นิพนธ์ต้นฉบับ

ระบบไหลเวียนเลือดแบบปิดในม้ามของค้างคาวแม่ไก่

ศิรินุช ศรีเจริญเวช* ภัสรา ลานเหลือ* นราวดี ชมภู* และ อภิชญา นิยมจันทร์*

บทคัดย่อ

วัตถุประสงค์: เพื่อศึกษารายละเอียดของระบบไหลเวียนเลือดในม้ามของค้างคาวแม่ไก่ โดยใช้เทคนิค vascular corrosion cast ร่วมกับกล้องจุลทรรศน์อิเล็กตรอนแบบส่องกราด

วิธีการ: สัตว์ทดลองเป็นค้างคาวแม่ไก่คละเพศ จำนวน 12 ตัวใช้ในการศึกษาโครงหลอดเลือดอย่างละเอียดโดยเทคนิค vascular corrosion cast ร่วมกับกล้องจุลทรรศน์อิเล็กตรอนแบบส่องกราด

ผลการศึกษา: พบว่า ม้ามของค้างคาวแม่ไก่ ได้รับเลือดมาจาก splenic artery และ short gastric artery โดยให้แขนงเรียก trabecular artery ซึ่งแตกแขนงต่อไปเป็น central artery โดยวางตัวชิดขอบด้านใดด้านหนึ่งของ white pulp และให้แขนงขนาดเล็กเพื่อไปเลี้ยง white pulp โดยตรง เมื่อ central artery ผ่านมาในส่วน red pulp จะแตกแขนงออกโดยรอบเป็น penicillar arteriole ซึ่งเลี้ยงในส่วนของ white pulp, red pulp และ marginal zone แขนงปลายของ penicillar arteriole เรียกว่า terminal arterial capillary จะติดต่อกับ red pulp sinusoid โดยตรง ก่อนจะรวมกันเป็น pulp venule เทเข้าสู่ trabecular vein และ splenic vein ตามลำดับ จึงสรุปได้ว่า ระบบไหลเวียนเลือดในม้ามของค้างคาวแม่ไก่เป็นระบบไหลเวียนเลือดแบบปิด **วารสารเทคนิคการแพทย์เชียงใหม่ 2550; 40: 61-70.**

ภาควิชากายวิภาคศาสตร์ คณะแพทยศาสตร์ศิริราชพยาบาล มหาวิทยาลัยมหิดล กรุงเทพฯ 10700 ประเทศไทย

Abstract : An investigation of the splenic closed microcirculation in Lyle's flying fox (*Pteropus lylei*)

Sirinush Sricharoenvej* Passara Lanlua* Narawadee Choompoo* Apichaya Niyomchan*

Objective: The purpose of this study was to elucidate the splenic microvasculature of *P. lylei*. The vascular corrosion cast technique with the SEM was applied in order to clarify this investigation.

Methods: The spleens of twelve adult *P. lylei* in both sexes were processed by using vascular corrosion cast technique combined with the SEM.

Results: It was revealed that the spleen receives the main arterial supply from the splenic and short gastric arteries. These arteries divide into many trabecular branches. Each trabecular artery branches into central arteries, that eccentrically situates in the white pulp. Along their courses in the white pulps, the central arteries give off small branches supplying the white pulps. Then, these arteries pass into the red pulp and ramifies into many small penicillar arterioles that nourishe the white and red pulps as well as marginal zones. The termination of these arterioles directly connect with the red pulp sinusoids that empty the blood into the pulp venules. The pulp venules proceed to the trabecular and splenic veins, respectively. It was concluded that splenic circulation of *P. lylei* is the closed type of blood circulation. **Bull Chiang Mai Assoc Med Sci 2007; 40: 61-70.**

Key words: Spleen, *Pteropus lylei*, Microvascularization, SEM

*Department of Anatomy, Faculty of Medicine Siriraj Hospital, Mahidol University, Bangkok, 10700, Thailand

Introduction

One of the interesting organs in abdomen is spleen. Its functions are both morphologic and immunologic filtrations.[1-4] The splenic parenchyma or splenic pulp consists of white and red pulps. The white pulp appears as circular or elongated whitish-gray spheres that are surrounded by the red pulp.[3] In general, red pulp is the most part that is concerned to the splenic ability to dispose of worn-out red blood cells and blood – borne pathogens, while the white pulp mainly serves the immune functions of the spleen. Consequently, the splenic circulation provides the channels for cell-cell interaction in these functions. However, the manner, in which blood flow from the arterial capillaries to the red pulp sinusoids, has been controversially discussed. Additionally, the

pattern of blood flow from red pulp sinusoids to trabecular veins has not yet been completely explained. Some investigators considered that the capillaries of the spleen directly drain into the red pulp sinusoids and trabecular veins, respectively, so it has been concluded as a closed type of blood circulation. The others found that the blood from capillaries passes through the spaces between the splenic cords, and then it is collected by the red pulp sinusoids and trabecular veins. In this category, it has been classified as an open type.[1-4]

Since the splenic microcirculation is one of the most debated topic, many researchers had been intensively studied the modes of arterial termination by using various techniques.[5-21] The vascular corrosion cast technique in conjunction with scanning electron microscopy (SEM) have been employed for detailed studies of the microarchitecture in various organs, including that in the spleen.[5-12,14,18,19] However, the splenic microcirculation of the *P. lylie* has not been investigated whether it is the closed or open type. Therefore, it is of interest to apply vascular corrosion cast technique with the SEM in order to clarify the patterns of the blood flow in this mammal.

Materials and Methods

Twelve adult Lyle's flying foxes (*Pteropus lylei*) of both sexes, weighing between 350-450 g, were used. After each animal was anesthetized by halothane inhalation, Batson's no.17 plastic mixture injection and the preparation of the splenic microvascular casts for the study with the SEM were done according to the process that had been previously described by Bamroongwong and her coworkers.[5]

Results

After the splenic microvascular casts were examined under the SEM, it was revealed that the main blood supply of this organ was splenic and short gastric arteries as well as the corresponding veins. After the entering of the arteries at its hilum, they divided into several branches, which deeply passed into the splenic parenchyma and ran along trabeculae as trabecular arteries (Fig 1). After leaving the trabeculae and passing into the white pulp, the trabecular artery subdivided into central or white pulp arteries (Fig 2). After the splenic microvascular casts were cut in the cross section, two distinguished areas were demonstrated, which were white and red pulps. These two pulps were supplied by different patterns of the microcirculation. In the white pulp, the central arteries, with varying in sizes, gave off small branches to supply the white pulp along their courses as shown in Figure 3. In the longitudinal view, the capillaries of this pulp formed a tubular structure surrounding the central artery (Fig 4). Moreover, the central artery eccentrically located in the pulp. Then, this artery passed into the red pulp. In the red pulp, the central artery ramified into many branches with the reduced sizes. These were called penicillar arterioles (Fig 5, 6). It is noticeable that the termination of these arterioles or terminal arterial capillaries acted as two patterns. Firstly, the branches of the penicilli in the marginal zone turned back to the marginal sinusoids and the white pulp (Fig 4-6). Secondly, they passed into the red pulp and directly connected with the red pulp sinusoids (Fig 7). Therefore, it was concluded that this characteristic of the spleen in the *P. lylei* was the closed type of microcirculation. Then, both marginal and red pulp sinusoids drained into the pulp venules that proceeded to the trabecular and splenic veins, respectively (Fig 8).

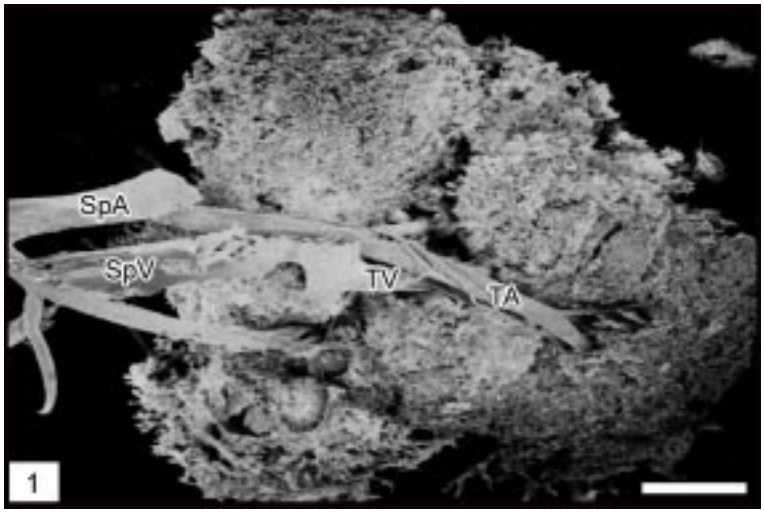

Figure 1 SEM micrograph of the splenic vascular cast in *P.lylei* showing the splenic artery (SpA) supplying the spleen. Contrastly, the trabecular vein (TV) drained into the splenic vein (SpV). Trabecular artery (TA). Bar = 1,000 μm

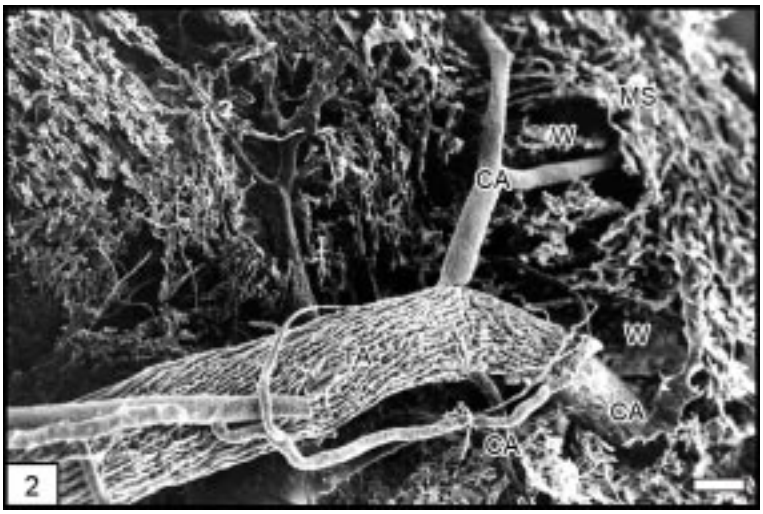

Figure 2 The trabecular artery (TA) divided into many branches of central artery (CA) that passed into the white pulp (W). Marginal sinusoid (MS). Bar = 100 μm

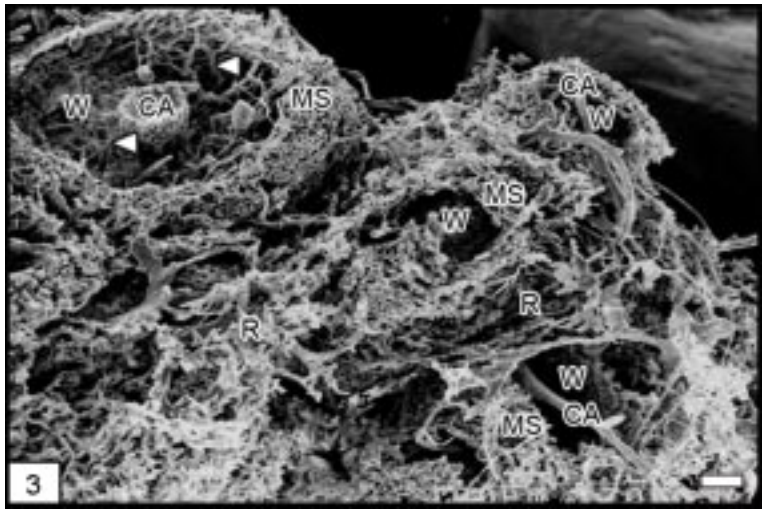

Figure 3 The central artery (CA) gave off many small branches (arrowheads) supplying the white pulp (W) and marginal zone through marginal sinusoid (MS). Red pulp (R). Bar = 100 μm

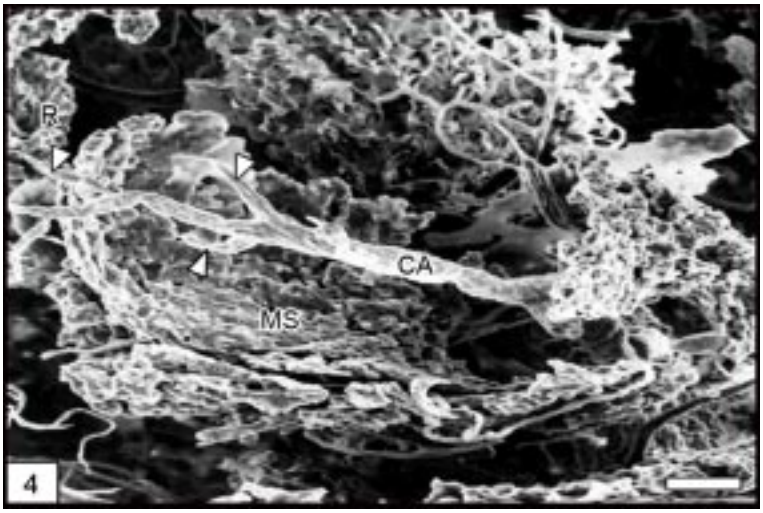

Figure 4 Longitudinal section of the splenic vascular cast in the white pulp showing the central artery (CA) gave rise to penicillar arterioles (arrowheads) connecting to the marginal (MS) and red pulp sinusoids (R). Bar = 100 μm

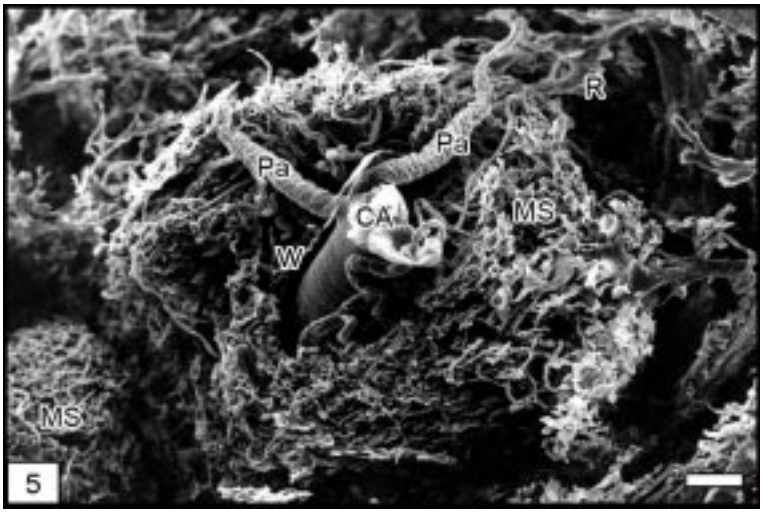

Figure 5 Cross section of splenic vascular cast. The central artery (CA) eccentrically located in the white pulp (W) and ramified into penicillar arterioles (Pa). Then, these arterioles terminated into marginal (MS) and red pulp sinusoids (R). Bar = 100 μm

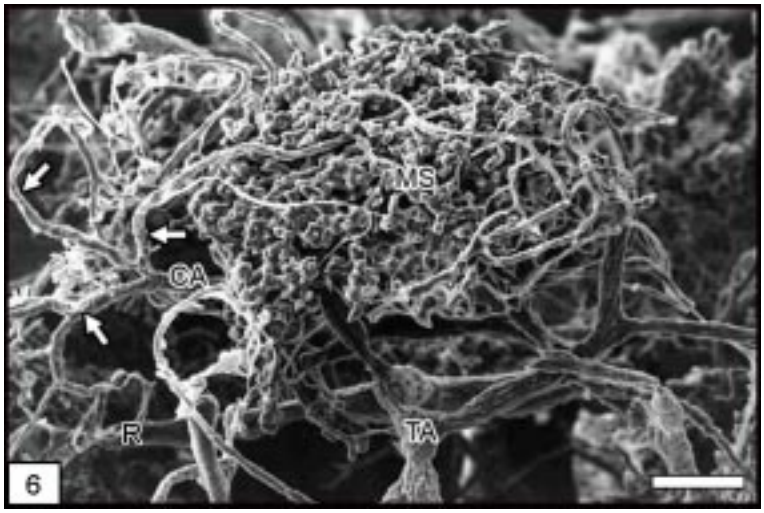

Figure 6 Lateral view of the white pulp that was surrounded by marginal sinusoids (MS). The terminal end of the central artery (CA) sent out radiating penicillar arterioles (arrows), that rapidly reduced in diameter and supplied marginal and red pulp sinusoids (R). Trabecular artery (TA). Bar = 100 μm

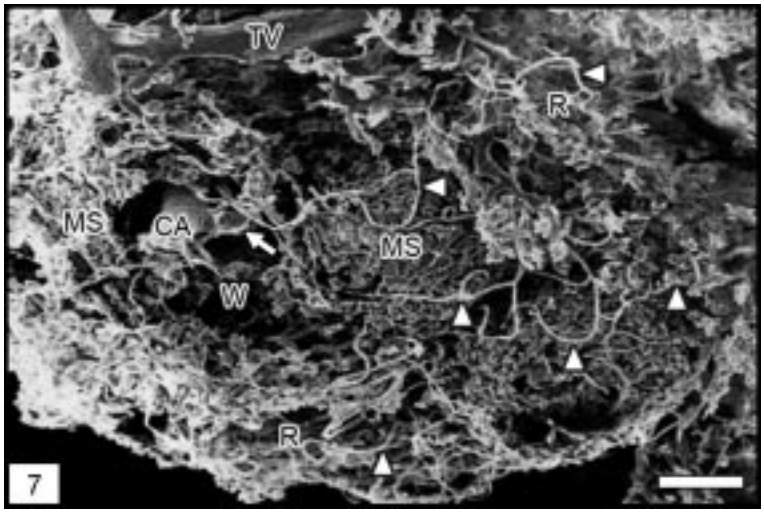

Figure 7 **The penicillar arteriole (an arrow) supplied marginal sinusoid (MS) and finally became terminal arterial capillaries (arrowheads) that directly connected to red pulp sinusoids (R). Central artery (CA), trabecular vein (TV) and white pulp (W). Bar = 250 μm**

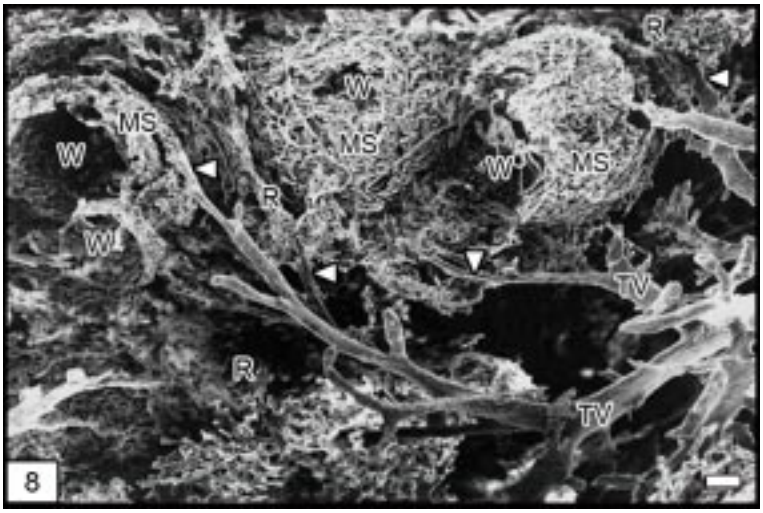

Figure 8 **The marginal (MS) and red pulp sinusoids (R) directly drained into pulp venules (arrowheads) that collected into trabecular vein (TV). White pulp (W). Bar = 100 μm**

Discussion

With the combination of the injected plastic replica and the SEM techniques, it is clearly shown that the spleen of the Lyle's flying fox receives the main blood supply and their branching patterns as those of human[2,3], common tree shrew[5], pig, horse, cow[6], dog[6,9], cat[6,9,22] and rat.[6,8] After that the central arteries branch into penicillar arterioles, which

supply the marginal zone and the red pulp. Surprisingly, these arterioles turn back into the marginal zone as recurrent branches. This phenomenon might be related to provide either double immunological filtration or sufficient blood supply in this zone. Moreover, it is appreciable that the penicillar arterioles rapidly reduce in size into terminal arterial capillaries in the marginal zone. This characteristic has never been mentioned before in the other mammals.[2,3,5-11] It is possible that rapid decrease in size of the penicillar arterioles might be modified to control the blood flow into the spleen, when this bat changes its positions. Furthermore, the capillary of the marginal zone is the sinusoidal type that is suitable for its function. The sinusoids of the red pulp usually act as a reservoir of the blood in the spleen.

By using vascular corrosion cast technique combined with the SEM, Kashimura and Fujita[11] had studied the blood circulation of the spleen in both instant and prolonged dead experimental animals. In the instant dead model, the walls of capillaries were intact. After the plastic mixture was injected into these capillaries, the plastic media filled in the vessels. Therefore, the splenic circulation of the instant dead model is the closed type. On the other hand, the autolysis occurred on the walls of the capillaries in the prolonged dead experimental animals. When the plastic mixture was injected into the capillaries of these models, the media leaked through the walls of these capillaries. Consequently, it has been the open type of splenic circulation in these prolonged dead animals. In addition, Bamroongwong and her coworkers[5] had studied splenic circulation in the common tree shrew by using vascular corrosion cast technique with the SEM. Several rates and volumes of plastic mixture were injected into the spleen. With the optimal rate and volume of the plastic injection, it was found that the splenic circulation of the common tree shrew is the closed type. If the plastic media is perfused at the high rate and with large volume, the leakage of the media will be found in the splenic cords. It is shown the open type in this situation. Because of either the morphological changes on the vascular wall after death or unsuitable technique of the plastic injection, this can lead to weakening on vascular wall that causes the leakage of injected media. Therefore, these two mentioned conditions effect on the interpretations of either closed or open type in the splenic circulation. It is concluded that the splenic circulation in the *P. lylei* is the closed type that is similar to those in common tree shrew[5], horse and cow.[6] However, the open type can be demonstrated in dog[6,9], rat[6,8,14,17] and cat.[6,10,22]

Conclusions

The spleen of the *P. lylei* receives main blood supply from the splenic and short gastric arteries. These arteries penetrate at the splenic hilum and divide into trabecular arteries. The trabecular artery branches into central arteries which give small branches to supply the white pulp and marginal zone. After leaving the white pulp, the central artery ramifies into many penicillar arterioles that rapidly reduce in diameter and end as terminal arterial capillaries. The capillaries directly join to the red pulp sinusoid. Therefore, the splenic closed microcirculation is demonstrated in this mammal under the suitable conditions of vascular corrosion cast technique.

Acknowledgement

The authors wish to thank Prof. Dr. Sanjai Sangvichien and Assoc. Prof. Dr. Sitha Piyawinijwong for their valuable comments, suggestion and consultation throughout this study. We would like to extend our gratitude to every staffs in the Electron Micro-

scopic Laboratory, Department of Pathology, and every microtechnique laboratory staffs in Department of Anatomy, Faculty of Medicine Siriraj Hospital for their technical helps.

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