blotting results. Further, the IHC sample size ($n = 6$) of the present study can be regarded as another limitation.

The current study provides evidence for LV accompanying the branches (anterior and posterior) of the MMA, AMA, and PMA which has not been reported previously. Furthermore, it provides diameter measurement which may show the role and the efficiency of each LV along the meningeal arteries. The LVs accompanying the meningeal arteries may have a supporting and/or compensatory function to the mLVs in preventing the accumulation of proteins that may result in various neurological diseases. Thus, understanding the organization of the extracranial transport of LVs of the brain is necessary to reveal the mechanism of lymphatic system-related diseases. Further, during surgery of the region care should be taken not to disturb the lymphatic circulation.

Ethical Statement

The approval of the Istanbul Forensic Medicine (ATK) Research Ethical Committee number: 21589509/2023/111 was obtained for this study.

Funding Statement

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

CRediT authorship contribution statement

Safiye Çavdar: Writing – original draft, Methodology, Investigation, Conceptualization. **Damlasu Altınöz:** Writing – review & editing, Visualization, Methodology, Investigation, Data curation. **Tevriz Dilan Demir:** Visualization, Methodology, Formal analysis, Data curation. **Acar Bayraktaroğlu:** Investigation, Data curation. **İlke Ali Gürses:** Investigation. **Gülnihal Özcan:** Writing – review & editing.

Declaration of Competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

The authors gratefully acknowledge the use of the services and facilities of the Koç University Research Center for Translational Medicine (KUTTAM), funded by the Presidency of Turkey, Head of Strategy and Budget. Graphical abstract was designed using Biorender (Agreement number: MT26WC3AMF).

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.tria.2024.100321.

References

[1] M. Absinta, S.K. Ha, G. Nair, P. Sati, N.J. Luciano, M. Palisoc, A. Louveau, K. A. Zaghloul, S. Pittaluga, J. Kipnis, D.S. Reich, Human and nonhuman primate meninges harbor lymphatic vessels that can be visualized noninvasively by MRI, Elife 6 (2017), https://doi.org/10.7554/elife.29738.001.

[2] G. Ringstad, P.K. Eide, Cerebrospinal fluid tracer efflux to parasagittal dura in humans, Nat. Commun. 11 (2020) 1–9, https://doi.org/10.1038/s41467-019-14195-x.

[3] K. Yağmurlu, J.D. Sokolowski, M. Çırak, K. Urgun, S. Soldozy, M. Mut, M. E. Shaffrey, P. Tvrdik, M.Y.S. Kalani, Anatomical features of the deep cervical lymphatic system and intrajugular lymphatic vessels in humans, Brain Sci. 10 (2020) 1–12, https://doi.org/10.3390/BRAINSCI10120953.

[4] K. Yağmurlu, J. Sokolowski, S. Soldozy, P. Norat, M. Çirak, P. Tvrdik, M. E. Shaffrey, M.Y.S. Kalani, A subset of arachnoid granulations in humans drain to the venous circulation via intradural lymphatic vascular channels, J. Neurosurg. 136 (2021) 917–926, https://doi.org/10.3171/2021.2.JNS204455.

[5] J.J. Iliff, M. Wang, Y. Liao, B.A. Plogg, W. Peng, G.A. Gundersen, H. Benveniste, G. E. Vates, R. Deane, S.A. Goldman, E.A. Nagelhus, M. Nedergaard, A paravascular pathway facilitates CSF flow through the brain parenchyma and the clearance of interstitial solutes, including amyloid β, Sci. Transl. Med. 4 (2012) https://doi.org/10.1126/scitranslmed.3003748.

[6] S. Da Mesquita, A. Louveau, A. Vaccari, I. Smirnov, R.C. Cornelison, K. M. Kingsmore, C. Contarino, S. Onengut-Gumuscu, E. Farber, D. Raper, K.E. Viar, R.D. Powell, W. Baker, N. Dabhi, R. Bai, R. Cao, S. Hu, S.S. Rich, J.M. Munson, M. B. Lopes, C.C. Overall, S.T. Acton, J. Kipnis, Functional aspects of meningeal lymphatics in ageing and Alzheimer's disease, Nature 560 (2018) 185–191, https://doi.org/10.1038/s41586-018-0368-8.

[7] L.M. Hablitz, M. Nedergaard, The glymphatic system: a novel component of fundamental neurobiology, J. Neurosci. 41 (2021) 7698–7711, https://doi.org/10.1523/jneurosci.0619-21.2021.

[8] A. Scott-Massey, M.K. Boag, A. Magnier, D.P.C.F. Bispo, T.K. Khoo, D.L. Pountney, Glymphatic system dysfunction and sleep disturbance may contribute to the pathogenesis and progression of Parkinson's disease, Int. J. Mol. Sci. 23 (2022) 12928, https://doi.org/10.3390/ijms232112928.
[9] A.F.A. Mentis, E. Dardiotis, G.P. Chrousos, Apolipoprotein E4 and meningeal lymphatics in Alzheimer disease: a conceptual framework, Mol. Psychiatr. 26 (2020) 1075–1097, https://doi.org/10.1038/s41380-020-0731-7, 2020 26:4.
[10] S. Chen, L. Shao, L. Ma, Cerebral edema formation after stroke: emphasis on blood–brain barrier and the lymphatic drainage system of the brain, Front. Cell. Neurosci. 15 (2021) 716825, https://doi.org/10.3389/fncel.2021.716825.
[11] E. Bruner, S. Sherkat, The middle meningeal artery: from clinics to fossils, Child's Nerv. Syst. 24 (2008) 1289–1298, https://doi.org/10.1007/s00381-008-0685-6.
[12] T. Greitz, T. Laurén, Anterior meningeal branch of the vertebral artery, Acta Radiol. Diagn. 7 (1968) 219–224, https://doi.org/10.1177/028418516800700303.
[13] T.H. Newton, The Anterior and Posterior Meningeal Branches of the Vertebral Artery1, 10.1148/91.2.271 91, 1968, https://doi.org/10.1148/91.2.271, 271–279.
[14] A.L. Rhoton, The cerebrum. Anatomy, Neurosurgery 61 (2007) 37–118, https://doi.org/10.1227/01.neu.0000255490.88321.ce. ; discussion 118.
[15] P. Lasjaunias, A. Berenstein, K.G. ter Brugge, Clinical Vascular Anatomy and Variations, Springer Berlin Heidelberg, 2001, https://doi.org/10.1007/978-3-662-10172-8.
[16] M. Absinta, S.K. Ha, G. Nair, P. Sati, N.J. Luciano, M. Palisoc, A. Louveau, K. A. Zaghloul, S. Pittaluga, J. Kipnis, D.S. Reich, Human and nonhuman primate meninges harbor lymphatic vessels that can be visualized noninvasively by MRI, Elife 6 (2017), https://doi.org/10.7554/elife.29738.001.
[17] S. Çavdar, B. Köse, D. Altınöz, G. Söyler, A. Cingöz, İ.A. Gürses, M. Özkan, H. Aslıyüksek, H. Çakır, Lymphatic vessels accompanying dorsal and basal dural sinuses in the human brain, J. Chem. Neuroanat. 134 (2023) 102357, https://doi.org/10.1016/j.jchemneu.2023.102357.
[18] A. Aspelund, S. Antila, S.T. Proulx, T.V. Karlsen, S. Karaman, M. Detmar, H. Wiig, K. Alitalo, A dural lymphatic vascular system that drains brain interstitial fluid and macromolecules, J. Exp. Med. 212 (2015) 991–999, https://doi.org/10.1084/JEM.20142290/VIDEO-1.
[19] A. Louveau, I. Smirnov, T.J. Keyes, J.D. Eccles, S.J. Rouhani, J.D. Peske, N. C. Derecki, D. Castle, J.W. Mandell, K.S. Lee, T.H. Harris, J. Kipnis, Structural and functional features of central nervous system lymphatic vessels, Nature 523 (2015) 337–341, https://doi.org/10.1038/nature14432.
[20] M. Golberg, G. Wysiadecki, J. Kobos, P. Brzeziński, M. Polguj, E. Clarke, K. Barszcz, K. Balawender, M. Radek, A. Żytkowski, Application of automated immunohistochemistry in anatomical research: a brief review of the method, Translational Research in Anatomy 28 (2022) 100211, https://doi.org/10.1016/J.TRIA.2022.100211.
[21] S. Çavdar, D. Altınöz, T. Dilan Demir, İ. Ali Gürses, G. Özcan, Extracranial transport of brain lymphatics via cranial nerve in human, Neurosci. Lett. 827 (2024) 137737, https://doi.org/10.1016/J.NEULET.2024.137737.
[22] M.S. Albayram, G. Smith, F. Tufan, I.S. Tuna, M. Bostancıklıoğlu, M. Zile, O. Albayram, Non-invasive MR imaging of human brain lymphatic networks with connections to cervical lymph nodes, Nat. Commun. 13 (2022) 1–14, https://doi.org/10.1038/s41467-021-27887-0, 2022 13:1.
[23] M. Johnston, D. Armstrong, L. Koh, Possible role of the cavernous sinus veins in cerebrospinal fluid absorption, Cerebrospinal Fluid Res. 4 (2007) 1–5, https://doi.org/10.1186/1743-8454-4-3.
[24] M. Furukawa, H. Shimoda, T. Kajiwara, S. Kato, S. Yanagisawa, Topographic study on nerve-associated lymphatic vessels in the murine craniofacial region by immunohistochemistry and electron microscopy, Biomedical Research 29 (2008) 289–296, https://doi.org/10.2220/biomedres.29.289.
[25] É. Mezey, I. Szalayova, C.T. Hogden, A. Brady, Á. Dósa, P. Sótonyi, M. Palkovits, An immunohistochemical study of lymphatic elements in the human brain, Proc Natl Acad Sci U S A 118 (2021), https://doi.org/10.1073/pnas.2002574118.
[26] R.O. Carare, M. Bernardes-Silva, T.A. Newman, A.M. Page, J.A.R. Nicoll, V. H. Perry, R.O. Weller, Solutes, but not cells, drain from the brain parenchyma along basement membranes of capillaries and arteries: significance for cerebral amyloid angiopathy and neuroimmunology, Neuropathol. Appl. Neurobiol. 34 (2008) 131–144, https://doi.org/10.1111/j.1365-2990.2007.00926.x.
[27] B. Engelhardt, P. Vajkoczy, R.O. Weller, The movers and shapers in immune privilege of the CNS, Nat. Immunol. 18 (2017) 123–131, https://doi.org/10.1038/ni.3666, 2017 18:2.
[28] E.N.T.P. Bakker, B.J. Bacskai, M. Arbel-Ornath, R. Aldea, B. Bedussi, A.W.J. Morris, R.O. Weller, R.O. Carare, Lymphatic clearance of the brain: perivascular, paravascular and significance for neurodegenerative diseases, Cell. Mol. Neurobiol. 36 (2016) 181–194, https://doi.org/10.1007/S10571-015-0273-8.
[29] E. Christensen, Studies on chronic subdural hematoma, Acta Psychiatr. Scand. 19 (1944) 69–148.
[30] M. Park, J.P. Park, S.H. Kim, Y.J. Cha, Evaluation of dural channels in the human parasagittal dural space and dura mater, Ann. Anat. 244 (2022), https://doi.org/10.1016/j.aanat.2022.151974.
[31] M.D. Johnson, Evaluation of fluid channels in the human dura and superior sagittal sinus in older patients, Neurosci. Lett. 746 (2021) 135649, https://doi.org/10.1016/j.neulet.2021.135649.
[32] Q. Ma, B.V. Ineichen, M. Detmar, S.T. Proulx, Outflow of cerebrospinal fluid is predominantly through lymphatic vessels and is reduced in aged mice, Nat. Commun. 8 (2017) 1–13, https://doi.org/10.1038/s41467-017-01484-6.
[33] R.M. Izen, T. Yamazaki, Y. Nishinaka-Arai, Y.K. Hong, Y.S. Mukouyama, Postnatal development of lymphatic vasculature in the brain meninges, Dev. Dynam. 247 (2018) 741–753, https://doi.org/10.1002/dvdy.24624.
[34] A. Louveau, I. Smirnov, T.J. Keyes, J.D. Eccles, S.J. Rouhani, J.D. Peske, N. C. Derecki, D. Castle, J.W. Mandell, K.S. Lee, T.H. Harris, J. Kipnis, Structural and functional features of central nervous system lymphatic vessels, Nature 523 (2015) 337–341, https://doi.org/10.1038/nature14432, 2015 523:7560.
[35] P. Yanev, K. Poinsatte, D. Hominick, N. Khurana, K.R. Zuurbier, M. Berndt, E. J. Plautz, M.T. Dellinger, A.M. Stowe, Impaired meningeal lymphatic vessel development worsens stroke outcome, J. Cerebr. Blood Flow Metabol. 40 (2020) 263–275, https://doi.org/10.1177/0271678x18822921.