

## Research Article

## Development and Maturation of the Dromedary Spleen: Anatomical and Histological Analysis During the First Three Years of Life



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**ABSTRACT**

**Background:** The dromedary spleen plays a crucial role in the immune system and maintaining homeostasis. However, there is limited research on the developmental changes in the spleen during the first few years of a dromedary's life.

**Objectives:** The objective of this study was to comprehensively investigate the developmental changes in the dromedary spleen during the crucial first three years of life. This aim was achieved through a multi-faceted approach involving macroscopic examination, which entailed measuring the absolute and relative mass of the spleen and morphometric analysis. Additionally, histological and histomorphometric analyses were employed to study the cellular composition of the spleen at different ages.

**Methods:** Five groups of dromedaries were examined in Southeastern Algeria. Their spleens were analyzed using both gross anatomy and histological examination. The cellular composition of their spleens was studied on histological slides of different ages, stained with hematoxylin and eosin.

**Results:** The results showed that the spleen's size, weight, and volume significantly increased as the animals grew. The splenic mass of the animal of the first group showed an average absolute mass of 251±14.19 g, whereas in the fourth group, they showed an average mass of 318±23.91 g. Also, the length, width, and thickness showed average values of 43.78±1.95 cm, 19.44±1.52 cm, and 1.88±0.16 cm, respectively, at 8 months. However, they showed average values of 49.6±1.86 cm, 24.32±1.69 cm, and 3.18±0.34 cm, respectively, at the age of 3 years. The cellular composition of the spleen changed over time, with a higher percentage of lymphoid tissue at 8 months, more red pulp at 1 year, and a higher percentage of white pulp at 2 years. Finally, the spleen had a mature mixture of red and white pulp and fully developed immune function by 3 years.

**Conclusion:** This study provides new insights into the development and maturation of the dromedary spleen. The findings have important implications for understanding the health and well-being of these animals. The results could potentially contribute to the development of better strategies for the management and care of dromedaries.

**Keywords:** Cytology, Dromedary, Histology, Spleen, Maturation

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## Introduction

The dromedary, or one-humped camel (*Camelus dromedarius*), is an important livestock animal in dry, arid regions of the world (Nagy et al., 2020). Dromedaries are adapted to survive in harsh environments with limited access to water and are used for various purposes, including transportation, milk production, and meat (Ho et al., 2022). Their unique physiology allows them to survive prolonged periods without water, tolerate high body temperatures, and maintain their body water balance. In addition to their physiological adaptations, dromedaries have a complex immune system that helps them combat disease and preserve health (Gossner et al., 2016; Hajinejad-Bamroud et al., 2020; Mohamed Amine et al., 2023; Fares et al., 2023c).

The spleen is a vital organ in the immune system of all mammals, including dromedaries. It functions as a blood filter, storing red blood cells, releasing them when necessary, and storing and activating immune cells. The spleen also removes senescent erythrocytes, or red blood cells that are no longer functional. In addition, the spleen is involved in the immune response to infections, and it contains various immune cells such as T cells, B cells, and macrophages (Mebius & Kraal, 2005).

Dromedaries' first three years of life are critical for their overall development and productivity. During this period, they undergo significant physiological changes that affect their growth and immune system. Understanding the changes during this period is essential for dromedaries' production and management practices. Also, it can help identify potential health problems and implement appropriate interventions.

One crucial aspect of dromedary development during the first 3 years of life is the maturation of the spleen, a vital organ in the immune system of all mammals. The spleen is crucial in blood filtering and activating immune cells to combat infections. However, little is known about the development and maturation of the dromedary spleen during this critical period (Fares et al., 2023a; Fares et al., 2023b).

The development and maturation of the dromedary spleen are poorly understood with the current state of knowledge, and more research is needed to fully understand the animals' immune function (Hussen & Schuberth, 2021). The findings of this study contribute to our understanding of the development of the drom-

edary spleen and may have practical implications for the management and care of these animals. Also, understanding the changes in the spleen during the first 3 years of life may help veterinarians identify potential health problems and implement appropriate interventions. Additionally, this information may be helpful for researchers studying the immune function of dromedaries and developing vaccines and other preventive measures to protect these animals from diseases.

Despite the importance of the spleen in the immune system of dromedaries, few research studies were carried out on the development and maturation of this organ (Lewis et al., 2021). In particular, there is a lack of information on the changes in the gross and microscopic anatomy, size, and cellular composition of the dromedary spleen during the first 3 years of life. This gap limits our understanding of the immune function of these animals and hinders the development of effective management strategies for dromedary health. However, there have been some studies conducted on the postnatal development of the spleen in other domestic animals, such as rabbits (Rahmoun et al., 2019) and lambs (Rahmoun et al., 2020), which may provide some insights into the developmental changes that occur in the dromedary spleen.

## Materials and Methods

This study selected 25 healthy dromedary camels and divided them into 5 age groups (8 months, 1 year, 18 months, 2 years, and 3 years), consisting of 5 animals per group. The animals were sourced from a single herd to control for extraneous variables that might affect the study's outcome (Cero et al., 2021). The animals were humanely slaughtered in adherence to Algerian Islamic customs and in accordance with stringent sanitary protocols under the supervision of licensed veterinarians. The ages of the dromedaries were obtained through a survey of their owners. The body weight of the dromedaries was determined by using a specific equation, which takes into account various physical measurements of the dromedaries to estimate their body weight, according to some studies (Kamili et al., 2006; Gherissi et al., 2022). Their spleens were meticulously collected and subjected to both morphometric and mass analysis. The gross anatomy of the spleen was carefully documented through detailed descriptions and photographs taken with a digital camera. The size and weight of the spleen were also measured using established techniques to ensure accurate and precise data collection.

For the microscopic study, the spleen fragments were fixed in formalin and processed for paraffin embedding. Five-micrometer sections were cut and stained with hematoxylin, eosin, and Masson trichrome staining techniques to highlight the spleen's various cell types and tissues. The stained sections were examined under a B382PLi-ALC light microscope at x400 magnification (Iezzoni, 2018).

The statistical results were expressed as Mean±SD of the means. Also, a one-way analysis of variance (ANOVA) was performed to compare the differences between the different age groups. Additionally, the Pearson correlation analysis was conducted to evaluate the relationship between the various physical factors of the spleen. The statistical analysis was performed using R software, version R 4.3.0 (Posit, PBC, USA, 2011-2023). Finally, we could determine if there were any significant differences between the groups and identify any potential correlations between the various physical characteristics of the spleen.

The equation of estimation of the dromedary's body weight according to some studies (Kamili et al., 2006) is as follows (Equation 1):

$$1. \text{Body weight (kg)} = 4.06 \times \text{age (y)} + 3.05 \times \text{circumference of the neck (cm)} + 3.38 \times \text{circumference of the thigh (cm)} + 1.38 \times \text{Hump length (cm)} - 191.$$

**Table 1.** Morphometric Mean±SD values of the dromedary spleen across different age groups

Age	Group (n)	Mean±SD			
		Mass (g)	Length (cm)	Width (cm)	Thickness (cm)
8 months	Groupe 1 (5)	251±14.19	43.78±1.95	19.44±1.52	1.88±0.16
1 year	Groupe 2 (5)	267±12.59	45.7±1.42	19.8±1.69	2.24±0.29
18 months	Groupe 3 (5)	280±21.02	46.2±1.62	21.7±1.78	2.58±0.24
2 years	Groupe 4 (5)	297±12.98	47.42±1.46	22.1±1.99	2.89±0.32
3 years	Groupe 5 (5)	319.8±21.91	49.6±1.86	24.32±1.69	3.18±0.34

**Table 2.** Analysis of variance for the mass of the dromedary spleen across different age groups

Source of Variation	Sum of Squares	df	Mean Square	F	P
Age groups	14196.1600	4	3549.0400	11.5064	5.1841e-05
Error	6168.8000	20	308.4400		
Total	20364.9600	24			

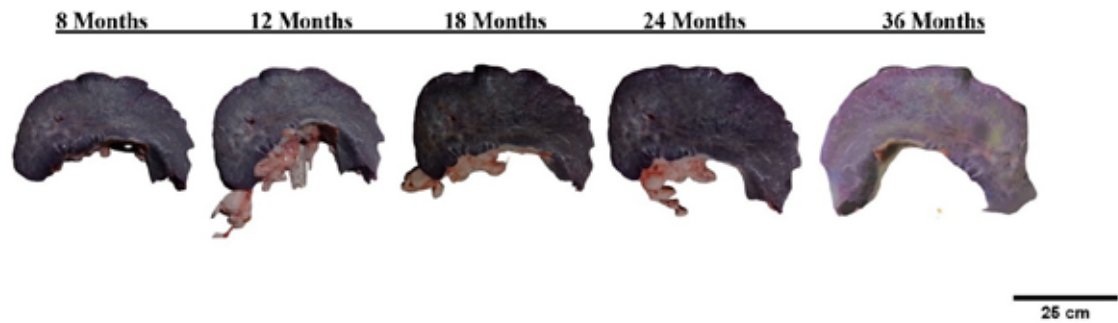
F and P<0.001: Significant difference in the splenic mass between the age groups.

## Results

Our investigation into the characteristics of the one-humped camel's spleen has revealed several intriguing details about this organ. Notably, we discovered its soft texture, colored in a distinctive red-brown shade (Figure 1). In addition, the spleen of camels possesses a concave surface, which represents a unique and previously unknown feature of this anatomical structure.

Our results also revealed that the spleen of the one-humped camel is located under the transverse processes of the lumbar vertebrae and extends from the upper caudal end of the dorsal sac of the rumen to the caudal end of the left kidney. It has two surfaces: A convex parietal surface in contact with the internal obliques muscle and the subcutaneous region and a concave visceral surface in contact with the upper-caudal surface and the left face of the dorsal sacrum for the rumen. The dorsal end of the spleen attaches to the upper surface of the dorsal sac of the rumen, while the caudal edge wraps around.

The results of this study revealed a range of variation in the physical characteristics of dromedaries within different age groups; the dromedaries in the first group, which were 8 months old, presented with splenic masses of 251±14.19 g and body weights of 339.4±9.24 kg, whereas the relative mass of the spleen in the animals of this group was 0.0738%±0.0036%, with a splenic



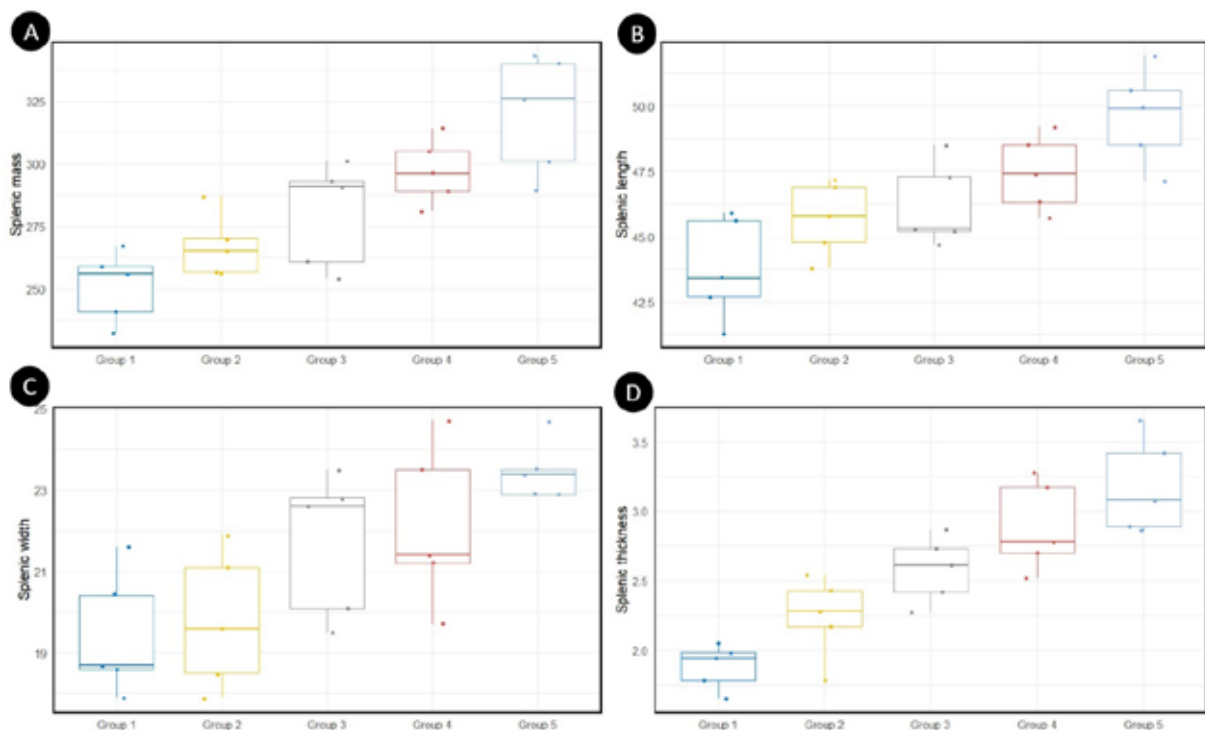
**Figure 1.** Spleen morphology according to age groups

length of  $43.78 \pm 1.95$  cm. The width of their spleen was  $19.44 \pm 1.52$  cm, with a splenic thickness of  $1.88 \pm 0.16$  cm (Figure 2).

On the other hand, in the dromedaries of the second group (1 year old), the splenic mass was  $267 \pm 12.59$  g, and the body weight was  $364.2 \pm 9.36$  kg. The relative mass of their spleen was  $0.0732\% \pm 0.0043\%$ , with a mean splenic length of  $45.7 \pm 1.42$  cm, width of  $19.8 \pm 1.69$  cm, and a thickness of  $2.24 \pm 0.29$  cm (Figure 2). Similarly, the dromedaries of the third group (18 months old) had a splenic mass of  $280 \pm 21.02$  g with a mean body weight of  $368.5 \pm 5.31$  kg, so the relative mass of their spleen

was  $0.0774\% \pm 0.0083\%$ , with a mean splenic length of  $46.2 \pm 1.62$  cm, width of  $21.7 \pm 1.78$  cm, and a thickness of  $2.58 \pm 0.24$  cm (Figure 2).

The dromedaries in the fourth group, which were two years old, presented with a splenic mass of  $297 \pm 12.98$  g with a body weight of  $404.6 \pm 10.41$  kg, so the relative mass of their spleen was  $0.0736\% \pm 0.0021\%$ . The mean splenic length in the animals of the fourth group was  $47.42 \pm 1.46$  cm, with a width of  $22.1 \pm 1.99$  cm and thickness of  $2.89 \pm 0.32$  cm. Finally, the dromedaries in the fifth group (three years old) had a splenic mass of  $318 \pm 23.91$  g and a body weight of  $426.2 \pm 5.4$  kg. The relative mass



**Figure 2.** Dynamics of A) The splenic mass, B) Length, C) Width, D) Thickness according to age groups

**Table 3.** Analysis of variance for the length of the dromedary spleen across different age groups

Source of Variation	Sum of Squares	df	Mean Square	F	P
Age groups	92.8840	4	23.2210	8.2438	0.0004
Error	56.3360	20	2.8168		
Total	149.2200	24			

F and P<0.001: Significant difference in the splenic length between the age groups.

**Table 4.** Analysis of variance for the width of the dromedary spleen across different age groups

Source of Variation	Sum of Squares	df	Mean Squares	F	P
Age groups	77.4104	4	19.3526	6.3976	0.0017
Error	60.5000	20	3.0250		
Total	137.9104	24			

F and P<0.001: Significant difference in the splenic width between the age groups.

**Table 5.** Analysis of variance for the thickness of the dromedary spleen across different age groups

Source of Variation	Sum of Squares	df	Mean Square	F	P
Age groups	5.2916	4	1.3229	16.8587	3.4167e-06
Error	1.5694	20	0.0785		
Total	6.8610	24			

F and P<0.001: Significant difference in the splenic thickness between the age groups.

of their spleen comprised  $0.0748\pm 0.0058\%$ . Their mean splenic length was  $49.6\pm 1.86$  cm, with a width of  $24.32\pm 1.69$  cm and a thickness of  $3.18\pm 0.34$  cm (Table 1) (Figure 2).

To further analyze the differences between the groups, a one-way ANOVA was performed. The results of the ANOVA showed a significant difference between the groups in terms of splenic mass (P<0.001), relative mass of the spleen (P<0.001), splenic length (P<0.001), splenic width (P=0.017), and thickness (P<0.001) (Tables 2, 3, 4 and 5). In all tables, the P<0.05, indicating a significant difference in the dromedary spleen's mass, length, width, and thickness across different age groups.

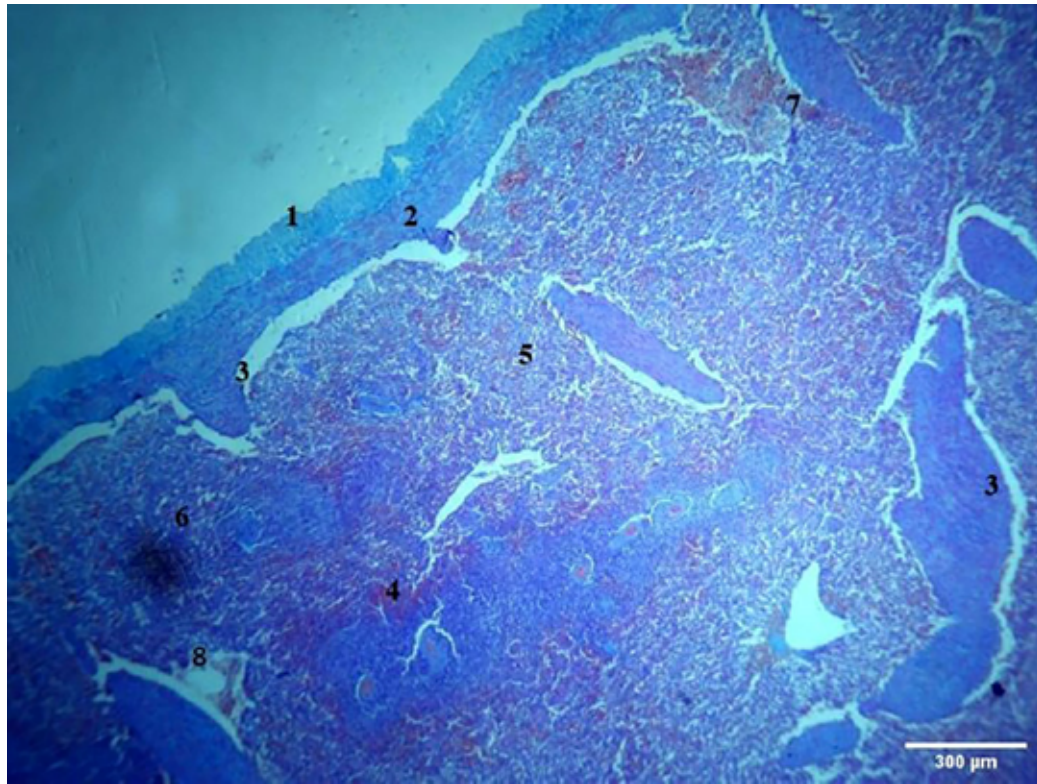
Regarding the histological analysis of the spleen of the dromedary (*C. dromedarius*), our results revealed a unique structure characterized by the presence of white pulp and red pulp. The white pulp was composed of lymphoid follicles, densely packed with lymphocytes, macrophages, and dendritic cells and surrounded by a dense network of reticular fibers, forming the framework for the follicles and providing structural support (Figure 3).

On the other hand, the red pulp of the spleen was composed of large sinusoids and thin-walled blood vessels lined with macrophages and reticular cells. It is also rich in blood vessels and is responsible for maintaining blood volume and blood pressure.

Additionally, we found that the splenic capsule is a thin layer of fibrous tissue that surrounds the spleen, separates it from the surrounding organs, and projects trabecular tissue toward the inner layers of the organ (Figure 4).

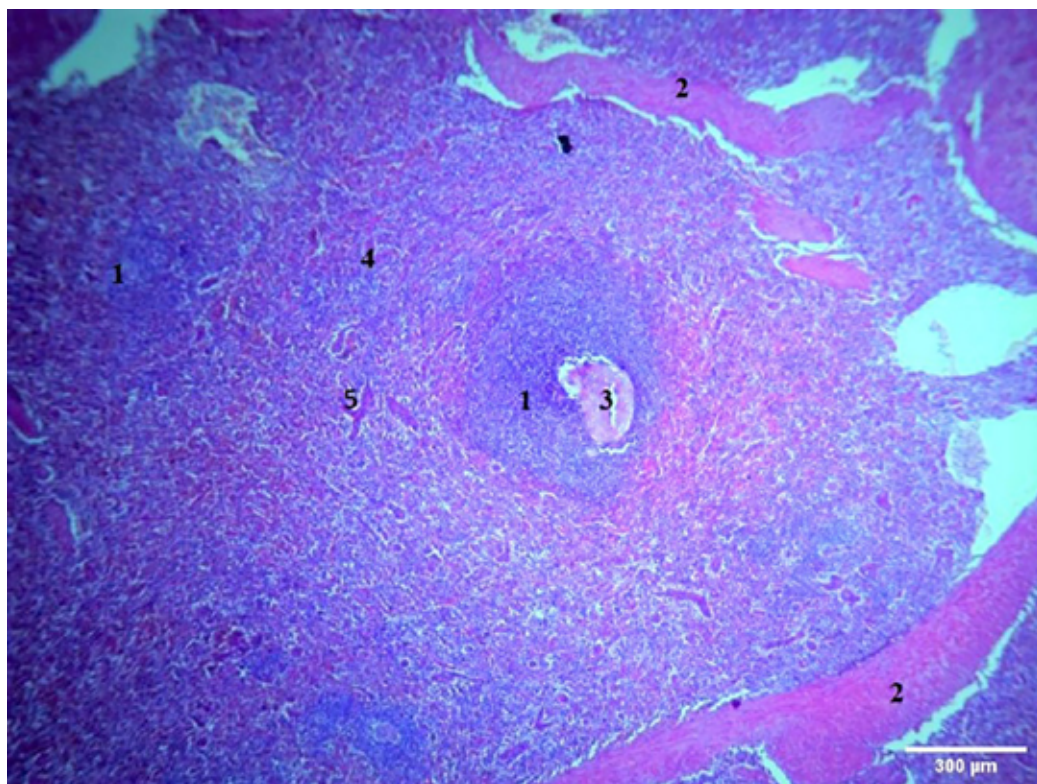
The cellular composition of the spleen changes significantly over time regarding cellular populations and density. At 8 months, the spleen contains mostly lymphocytes and a small number of erythrocytes and macrophages. In contrast, at 1 year of age, the number of erythrocytes increases, but the number of lymphocytes decreases. At 2 years of age, the spleen has a higher rate of macrophages and a lower number of erythrocytes, while at 3 years, the spleen contains the highest rate of lymphocytes, suggesting that the immune function of the spleen is fully developed and functional then in carrying out its various roles in the immune system.



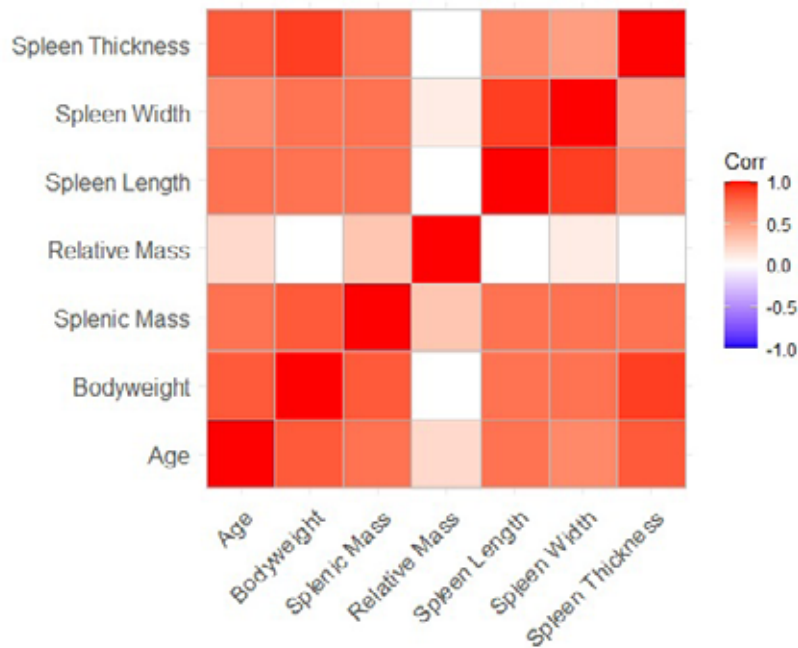


**Figure 3.** Histological section of the spleen in dromedary during the first three years of life (x40 Masson trichrome staining)

1) Capsule, 2) Mesothelium (smooth muscle bundles), 3) Trabeculae, 4) Red pulp, 5) White pulp, 6) Lymphatic follicle, 7) Periarterial zone, 8) Splenic ellipsoids



**Figure 4.** Histological section of the parenchyma of the spleen in dromedary during the first three years of life (x40 Hematoxylin and Eosin staining), 1) White pulp (mantle zone), 2) Trabeculae, 3) Pulp artery, 4) Red pulp, 5) Sinusoidal frames



**Figure 5.** Correlogram of age and morphometric parameters

Our results revealed that the spleen undergoes developmental changes in its structure and immune function over time. At 8 months, the spleen mainly comprises lymphoid tissue and a sparse population of splenic cords. In contrast, at 1 year, the number of splenic cords increases, and the spleen contains more red pulp, which is important for removing senescent red blood cells and storing iron. At two years of age, the spleen contains a higher ratio of white pulp, and the splenic cords become more prominent. By 3 years of age, the spleen presents a mature mixture of red and white pulp and has fully developed its immune function,

Our statistical results revealed a moderate to strong positive correlation between age and various physical characteristics of the spleen in one-humped camels. As the age of the animals increased, there was a corresponding increase in the spleen's thickness, length, and mass. These findings suggest a physiological relationship between the dromedaries' age and their spleen's development.

Additionally, there was a moderate positive correlation between the width of the spleen and age, indicating that as the dromedary ages, there may also be a slight increase in the width of their spleen. Furthermore, our study found a strong correlation between splenic mass and its thickness, width, and length. This finding suggests a strong relationship between the size and dimensions of the spleen and its overall mass. Also, we observed a strong positive correlation between the camels' body weight and the splenic mass, indicating that as the

body weight of the camels increases, so does the mass of their spleen (Figure 5).

Our findings indicate obvious correlations between dromedaries' age and their spleen's physical characteristics. Also, the correlations between splenic mass and its thickness, width, and length, as well as the correlation between the body weight of the dromedaries and the mass of their spleen, suggest strong relationships between these factors.

## Discussion

The one-humped camel's spleen is a vital organ that has been the subject of numerous studies. However, our recent research has revealed a surprising discrepancy in earlier reports regarding the anatomy of the spleen.

Specifically, we found that our observations did not show the groove between the dorsal end of the spleen and the body, as described in previous studies (Zidan et al. (2000a); Nawal & Maher, 2018). This unexpected finding raises important questions about the structure and function of the one-humped camel's spleen and highlights the need for further investigation.

Our results on the anatomy of the dromedary's spleen have yielded some intriguing findings. Perhaps the most fascinating is the discovery of the concave surface of the spleen, unlike the convex surface typically found in other mammals such as cows, sheep, and horses.

This unique characteristic of the camel's spleen sets it apart from other animals. It underscores the importance of comparative anatomy in understanding this organ's evolutionary history and functional significance. Previous studies (Chadburn, 2000, Khalel, 2010; Maina et al., 2014; Nawal & Maher, 2018) have also highlighted the distinctive features of the camel's spleen, including its coloration and texture.

Moreover, our study revealed that the shape of the camel's spleen is distinct from other animals. Cows have an elongated and elliptical spleen, while sheep have an almost round shape. Dogs have a sickle-shaped spleen, and cats have a broad, curved, flattened, and elongated one (Hassankhani et al., 2017; Wang et al., 2021).

Also, Jaji et al. (2019) conducted a study on the anatomical and histological characteristics of the spleen in one-humped camels. They reported similar observations regarding the location and surface structure of the camel's spleen. The concave surface of the spleen is particularly noteworthy, as it differs from the convex surface typically found in other mammals. Sty and Conway (1985) studied the histology of the camel's spleen and reported that the spleen undergoes a significant increase in size and mass as the animal grows. Similarly, Kamath et al. (2000) investigated the gross anatomy of the camel's spleen and found that the organ's size and weight increased with age.

Our results about the morphometric variables provide further evidence of the changes in the one-humped camel's spleen as the animal grows. The significant differences in the mass, length, width, and thickness of the spleen across different age groups suggest that the organ undergoes structural adaptations to meet the needs of the growing animal.

Regarding the histological structure of the dromedary's spleen, Zidan et al. (2000b) reported that the red pulp of the camel's spleen is composed of sinusoids lined with macrophages and reticular cells. Our study supports this finding, indicating that the red pulp of the spleen in one-humped camels is also composed of sinusoids and rich in blood vessels, suggesting that the spleen plays a vital role in regulating blood volume and pressure.

Furthermore, our study found that the splenic capsule is a thin layer of fibrous tissue that separates the spleen from surrounding organs and sends trabecular tissue toward the inner layers of the organ. These results are consistent with previous research (Zidan et al., 2000a; Zidan et al., 2000b), indicating that the structure of the splenic capsule in one-humped camels is similar to that of other mammalian species.

The results of our study also showed that the red pulp of the spleen is composed of sinusoids that are rich in blood vessels and responsible for maintaining blood volume and pressure, which is consistent with the function of the red pulp reported by Fares et al. (2023a). Furthermore, our study found that the splenic capsule is a thin layer of fibrous tissue that surrounds the spleen and separates it from the surrounding organs, which agrees with the results reported by Fares et al. (2023a).

Finally, the correlations observed in our study between the physical characteristics of the spleen and the age and body weight of the dromedaries are consistent with the results reported by Brendolan et al. (2007) and Burn et al. (2008). This finding suggests that the physical changes in the spleen of dromedaries over time are not unique to our study. Still, they have been widely observed and reported in the scientific literature. These correlations can provide essential insights into the development of the spleen and its functions in the dromedary, as well as potential implications for the health and well-being of the animal.

Furthermore, the relationships between splenic mass, thickness, width, length, and body weight of the dromedaries are significant, indicating that as the dromedary grows, its spleen also grows proportionally. This finding is consistent with the results of previous studies that have investigated the correlation between body weight and splenic mass in other animal species (Brendolan et al., 2007; Wang et al., 2021). Our findings may have implications for veterinary medicine and animal welfare, particularly in relation to the diagnosis and treatment of diseases that affect the spleen, as well as the potential effects of physiological changes associated with growth and development.

## Conclusion

The dromedary spleen undergoes significant changes in size, weight, and cellular composition during the first 3 years of life. These changes are important for understanding the immune function of the dromedary and may have practical implications for the management and care of these animals. Understanding the changes in the spleen's size and weight may help develop body weight standards for dromedaries, which are important for the evaluation of animal health and well-being. In this study, the size and weight of the spleen increased significantly between eight months and three years of age, as determined by statistical analysis. Specifically, the mean size of the spleen and the splenic mass increased with age, which concludes that the dromedary spleen undergoes significant growth and development during the first three years of life.



In addition, understanding the changes in the cellular composition of the spleen may help identify potential health problems or changes in immune function that could impact the health of the dromedary. In this study, the cellular composition of the spleen changed significantly over time. At 8 months, the spleen contained mainly lymphocytes and a small number of erythrocytes and macrophages. At 1 year, the number of erythrocytes increases, and the percentage of lymphocytes decreases. By 2 years of age, the spleen contains a higher percentage of macrophages, and the number of erythrocytes decreases. At 3 years, the spleen contains a mixture of all three cell types, with the highest percentage of lymphocytes. These changes in cell composition may be due to the different functions of the spleen at different ages. For example, the increase in erythrocytes at 1 year of age may be related to the increased storage of red blood cells in the spleen at this age. In contrast, the increase in macrophages at 3 years of age may be related to the increased immune function of the spleen at this age.

## Ethical Considerations

### Compliance with ethical guidelines

There were no ethical considerations to be considered in this research.

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### Authors' contributions

Conceptualization, supervision and methodology: Fares Mohamed Amine, Khenenou Tarek and Rahmoun Djallal Eddine; Data collection: Fares Mohamed Amine, Derradji Harek, Houari Hemida and Rabah Mayouf; Investigation, data analysis and writing: Fares Mohamed Amine.

### Conflict of interest

The authors declared no conflict of interest.

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