

# Post-natal development of some lymphoid organs of West African Dwarf (WAD) goats: a histological perspective

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## *Abstract*

The study evaluated the developmental changes in the mesenteric lymph nodes, thymus and spleen of West African Dwarf (WAD) goat using histological techniques. Twenty-seven WAD goats of 1 day, 2 weeks and 1 to 7 months old were used for this study. Results showed that the mesenteric lymph node of the WAD goats had poorly organized cortical and medullary areas at 1 day postnatum, and dense infiltration of lymphocytes in the cortex with a few primary lymphoid follicles at 2 and 3 months. At 1 day old, the thymus exhibited thin cortical parenchyma and extensive medullary areas. The thymic parenchyma of 3 and 4 months old showed distorted outlines of the cortex and medulla with adipose tissue deposition in the trabeculae. The number of Hassal's corpuscles increased from the 1<sup>st</sup> day to the 7<sup>th</sup> month of birth. Furthermore, at 1 day old, the spleen demonstrated poorly defined red pulp and white pulp areas. The outlines of the red and white pulp areas were more obvious at 2 weeks of age and the red pulp areas became more expansive with increased concentration of red blood cells from 1 to 7 months old. It was concluded that the mesenteric lymph nodes of WAD goats may attain adult morphology as early as 1 month after birth, the thymus which appeared as a fully functional thymus at birth could regress at 3 months after birth and the spleen could perform limited lymphoid functions immediately after birth.

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**Keywords:** development, histology, lymphoid organs, postnatum, West African Dwarf goats

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## Introduction

Primary and secondary lymphoid organs have major roles in the immune system of mammals. They are strategically positioned in defense, against invading pathogens. Over the years, interest in the ontogenesis of mammalian organs has increased, leading to reports on the diversities and morpho-functional significance of several organs (Ngo et al., 2001; Boehm and Bleul, 2007; Randall et al., 2008; Landreth, 2012). While the primary lymphoid organs are believed to develop during embryogenesis, secondary lymphoid organs have been shown to develop during embryogenesis or in the first few weeks after birth (Randall et al., 2008, Farley et al., 2013). The timing of development and the structural organization of lymphoid organs postnatum may inform their functionality at different stages of adult life. In animal species largely exposed to wide range of pathogens, the prevention, control and treatment of diseases will become more feasible with adequate knowledge of the developmental trend of lymphoid organs in each species.

Except for the management and disease constraints, the West African Dwarf goat is an important domestic animal species in the tropics with huge potential to provide a good percentage of animal protein needs in the Sub-Saharan Africa (FAO, 1991). In most ruminants including WAD goats, the normal structures of the lymph nodes, thymus and spleen have been reported (Gomariz et al., 1989; Pearse, 2006; Willard-Mack, 2006; Cesta, 2006; Zidan and Pabst, 2015). In the development of these lymphoid organs, emphases have been on embryogenesis with little recourse to post-natal changes that may occur in these organs (Kutyrev et al., 2008; Landreth, 2012; Prasad et al., 2012; Farley et al., 2013). Although a few reports exist on the post-natal changes in the structure of lymph nodes, thymus, and spleen, available literature does not reveal the post-natal developmental features of these entities in the West African Dwarf (WAD) goat reared in Sub-Saharan African environment.

Therefore, the present study focused on the post-natal developmental changes in the structures of the mesenteric lymph nodes, thymus and spleen of WAD goats with the aim of establishing the periods of peak development, and their morpho-functional significance.

## Materials and Methods

**Experimental animals:** A total of 27 West African Dwarf (WAD) goats of 1 day, 2 weeks, 1 month, 2 months, 3 months, 4 months, 5 months, 6 months and 7 months old were used for this study. Each age group consisted of 3 animals. They were obtained from local markets in Nsukka local government area, Enugu State and acclimatized in the animal house of the Department of Veterinary Anatomy, University of Nigeria, Nsukka, Enugu State. Following acclimatization, the animals were sacrificed for human consumption.

**Histological procedures:** Samples of the mesenteric lymph nodes, spleen and thymus of the WAD goats were obtained and fixed by immersion in Bouin's fluid

for 36 hours. The fixed samples were then dehydrated in increasing concentrations of ethanol, cleared in three changes of xylene and embedded in paraffin wax. The mounted tissues were then sectioned in a rotary microtome to obtain a 5- $\mu$ m thick section. The tissues which were mounted on glass slides were stained routinely with haematoxylin and eosin for light microscopy. Photomicrographs were captured using a Moticam digital camera (Motic China Group Co., Ltd, Xiamen, China).

## Results

**Histology of WAD goat mesenteric lymph nodes:** The bean-shaped mesenteric lymph nodes of the WAD goats exhibited connective tissue capsules which extended trabeculae into the interior of the organ; a cortex beneath the capsule made up areas of lymphocytic infiltration, lymphoid nodules with or without germinal centres, macrophages lining the cortical sinuses, and reticular cells; and the medulla characterized by medullary cords and dilated medullary sinuses frequently bridged by reticular cells and fibres.

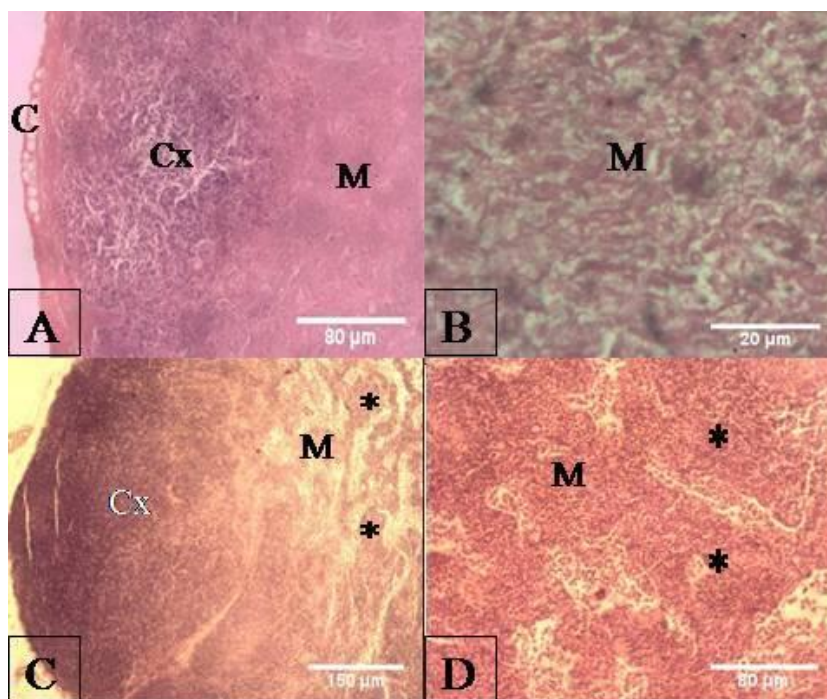
However, at day 1 postnatum, the cortical and medullary tissues of the lymph nodes were poorly organized (Fig. 1A). The cortex showed lightly infiltrated areas of lymphocytes without lymphoid nodules and the medulla demonstrated forming sinuses and unorganized medullary cords (Fig. 1). The short trabecula which extended from the capsule terminated on the periphery of the cortex. At 2 weeks, 1 month, 2 months, 4 months, 5 months, 6 months and 7 months postnatum, the lymph nodes showed organized cortical and medullary areas. At 2 weeks and 1 month old, the lymph node cortex contained densely infiltrated lymphocytes without lymphoid follicles while the medulla demonstrated medullary cords and medullary sinusoids (Fig. 1). At 2 and 3 months, the lymph node cortex showed dense infiltration of lymphocytes with few primary lymphoid follicles. At 4 to 7 months postnatum, large populations of lymphoid follicles were observed in the cortical parenchymal areas (Fig. 2).

**Histology of WAD goat thymus:** The thymus of the WAD goats were encapsulated. The connective tissue capsule penetrated the thymic parenchyma, dividing it into incomplete thymic lobules. Each thymic lobule showed peripherally darkly stained cortex and a central light medulla. The cortex was made up of large population of thymocytes in a stroma of epithelial reticular cells. The thymic medulla exhibited a cyto-reticulum of epithelial reticular cells with less densely packed lymphocytes and thymic corpuscles. Blood vessels and nerves were observed in the trabeculae.

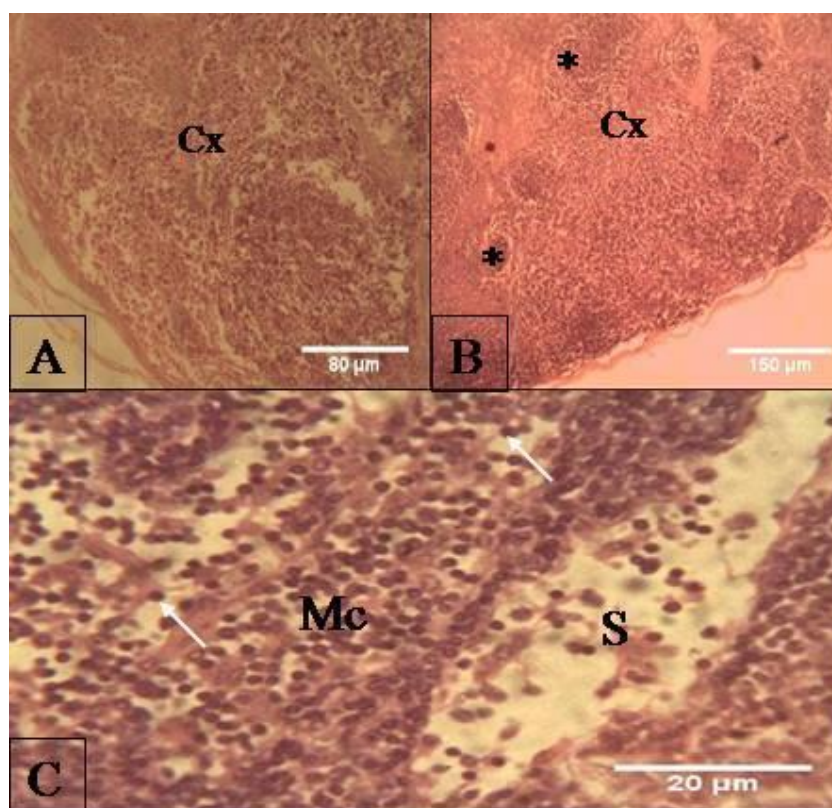
However, at day 1 postnatum, the thymus of the WAD goats were characterized by thin cortical parenchyma, and extensive medullary area (Figs. 3, 4). The thymic lobules were poorly defined at day 1 (Fig. 3A). Moreover, at week 2, months 1 and 2, the thymic lobules were well defined, and the medullary parenchymal areas contained thymic corpuscles (Fig. 3B). At 3 and 4 months old, the cortex and medulla of the thymic lobules were observable but their outlines

were distorted. Adipose tissues were deposited within the trabeculae of the 3- and 4-month-old WAD goats (Fig. 3C). Furthermore, at 5 to 7 months old, the cortical tissues of the thymus were lost and medullary areas

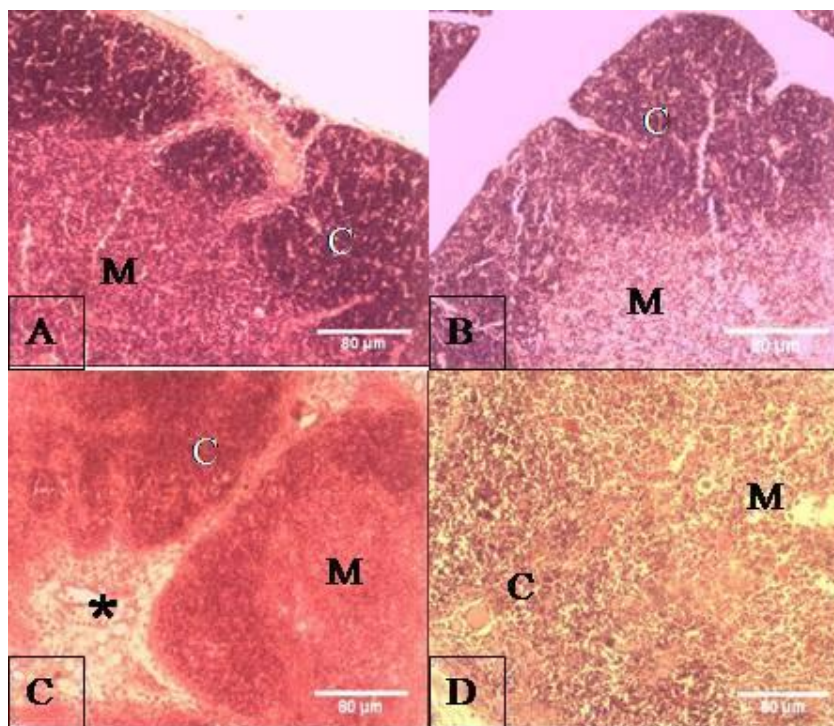
became extensive containing many Hassal's corpuscles (Figs. 3D, 4), whereas at day 1, few Hassal's corpuscles were present in the thymic medulla; the number increased as the animal aged (Fig. 4).



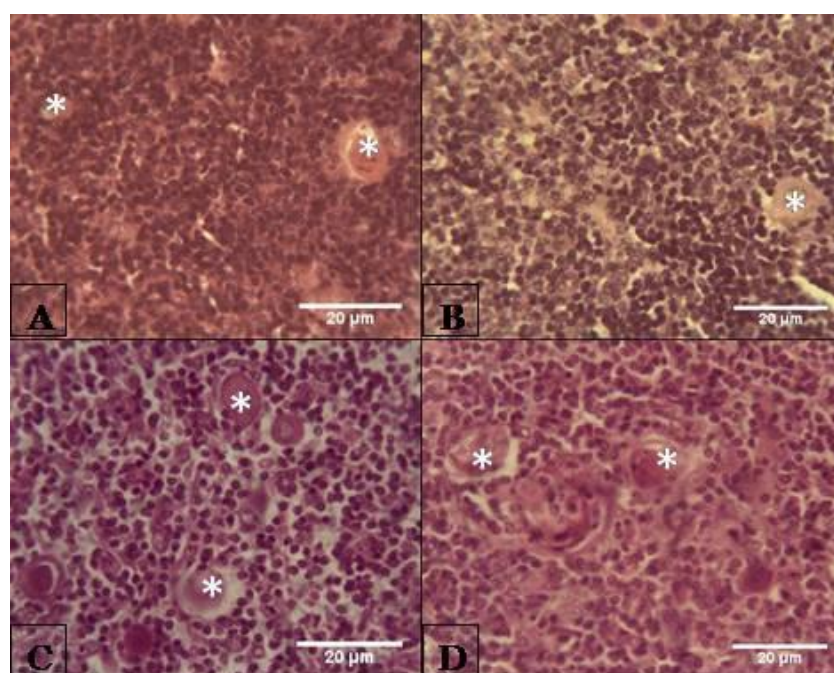
**Figure 1** Photomicrographs of the mesenteric lymph node of 1-day-old (A x100, B x400), 2-week-old (C x40), and 2-month-old (D x100) West African Dwarf (WAD) goats showing the cortex (c), cortex (Cx), medulla (M) and Medullary cords (asterisks). Note the unorganized medulla of 1-day-old lymph node (B). H & E stain.



**Figure 2** Photomicrographs of the mesenteric lymph node of 3-month-old (A x100), 4-month-old (B x40) and 7-month-old (C x400) West African Dwarf (WAD) goats showing cortex (Cx), medullary cord (Mc), and medullary sinus (S). Note the lymphocytes (arrows) and lymphoid follicles (asterisks). H & E stain.



**Figure 3** The thymus of 1-day-old (A), 2-week-old (B), 3-month-old (C) and 5-month-old (D) WAD goats showing thymic cortex (C), thymic medulla (M), and adipose tissue deposition (asterisk). H & E stain. x100.

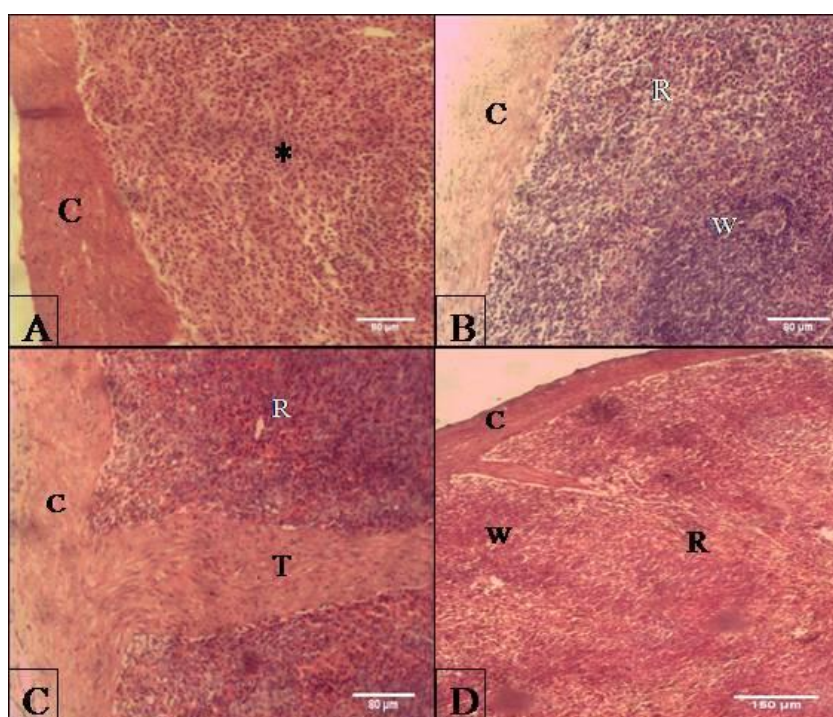


**Figure 4** Photomicrographs of the thymic medulla of 1-day-old (A), 2-week-old (B), 5-month-old (C) and 6-month-old (D) West African Dwarf (WAD) goats showing Hassal's corpuscles (asterisks). H & E stain. x400.

**Histology of WAD goat spleen:** The spleen of the WAD goats showed a dense connective tissue capsule with a splenic parenchyma which demonstrated white and red pulp. The white pulp contained lymphoid follicles and peri-arteriolar lymphoid sheath while the red pulp was made up of blood sinusoids and splenic cords.

The spleen of 1-day-old goats showed splenic parenchyma with poorly defined red pulp, white pulp areas. The entire splenic parenchyma appeared as white pulp (Fig. 5A). At 2 weeks, the red and white pulp areas were clearly defined and the splenic sinusoids contained red blood cells with demarcated

splenic cords (Fig. 5B). At 1 to 7 months postnatum, the red pulp areas became more expansive and there was increased concentration of red blood cells in the splenic sinusoids while the white pulp areas were characterized by dense lymphocytic infiltration, lymphoid nodules and peri-arteriolar lymphoid sheath (Figs. 5C, D). The splenic cords of the red pulp areas were clearly demarcated. At 1 day old, 2 weeks, 1 to 7 months old, thick capsules were observed and the trabeculae extended from the capsule deep into the splenic tissues (Fig. 5C).



**Figure 5** The spleen of 1-day-old (A, x100), 2-week-old (B, x100), 1-month-old (C, x100) and 3-month-old (D, x40) WAD goats showing unorganized splenic parenchyma (asterisk), capsule (C), white pulp (W), red pulp (R) and trabecula (T). H & E stain.

### Discussion

In the present study, the poorly organized cortical and medullary parenchymal areas in the lymph node of 1-day-old WAD goats suggest that further organization of the lymph node tissues occurs shortly after birth, probably due to minimal involvement of these nodes in immune response. This is further evidenced by few lymphocytes which infiltrated the cortex and the unorganized medullary area which was devoid of lymphocytes or reticular cells. Progressive increase in the concentration of lymphocytes in the cortex as reported in this study from 2-week-old to 7-month-old lymph nodes may represent increased participation of the nodes in the defense of the body against invading pathogens, most probably due to age and level of exposure to environmental factors (Parker et al., 2015). The 2-week-old and 1-month-old lymph nodes contained dense infiltration of lymphocytes, 2- and 3-month-old lymph nodes exhibited dense infiltration of lymphocytes with few lymphoid nodules while large population of lymphoid nodules were observed in the cortex of 4- to 7-month-old mesenteric lymph nodes. Following antigenic insults, the densely infiltrated lymphocytes often respond by intense multiplication and differentiation which often results in the formation of primary and secondary lymphoid nodules, structural modifications representing the production of antibodies (Zidan and Pabst, 2010; Parker et al., 2015; Udoumoh and Ezeasor, 2015). The report of this study is supported by the observations of previous authors (Kutyrev et al., 2008; Parker et al., 2015) in the mesenteric lymph nodes of Baikal Seal and rat, respectively. Therefore, the mesenteric lymph nodes of WAD goat have substantial exposure to antigens early

in life and so assumes adult morphology early in post-natal life even at 1 month old.

Moreover, the structural features of the thymus of 1-day-old WAD goats in the present study further demonstrate that the thymus may be fully functional in early post-natal life. This observation is in agreement with the reports of functional thymus in late pre-natal life and early post-natal life (Gui et al., 2012; Gillard and Farr, 2016). Although the cortex and medulla of 1-day-old thymus were well delineated, evidence of thymic regression; distorted cortical and medullary outlines, deposition of adipose tissue in the trabeculae; were observed in the thymus of 3-month-old WAD goats. Furthermore, the loss of the cortical tissues or decrease in the population of thymocytes were more pronounced in the 5- to 7-month-old thymus. Thymic regression could be triggered by age, environmental stress and nutritional limitations (Anderson et al., 1974; Lawrence et al., 1986; Dobson et al., 2010). However, in the present study the observed regressive changes in the thymic tissues may be due to age. The present study disagrees with the report of depopulation of thymocytes in the cortex of 2- to 6-month-old thymus of red deer (Dobson et al., 2010). The population of the Hassal's corpuscles increased from 1-day-old thymus to 7-month-old thymus, suggesting the changes to be age-related. Thus, since thymocyte population wanes rapidly following sexual maturity in all vertebrates (Teh, 1993; Shortman et al., 1998), WAD goat which may attain sexual maturity early in life has fully functional thymus at birth and their thymus could regress as early as 3 months postnatum.

In the present study, the most obvious change in the structure of the spleen were the gradual increases in the red pulp areas with increase in the

concentration of red blood cells from 2-week-old to 7-month-old spleen. Such changes may be a function of the evolving circulatory system and physiologic adaptations as the animal ages. Although the spleen of 1-day-old WAD goats in the present study appeared morphologically mature, there were limited red pulp areas, validating the popular hypothesis that the spleen contains limited reserves of hematopoietic cells into early post-natal life (Landreth, 2012). Similarly, the lymphoid functions of the 1-day-old spleen may be reduced as the white pulp areas were devoid of lymphoid nodules and periarteriolar lymphoid sheath (PALS). This observation is similar to that reported in golden hamster (*Mesocricetus auratus*) (Gomariz et al., 1989).

Finally, the present study has demonstrated that the mesenteric lymph nodes, thymus and spleen of WAD goats undergo further morphological modifications that could influence their physiology shortly after birth. While the mesenteric lymph nodes of WAD goats may attain adult morphology as early as 1 month after birth, the thymus which appears as fully functional at birth, could regress at 3 months after birth and the spleen could have limited lymphoid functions immediately after birth.

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## บทคัดย่อ

### การพัฒนาหลังคลอดของอวัยวะ lymphoid บางชนิดในแพะแคระแอฟริกาตะวันตก: มุมมองทางจุลพยาธิวิทยา

อนิเต ฟรานซิส อูโดโม<sup>1,2\*</sup> คลิฟฟอร์ด วาบัสวู อบิอูส<sup>1</sup>

การศึกษานี้ได้ประเมินการเปลี่ยนแปลงพัฒนาการของต่อมน้ำเหลือง mesenteric ต่อมไขมัน และม้าม ของแพะแคระแอฟริกาตะวันตก (WAD) โดยใช้เทคนิคทางจุลพยาธิวิทยา โดยศึกษาในแพะ WAD 27 ตัว ที่อายุ 1 วัน, 2 สัปดาห์ และ 1 ถึง 7 เดือน ผลการศึกษาพบว่า พัฒนาการของต่อมน้ำเหลือง mesenteric ในวันที่ 1 มีการเจริญของ cortical และ medullary ไม่เต็มที่ และในเดือนที่ 2 และ 3 มีการแทรกของเซลล์เม็ดเลือดขาวในเนื้อเยื่อชั้นนอก และพบ lymphoid follicles พัฒนาการของต่อมไขมัน ในวันที่ 1 พบเนื้อเยื่อ cortical parenchyma และ medullary บาง และในเดือนที่ 3 และ 4 พบเส้นโครงร่างระหว่าง cortical และ medullary และการสะสมของเนื้อเยื่อไขมันใน trabeculae นอกจากนี้พบการเพิ่มจำนวนของ Hassal's corpuscles เพิ่มขึ้นจากวันที่ 1 ถึงเดือนที่ 7 หลังเกิด พัฒนาการของม้าม ในวันที่ 1 พบบริเวณ red และ white pulp โดยพบขอบเขตมีความชัดเจนขึ้นเมื่ออายุ 2 สัปดาห์ และบริเวณ red pulp เริ่มขยายตัวมากขึ้นและมีเม็ดเลือดแดงเพิ่มขึ้นในเดือนที่ 1 ถึง 7 โดยสรุปการศึกษานี้พบว่า ต่อม้ำเหลือง mesenteric ของแพะ WAD เจริญเต็มที่ ได้ตั้งแต่ 1 เดือนหลังคลอด ส่วนต่อมไขมันมีพัฒนาการเต็มที่หลังคลอด และ ลดลงเมื่ออายุ 3 เดือน ส่วนม้ามสามารถทำหน้าที่ lymphoid ได้จำกัดทันทีหลังคลอด

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**คำสำคัญ:** พัฒนาการ จุลพยาธิวิทยา อวัยวะ lymphoid หลังคลอด แพะแคระแอฟริกาตะวันตก

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