

Changes in Thymus and Spleen Morphology Under Stress in Early and Adult Generations

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Received: 30 Jan 2026 | Received Revised Version: 14 Feb 2026 | Accepted: 26 Feb 2026 | Published: 17 Mar 2026

Volume 08 Issue 03 2026 | Crossref DOI: 10.37547/tajmspr/Volume08Issue03-11

Abstract

The article studied the morphological and morphometric characteristics of the thymus organ of 3- and 21-day old offspring born from mother rats exposed to chronic stress and compared them with offspring in the control group, and developed an appropriate assessment criterion. The expected results showed that the thymus of the offspring in the experimental group showed profound and significant morphological changes compared to those in the control group.

Keywords: Thymus, spleen, stress, morphology, morphometry, Hassall bodies.

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Cite This Article: Normuradov Aliyor Doniyor o'g'li, & Axmedova Sayyora Muhamadovna. (2026). Changes in Thymus and Spleen Morphology Under Stress in Early and Adult Generations. *The American Journal of Medical Sciences and Pharmaceutical Research*, 8(03), 132–136. <https://doi.org/10.37547/tajmspr/Volume08Issue03-11>

1. Introduction

Chronic maternal stress during pregnancy leads to significant structural and functional changes in the immune organs of the offspring. Studies have shown that prolonged stress in pregnant rats causes a noticeable reduction in the white pulp area of the spleen in their offspring. This reduction is associated with a lower density of lymphocytes and a decrease in the size of lymphoid follicles, indicating suppression of normal immune development. At the same time, the red pulp region of the spleen tends to increase in size, which may reflect compensatory hematopoietic activity aimed at maintaining blood cell production under stress conditions.

In addition to the spleen, prenatal stress also affects the

morphology of the thymus, one of the primary immune organs responsible for T-lymphocyte maturation. In offspring exposed to maternal stress, the thymus shows structural alterations such as a reduction in cortical thickness and disturbances in the normal distribution of lymphocytes between the cortex and medulla. These morphological changes suggest impaired thymocyte proliferation and differentiation.

Overall, these findings demonstrate that prenatal exposure to chronic stress can have long-lasting effects on the development and structural organization of immune organs in rat offspring. Such alterations may lead to weakened immune responses and increased susceptibility to various diseases later in life, highlighting the importance of maternal physiological and psychological stability during

pregnancy.

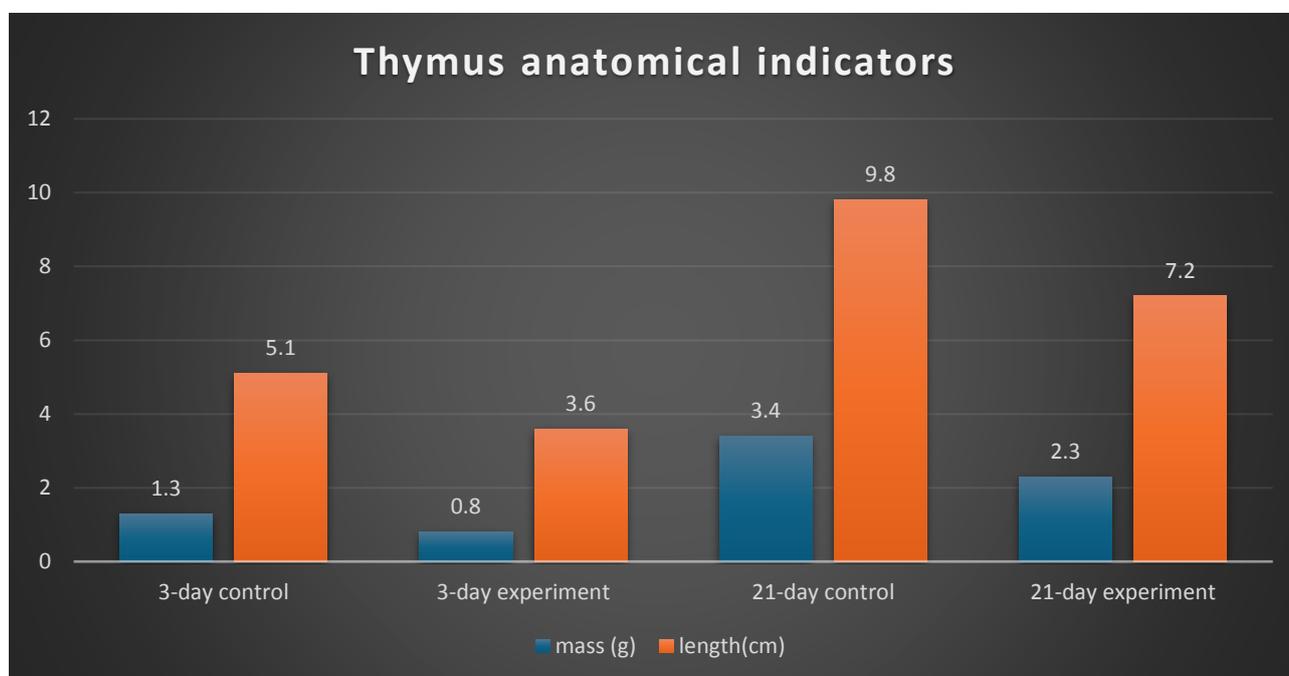
2. Methods

To achieve this goal, 40 albino laboratory rats weighing 160–170 grams and 80 offspring were used. The rats were divided into two groups. Group 1 consisted of 20 healthy rats serving as a control group. Pregnant rats in the control group were injected with 1.0 ml of saline solution into the stomach every morning. A subclavian catheter was used as a probe. The second group was experimental. 20 pregnant albino laboratory rats were kept in specially prepared maze cages to create experimental stress. Several methods were used to obtain results: general histological, organometric, and morphometric methods. After opening the chest and separating the thymus, the anatomical parameters of the organ were determined. Thymus size was measured with calipers. Electronic scales were used to measure thymus weight in rats. Histological preparations. Sections 8-10 μm thick, prepared on a rotary microtome, were stained with hematoxylin and eosin. Collagen fibers in the connective tissue were determined using the van Gieson method, and elastic fibers were determined using the Weigert method.

3. Results

On the third day, the boundaries between the parenchymal fragments in the histological preparation were very poorly defined due to severe thinning of the connective tissue septa. Adipose tissue adhered very closely to the organ surface externally, in places growing into the thymus parenchyma. In the fragments, the boundary between the cortex and medulla was blurred, a consequence of cortex illumination. This phenomenon is associated, on the one hand, with a less dense arrangement of lymphocytes and, on the other hand, with less intense staining of their nuclei. Next, the average values of the anatomical parameters of the thymus in 3-day-old individuals were studied. Accordingly, in the control group, the average thymus weight in 3-day-old individuals was 1.3 ± 0.5 g; length – 5.1 ± 0.9 cm; By the 21th day, the thymus weight increased by 0.5 g to 1.8 ± 0.5 g, and the length — by 0.4 cm to 5.1 ± 0.9 cm, that is, the increase was 27%; in the experimental group, a significant decrease was observed: in particular, the average weight of 3-day-old offspring was 0.8 ± 0.1 g; length — 3.6 ± 0.7 cm, that is, these indicators were 38% less than in the control rats. Similar differences are shown in the diagram below (Diagram 1).

Diagramma 1



At the same time, lymphoid cells are evenly distributed in many places. However, there are entire areas where thymocytes undergo significant degenerative changes. By

day 21, many reticuloepithelial cells degenerate, and small cyst-like spaces appear in their place. Cases of lymphocyte "adhesion" to reticular cells were also observed. (Figure 1)

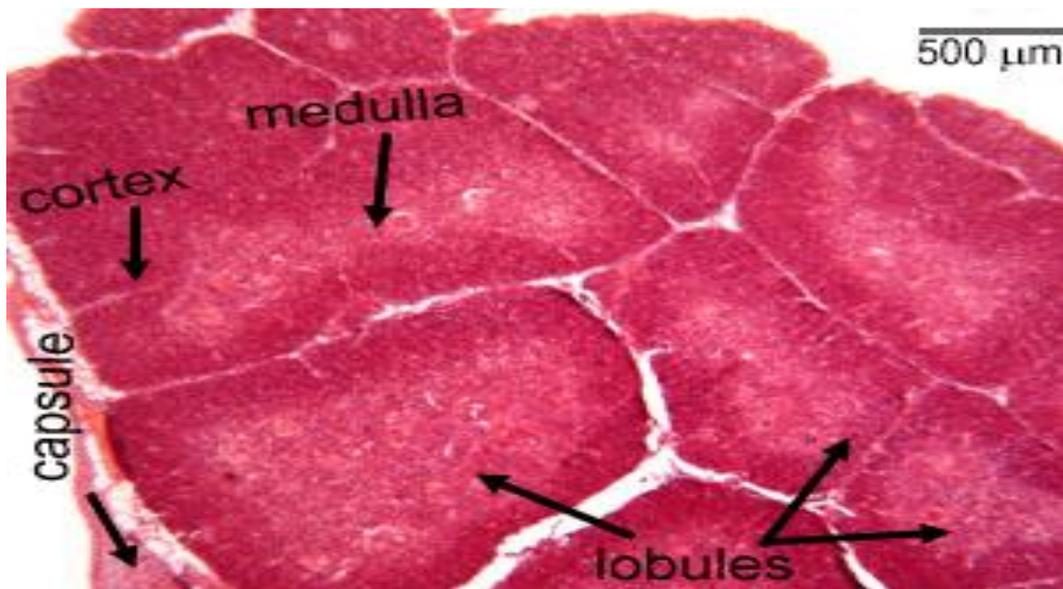


Figure 1: Thymic epithelial hyperplasia in a male B6C3F1 mouse from a chronic study. Hyperplastic epithelial cells form tubules (arrow) and cords (arrowhead) within the thymic medulla. (3 day)

Solid bodies are more common in experimental rats than in control animals. Smaller bodies have an empty lamina surrounded by concentrically arranged flattened reticuloepithelial cells with elongated nuclei. By day 21, the

lamina in larger bodies becomes filled with fragments of cellular protoplasm; their structure resembles lymphoid follicles. Small blood vessels are rarely found in the spleen tissue. (Figure 2)

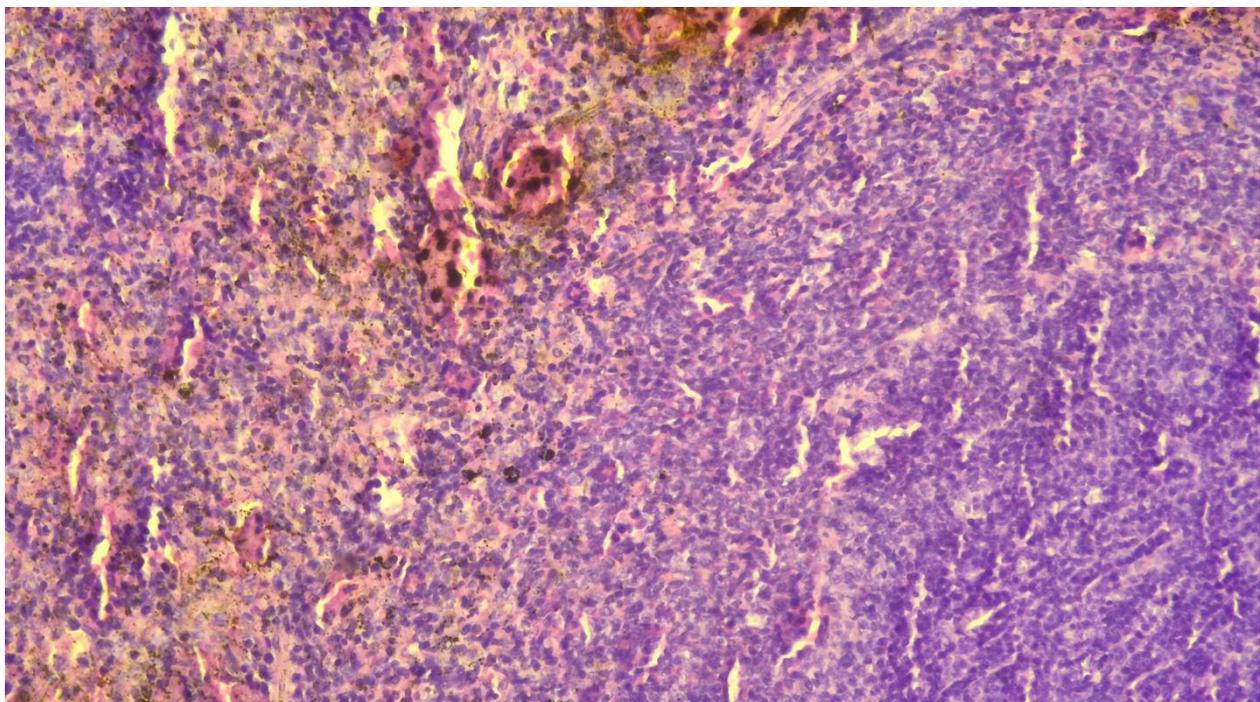


Figure 2: Spleen demonstrating amyloid deposition in blood vessels (hematoxylin and eosin stain, original magnification x200).

Table 1. Micromorphometric indicators

Term	Indicators	Control(M±m)	Experiment (M±m)	% difference	p
3 day	White pulp (%)	25 ± 2	18 ± 2	↓25%	<0,05
	Lymphoid follicle diameter (µm)	100 ± 8	70 ± 6	↓20%	<0,05
21 day	White pulp (%)	41 ± 3	33 ± 3	↓23%	<0,01
	Lymphoid follicle diameter (µm)	275 ± 15	240 ± 12	↓21%	<0,01

On the 3rd day of postnatal development, the offspring of rats exposed to prenatal chronic stress already demonstrated significant morphometric changes in the spleen compared to the control group. The relative area of the white pulp was reduced by 25% ($p < 0.05$), while the diameter of the lymphoid follicles decreased by 20% ($p < 0.05$). These findings indicate a delay in the early stages of lymphoid structure formation in the spleen and suggest an initial suppression of lymphocyte proliferation and differentiation processes caused by intrauterine stress exposure.

By the 21st day of postnatal development, these alterations persisted and became more indicative of long-term impairment in immune organ development. The area of the white pulp remained significantly decreased by 23% ($p < 0.01$), and the diameter of the lymphoid follicles was reduced by 21% ($p < 0.01$) compared to the control group. Unlike the early stage, when the changes reflected delayed formation of lymphoid structures, the findings at day 21 demonstrate a sustained disruption in the maturation of immunocompetent elements of the spleen. This persistent reduction in white pulp structures may indicate decreased adaptive immune capacity and a potential increase in susceptibility to infections in the offspring.

4. Conclusion

The study results showed that prenatal chronic stress has a

significant negative impact on the development of immune organs in rat offspring. The most profound changes were observed in the thymus, where a decrease in lobe size, damage to the karyolemma, a reduction in Hassall's bodies, and a significant decrease in T-lymphocyte counts were observed. Overall, the combination of these findings suggests that prenatal chronic stress causes systemic suppression of morphogenesis in both central (thymus) and peripheral (spleen) immune organs. The differences observed between the 3-day and 21-day postnatal periods indicate that prenatal stress not only disrupts the early formation of immune structures but also delays their structural maturation. As a result, offspring exposed to prenatal stress show signs of functional immune deficiency in the early stages of postnatal development.

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