

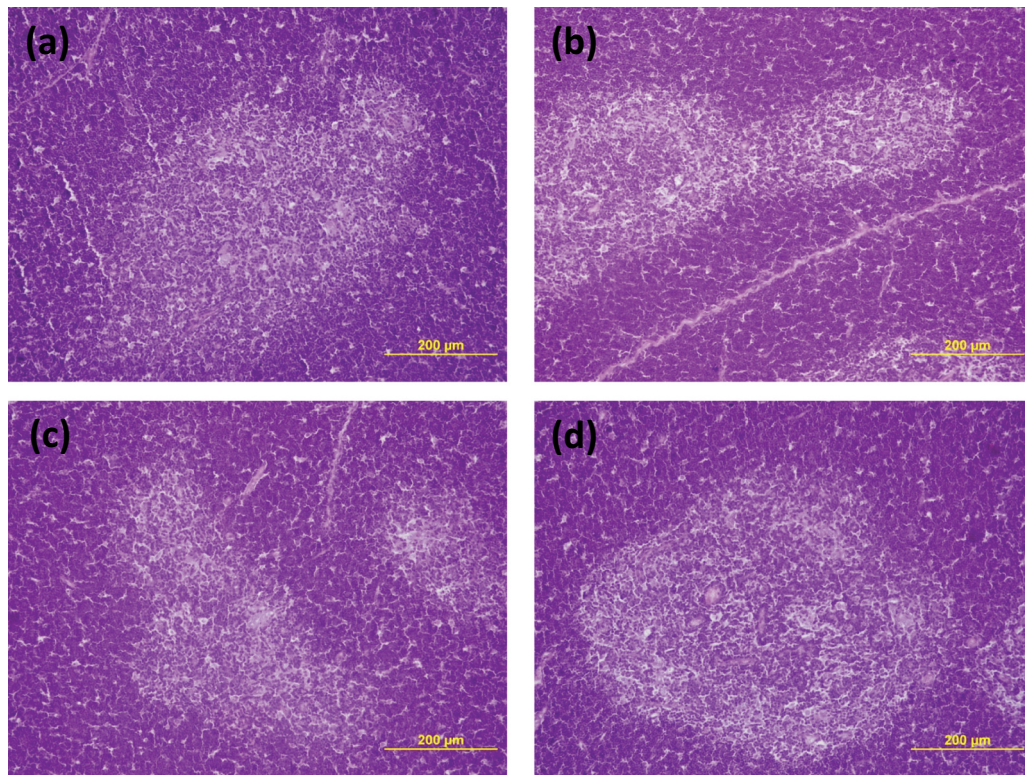


Figure 5. Histopathological changes in the thymus belonging to second generation rats exposed to EMF. In all groups of second generation; cortex, medulla and the residing cells were noticeable. The medulla of the organ consisted of numerous mature lymphocytes, reticuloepithelial cells and Hassall corpuscles. (a) Control group, (b) first group, (c) second group, (d) third group (H&E staining x400, Olympus, BX51, Tokyo, Japan).

when the second group of the fourth-generation was compared with the other experimental groups (Figure 7).

Discussion

The thymus is a fundamental lymphoid organ responsible for the maturation and selection of T cells. Thymus size and function are believed to decline with age (Gui et al. 2012). The thymus reaches its peak size during puberty, after which it gradually atrophies. If the thymus hypertrophy is not dependent on the patient's age, further investigation should be done (Khan and Anjum 2020). Thymic epithelial tumors manifest lobulated, contour, mediastinal fat invasion, hypertrophy, and vascular structure (Nishino et al. 2006).

This study investigated the effects of pre and postnatal 2450 MHz CWRFR on rats' thymus spanning four generations. Our study found morphological changes in the thymus tissue. Our most critical histopathological findings were visibly pronounced thymus hypertrophy in the exposed animals of the third and fourth generation and

the striking vascular structures in the second group of the fourth generation compared to the other experimental groups.

The results of this study were also consistent with earlier findings. Lee et al. investigated the effects of RF-EMF on AKR/J mice used for animal models of lymphoma. The mice were exposed to RF-EMF for 45 min/day, 5 days/week, over a total of 42 weeks. The thymus showed significant enlargement in most mice, and the thymoma was abnormal tissue (Lee et al. 2011). Misa-Agustino et al. exposed different levels of microwave radiation on rats. They reported histopathological changes induced 2450 MHz EMF in rat thymus. The researchers found extravascular erythrocytes in thymic tissue of animals exposed to EMF as in our study. Furthermore, they also reported an increased number of blood vessels in the hemorrhagic reticulo-epithelial cells and medulla (Misa-Agustino et al. 2015). It is evident from our results and the findings of Misa-Agustino et al. that EMF can cause changes in the morphology of thymic tissue.

Hanci et al. reported that light microscopy and transmission electron microscopy demonstrated pathological changes in splenic and thymic tissues of 900 MHz EMF

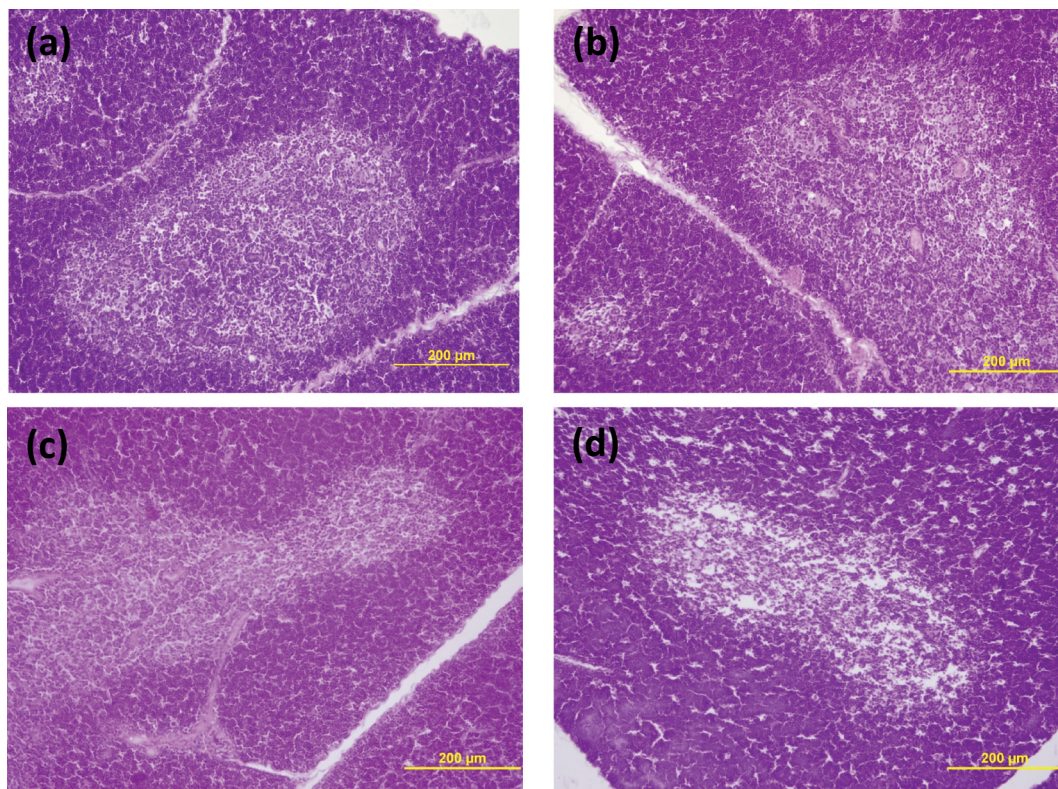


Figure 6. Histopathological changes in the thymus belonging to third generation rats exposed to EMF. In all groups of third generation; cortex, medulla and the residing cells were noticeable. The medulla of the organ consisted of numerous mature lymphocytes, reticuloepithelial cells and Hassall corpuscles. (a) Control group, (b) first group, (c) second group, (c) third group (H&E staining x400, Olympus, BX51, Tokyo, Japan).

exposed on rat offspring during the prenatal period. It was found that exposure during the prenatal period altered the antioxidant defense system and caused oxidative stress by increasing lipid oxidation. These researchers also reported mitochondrial degeneration, vacuolization, necrotic thymic lymphocytes, and epithelial cells in slices of EMF-exposed offspring (Hanci et al. 2015). Hashish et al. reported the effects of continuous whole-body exposure to both fields for 30 days on several blood and liver parameters in mice. They applied two magnetic fields: an extremely low-frequency electromagnetic field (ELF-EMF) and a static magnetic field (SMF). A significant reduction in the number of platelets, peripheral lymphocytes, monocytes, splenic total, T, and B lymphocyte levels was evaluated in the groups exposed to SMF and ELF-EMF (Hashisha et al. 2008). Kulaber et al. investigated changes in the thymic tissue of male rats exposed to continuous 900 MHz EMF for 1 h/day on postnatal days 22–59. They reported that a continuous 900 MHz EMF can change histopathology and increase the tissue MDA in male rat thymic tissue (Kulaber et al. 2017). Hekmat et al. applied 940 MHz RF-EMF on calf thymus DNA and stated that 940 MHz RF-EMF could alter the structure of DNA (Hekmat et al.

2013). Other researchers also reported the effects of ELF-EMF on the thymus and spleen tissues. Zhang et al. indicated that exposure to ELF-EMF in male mice decreased incidents of spleen and thymus tissues. Also, the exposure of ELF-EMF significantly decreased the NK-cell activity and proliferation of T and B lymphocytes (Zhang et al. 2016).

Contrary to our findings, some studies did not observe any change (Ohtani et al. 2015; Sommer et al. 2007, 2004). Sommer et al. (2004) exposed female AKR/J mice to 900 MHz EMF for 24 h/day, 7 days/week. Lymphoma incidence did not differ between the sham-exposed and exposed mice. Sommer et al. showed whether chronic exposure to the UMTS (Universal Mobile Telecommunication System) EMFs influenced the development of lymphoma in a lymphoma AKR/J mouse model. Exposure for 24 h/day, 7 days/week with an average whole-body SAR of 0.4 W/kg did not affect the lymphoma incidence and the malignancy of the lymphomas, which were genetically predisposed to this disease. Ohtani et al. reported the effects of continuous exposure to 2.14-GHz RF-EMF on T cell responses and its effects on the developmental immune systems. Their data indicated that long-term RF-EMF exposure had no

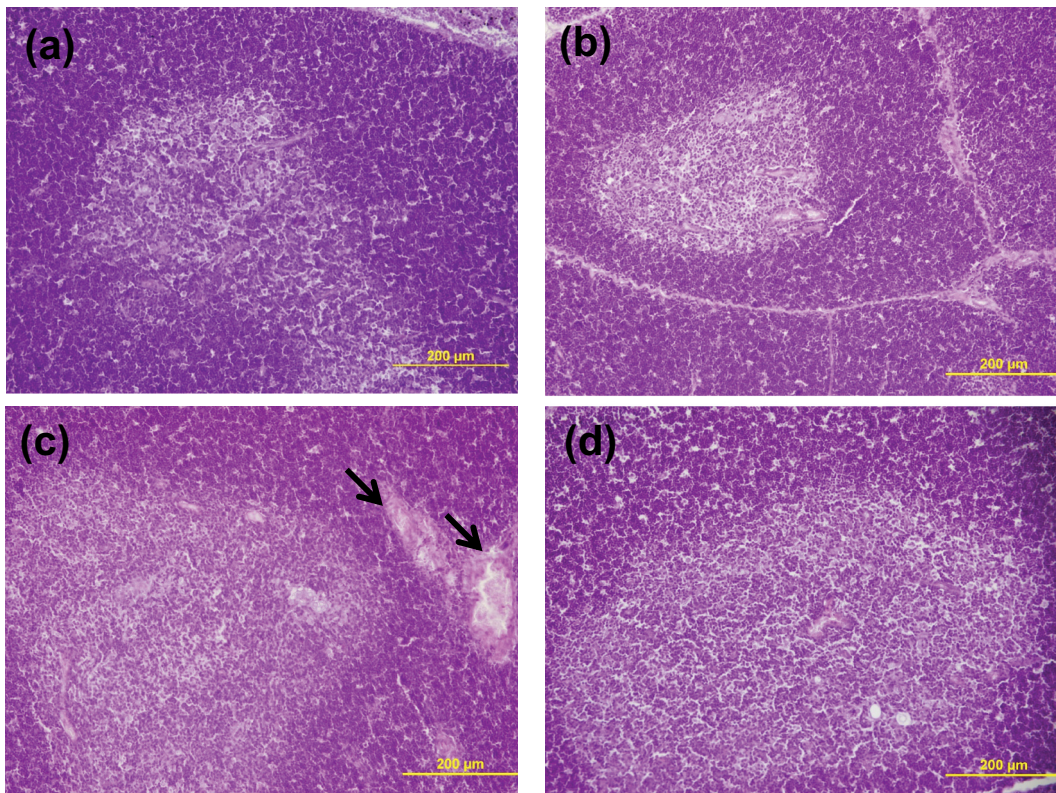


Figure 7. Histopathological changes in the thymus belonging to fourth generation rats exposed to EMF. The thymus cortex was formed multiple lymphocytes and reticuloepithelial cells. In first and third groups of fourth generation; cortex, medulla and the residing cells were noticeable. (a) Control group, (b) first group (c) in the second group, vascular structures were remarkable compared with other experimental groups, (d) third group (arrow; blood vessel, H&E staining x400, Olympus, BX51, Tokyo, Japan).

adverse impactation T cell activation, Th1/Th2 balance, and immune-like T cell populations in developing rats. However, important transcriptional effects were shown (Ohtani et al. 2015).

Conclusion

Our findings reveal that the effect of pre- and postnatal 2450 MHz CWRFR decreases the average body masses of rats. Pre- and postnatal 2450 MHz CW RFR exposure may potentially affect the thymus of future generations. Throughout three generations, increased vascularization and hypertrophy of the thymus can cause thymic epithelial tumors.

Disclosure statement

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F.C.T. and S.D. are responsible for the study design and writing of the article. B.Y. and A.H.Y. performed the microscopic analyses of slices. B.T. was responsible for the statistical analysis of the data. K.Y. was measured the SAR value.

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