

Research Article

Putative Primo-Vascular System in Rabbit Placenta



Fatma El-Zahraa A. Mustafa*

Department of Anatomy and Histology, Faculty of Vet. Medicine, Assiut University, Assiut, Egypt

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Abstract

The primo vascular system (PVS) is a very important topic of study nowadays because of their role in transport and regeneration of tissue and in cell migration and cancer metastasis. The PVS was detected in different organs of the rabbit but not in the placenta. In this work, we observe the PVS inside the blood vessels of the placenta for the first time. The main characteristic features of the primo vessels (PVs) from the rabbit placenta were in agreement with the PVS in different organs of animals, including the rod-shaped nuclei and their arrangement.

1. Introduction

The primo vascular system (PVS), known as the circulatory system recently, is observed and found throughout the body [1]. The PVS has been detected in the blood vessels of rodents [2–4], in the heart of bovine [5], in the lymph vessels of rabbits and rats [6,7], in the central nervous system of rabbits and rats [8], and on the surfaces of numerous organs of different rodents [9]. The PVS is expected to play a role in the cancer metastatic pathway in cancer-bearing rodents [10,11].

The medical importance of the PVS has been understood by the abundance of mast cells, hematopoietic stem cells, and embryonic-like stem cells in these vessels. Therefore, the PVS is considered a key player in immunity and renewal of cells.

The function of the PVS in cancer metastasis was reported [12]. Moreover, the PVS is considered important in the hormone transport pathway due to the abundance of catecholamine-producing cells in these vessels [13].

Therefore, studying placental PVS is very important to fully understand the immune importance, stem cells, and regeneration of these vessels. In this article, we explore the PVS in the rabbit placenta.

* Corresponding author.

E-mails: f.histology@aun.edu.eg, f.331986@yahoo.com.

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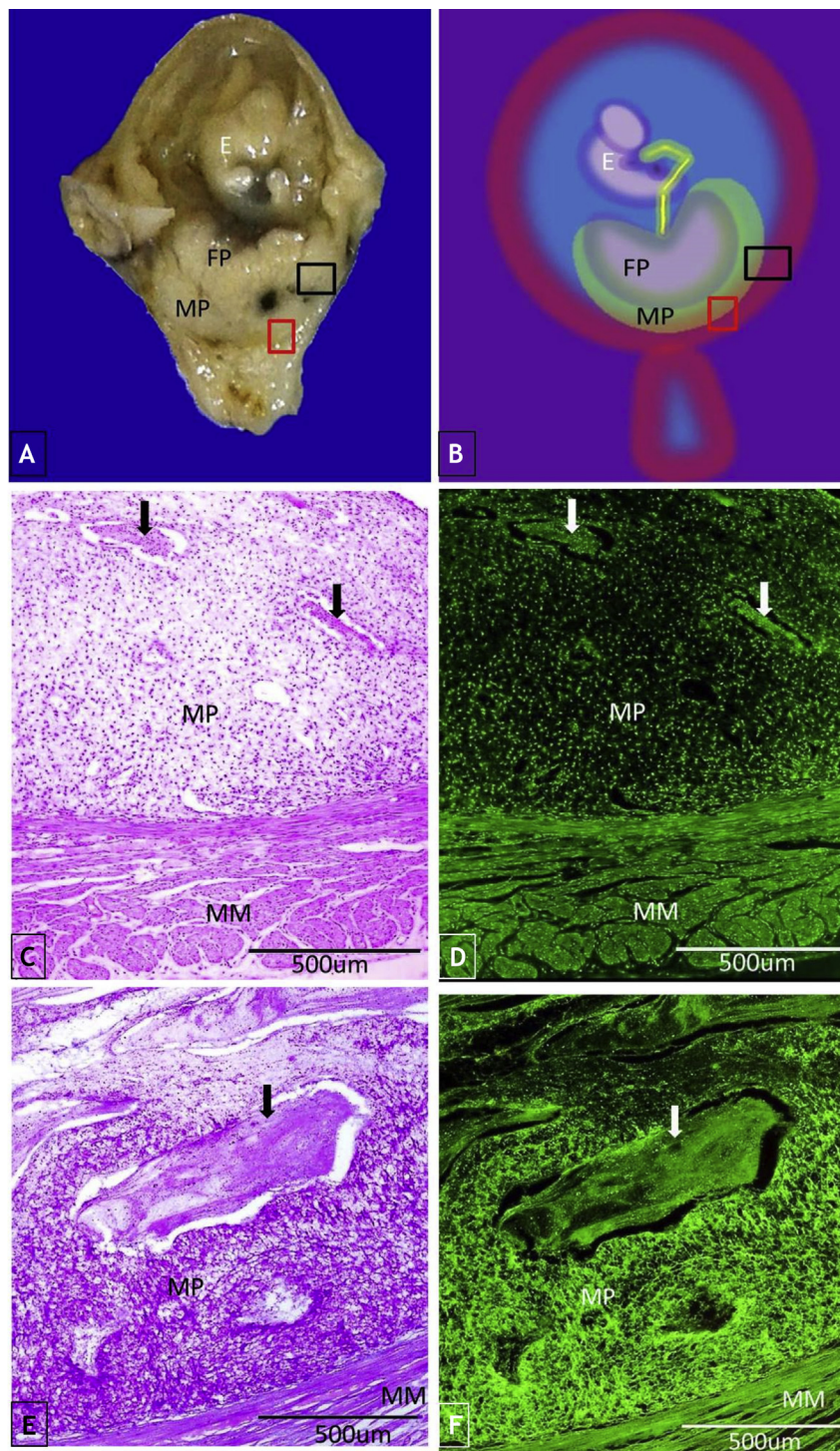


Figure 1 Primo vessels in the placenta of a rabbit. (A) Positions where primo vessels were observed. (B) Schematic illustration showing positions where primo vessels were observed (C–E) Primo vessels on the maternal side of the placenta (black arrow). MP (maternal side of the placenta), FP (fetal side of the placenta), E (embryo), MM (myometrium). Black and red frames represent samples in c and e, respectively. (C) Stained using hematoxylin and eosin. (E) Stained using combined Alcian blue–PAS (D and F) are negative images of (C and E), respectively. Black and red frames represent samples in (C) and (E), respectively. E = embryo; FP = fetal side of the placenta; MM = myometrium; MP = maternal side of the placenta.

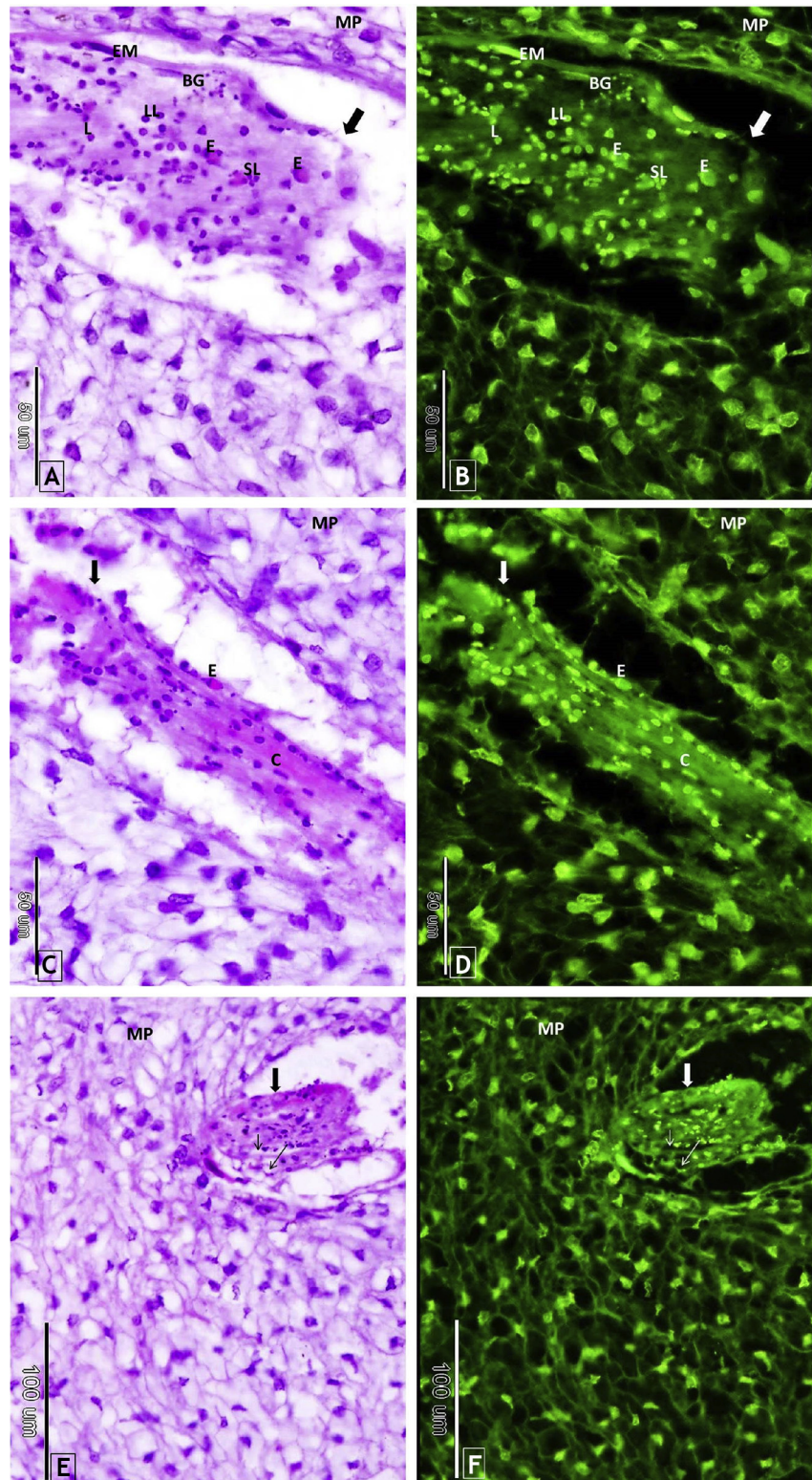


Figure 2 Images showing histological features of primo vessels After hematoxylin and eosin images (A, C, E). respectively and negative images are (B, D, F). Large black arrows denote primo vessels on the maternal side of the placenta (large black arrows), sinuses (small black arrows) MP = maternal side of the placenta; EM (epithelium), BG (basophilic granules), L (lymphocytes), IL (large lymphocytes), SL = small lymphocytes), E (eosinophils), C (hyalinized stroma).

2. Material and methods

2.1. Sample collection

The study was approved by the ethics panel of Assiut University, Egypt. The material included in this work was obtained from 3 mature does of the New Zealand white rabbit (4–5 months old), which grew up in the faculty of medicine animal house, Assiut University. The rabbits were housed separately under a consistent and controlled environment of ventilation, humidity, light (12-h light/dark cycle), and temperature (23–25°C). The sexually mature females were mated with bucks of the same species. The whole uterus was fixed immediately after slaughtering.

2.2. Histological preparation

The collected materials were dissected as quickly as possible and then fixed in Bouin's fluid for 22 hours. The fixed samples were dehydrated in an ascending series of ethanol, then cleared in methyl benzoate, and embedded in paraffin wax. Transverse paraffin sections of 1–7 μm thickness were cut and stained with Harris hematoxylin and eosin and combined Alcian blue–PAS [14].

3. Results

Fig. 1 showed the maternal side and fetal side of the placenta of the rabbit, myometrium, and embryo within the uterus. The PVs were detected in the maternal side of the placenta of the rabbit (Fig. 1A and B). Moreover, the PVs were observed on the maternal side of the placenta as a distinctive tissue element inside the blood vessels and not attached to the wall of the blood vessels (Fig. 1C–F). Morphometrical measurements revealed that the mean thickness of the PV was about $52.21 \pm 8.341 \mu\text{m}$.

Fig. 2A–B showed that boundaries of PVs are not surrounded by an external membrane of endothelium and that they are surrounded by an outer layer of epithelial cells with deeply stained nuclei.

The PVs contain basophilic granules ($1.48 \pm 0.14 \mu\text{m}$) mainly composed of large amounts of basophilic structures with nucleic structures resembling chromatin that are strongly stained with hematoxylin. In addition, PVs contain various kinds of cells, including granulocytes (eosinophils) and lymphocytes (of different size), which are haphazardly distributed in the matrix singly or in small groups, all of which are embedded in the matrix of the PV (Fig. 2A–D).

The PV stroma appeared as a fibrous bundle that is loosely arranged and infiltrated with inflammatory cells, fibrin-like fibers, and amorphous intercellular substance as a loose hyalinized stroma (Fig. 2C–D). The nuclei of endothelium cells in PVs are cigar-shaped ($7.60 \pm 0.4676 \mu\text{m}$), whereas they appear in the form of broken rods along the PV.

Histological examination of the placental PVs revealed the absence of erythrocytes in these vessels (Fig. 2A–D). In addition, the PV matrix consists of many sinuses of variable sizes with an average diameter of $4.13 \pm 0.24 \mu\text{m}$ (Fig. 2E–F).

4. Discussion

In this research on the PVs detected on the placental blood vessels, data were strengthened by previous observations of the PVs detected within the blood vessels of rodents [15].

Previous literature described the characteristics of PVs found on the surface of internal organs [16,17]. These structures showed sinuses, flattened nuclei that are longitudinally arranged, outer epithelium membranes, and many fibers. In this research, PVs were detected freely floating in placental vessels and resembled the features of PVs reported in previously published articles.

This PV showed the ordinary character of the PVs as shapes and sizes. In addition, the longitudinal and broken appearance of nuclei of the endothelial cells of placental PVs resembled the previously detected PVs [18], many fibers and external membranes were easily detected. Moreover, our observations revealed that these placental structures were related to the PVS family despite the observation of some contrast with the previously published work [16,17]. In this study, the outer membrane was clearly detected, but the inner one was not clear. We suggest that as the circulatory system showed different kinds of vessels, similarly, there should be different types of PVs. In this research article, we explored a new kind of PV within the placenta and expected that in the future, various kinds of PVs might be detected.

The morphometric characteristics of the PVS within the placental vessels in this study appeared in agreement with those of the PVS within other vessels previously studied in different animals. The mean thickness of PVs in our study was nearly similar to that of PVs in mice (20 μm) [3], rats (46 μm) [4,5], and rabbits (50 μm) [7,19]. Moreover, the thickness of PVs in different organs resembled that of those previously studied in the lymph organs of rats [20–22], on the surface of organs of rabbits [21], and in the brain of rats [22], which was 32 μm , 45 μm , and 23 μm , respectively. Our data revealed that the PV thickness was $52.21 \pm 8.341 \mu\text{m}$.

According to Kim et al [1], the PVS was found to be a key player in regeneration of cells and tissue. This was supported by the presence of stem cells in these vessels [12,23]. In the present study, various blood cells, including granulocytes (eosinophils) and lymphocytes, were found to be embedded in the matrix of PVs. More experiments in this way are needed in the future.

Disclosure statement

No conflict of interest.

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References

- [1] Kim JD, Ogay V, Lee BC, Kim MS, Lim I, Woo HJ, et al. Catecholamine producing novel endocrine organ: Bonghan system. *Med Acupunct* 2008;20:97–102.

- [2] Soh KS. Current state of research on the primo vascular system. Its role in cancer and regeneration. In: Soh KS, Kang KA, Harrison D, editors. *The Primo Vascular System*. New York: Springer; 2011. p. 25–38.
- [3] Yoo JS, Kim MS, Ogay V, Soh KS. In vivo visualization of Bonghan ducts inside blood vessels of mice by using an Alcian blue staining method. *Indian J Exp Biol* 2008;46:336–9.
- [4] Baik KY, Lee BC, Johng HM, Nam TJ, Sung B, Soh KS. Long threadlike structure inside the blood vessels of rats. *Newest Med* 2004;47:18–22.
- [5] Lee BC, Baik KY, Johng HM, Nam TJ, Lee J, Sung B, et al. Acridine orange staining method to reveal the characteristic features of an intravascular threadlike structure. *Anat Rec B New Anat* 2004;278:27–30.
- [6] Lee HS, Park WH, Je AR, Kweon HS, Lee BC. Evidence for novel structures (primo vessels and primo nodes) floating in the venous sinuses of rat brains. *Neurosci Lett* 2012;522:98–102.
- [7] Jiang X, Lee B-C, Choi CH, Baik K-Y, Soh K-S, Kim HK, et al. Tubular substructure of intravascular thread-like structures from rats and rabbits. *J Korean Phys Soc* 2004;44:1602–4.
- [8] Jia ZF, Soh KS, Zhou Q, Dong B, Yu WH. Study of novel threadlike structures on the intestinal fascia of dogs. *J Acupunct Meridian Stud* 2011;4:98–101.
- [9] Cho SJ, Kim BS, Park YS. Threadlike structures in the aorta and coronary artery of swine. *J Int Soc Life Inf Sci* 2004;22: 609–11.
- [10] Kwon BS, Ha CM, Yu S, Lee BC, Ro JY, Hwang S. Microscopic nodes and ducts inside lymphatics and on the surface of internal organs are rich in granulocytes and secretory granules. *Cytokine* 2012;60:587–92.
- [11] Langevin HM, Yandow JA. Relationship of acupuncture points and meridians to connective tissue planes. *Anat Rec* 2002;269: 257–65.
- [12] Ogay V, Soh KS. Identification and characterization of small stem-like cells in the primo vascular system of adult animals. In: Soh KS, Kang KA, Harrison D, editors. *The Primo Vascular System*. New York: Springer; 2011. p. 149–55.
- [13] Kim BH. The Kyungrak system. *J Jo Sun Med* 1965;108:1–38.
- [14] Bancroft JD, Gamble M. *Theory and Practice of Histological and Histochemical Techniques*. 3rd ed. New York NY USA: Churchill Livingstone; 2002. Butter Worths.
- [15] Lee B, Yoo JS, Baik KB, Kim KW, Soh K. Novel threadlike structures (Bonghan Ducts) inside lymphatic vessels of rabbits visualized with a Janus Green B staining method. *The Anatomical Record* 2005;286:1–7.
- [16] Lee BC, Yoo JS, Ogay V, Kim KW, Dobberstein H, Soh KS, et al. Electron microscopic study of novel threadlike structures on the surfaces of mammalian organs. *Microsc Res Tech* 2007;70: 34–43.
- [17] Yoo JS, Ayati MH, Kim HB. Characterization of the primo-vascular system in the abdominal cavity of the lung cancer mouse model and its differences from the lymphatic system. *PLoS ONE* 2010;5. e9940.
- [18] Fung PC, Kong RK. The integrative five-fluid circulation system in the human body. *Open J Mol Integr Physiol* 2016;6:45–97.
- [19] Lee BC, Soh KS. Contrast-enhancing optical method to observe a Bonghan duct floating inside a lymph vessel of a rabbit. *Lymphology* 2008;41:178–85.
- [20] Lee SH, Bae KH, Kim GO, Nam MH, Choi YB, Kwon HM, et al. Primo vascular system in the lymph vessel from the inguinal to the axillary nodes. *Evid Based Complement Alternat Med* 2013;2013:472704.
- [21] Shin HS, Johng HM, Lee BC, Cho SI, Soh KS, Baik KY, et al. Feulgen reaction study of novel threadlike structures (Bonghan ducts) on the surfaces of mammalian organs. *Anat Rec B New Anat* 2005;284:35–40.
- [22] Lim J, Jung JH, Lee S, Su Z, Qiang Z, Cha JM, et al. Estimating the density of fluorescent nanoparticles in the primo vessels in the fourth ventricle and the spinal cord of a rat. *J Biomed Opt* 2011;16:116010.
- [23] Lee SJ, Park SH, Kim YI, Hwang S, Kwon PM, Han IS, et al. Adult stem cells from the hyaluronic acid-rich node and duct system differentiate into neuronal cells and repair brain injury. *Stem Cells Dev* 2014. <https://doi.org/10.1089/scd.2014.0142>.