

In vivo Assessment of initial Thymus Size and Age-related Thymic Involution in a C57BL/6J Mouse Strain

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Introduction

The thymus is a primary site for the development and output of the majority of naïve T-lymphocytes (T cells), a heterogeneous group of cells essential in protecting the body against invasions by foreign organisms¹. T cells are critical cells of the adaptive immune system. Despite this critical immune function, the thymus undergoes physiological changes associated with aging that is termed thymic involution². The thymus gland is largest and most active (T cell output) during the neonatal and pre-adolescent periods and begins to atrophy after puberty. Thymic involution is closely associated with immunosenescence, a degeneration of the immune system primarily due to the alterations in T cell composition.

The C57BL/6J mouse strain can serve as a convenient model for investigations of immune-based therapeutics that can enhance thymic function. As a result, measuring mouse thymus weight after pharmacological intervention can be used as an early marker for potency. Thymus weight is usually measured post mortem precluding the possibility of repeated measurements. Initial thymic size and rate of involution has been shown to be different across gender and strains of mice³.

In this study, we aimed to investigate in vivo by means of MRI the thymus time dependant natural growth and involution pattern in a group of male and female C57BL/6J mice.

Methods

The study was performed in 19 mice in bred in house, 7 male mice (pups) and 12 female mice divided in two equal groups as they came from a different litter. The local Animal Research Ethics Board Committee (Göteborg) approved the study. Anesthetized animals were imaged once a week when mice were in the range 4 to 10 weeks old. Imaging was performed on a 9.4 T Biospec Bruker MR scanner. Initially, scout images of the heart anatomy were acquired for accurate planning of the subsequent long axis images covering the entire thymus using the IntraGate gradient echo scan sequence with the following parameters: FOV: (25x25) mm², 20 contiguous slices of 0.6 mm thickness, matrix dimension: 256x256, spatial resolution: (98x98) µm², TR/TE/alpha/NR: 152 ms/2.5 ms/22 deg/20, total acquisition time: 6 min. After the last imaging session mice were euthanized and the thymus was excised for measure of the wet weight (ww) mass. Multi slice images were exported and thymus volume was estimated by manual segmentation using the Analyze software v.11 (AnalyzeDirect Inc., Overland Park, KS, US).

Results

A fast imaging protocol allowing high throughput and minimal stress was designed. High resolution gated images (Fig. 1A) showed a high contrast between thymus and peripheral tissues allowing a simple segmentation procedure of the thymus. Fig. 1B displays a representative reconstructed 3D surface of a 7 weeks old thymus. Reproducibility assessed by repeated measurement showed good agreement (mean = -1.2; 95% limits of agreement = -5.0 and 2.6). Fig. 2 represent the time dependant thymus volume growth and involution in male (●) and female (■) mice (mean ± STDV). A distinct pattern was observed between male and female mice. Bland-Altman plot (Fig. 3) for the agreement between ww and estimated MRI weight (density: 1.1 g/ml) showed a good agreement. All points to fall within 2 standard deviation of mean (outer dashed lines).

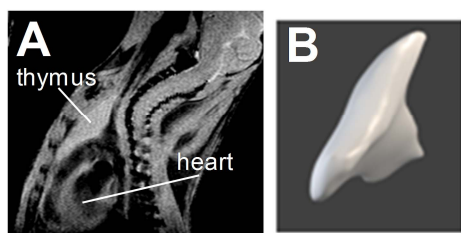


Fig. 1

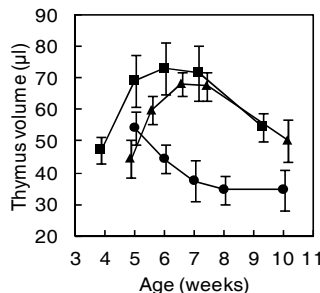


Fig. 2

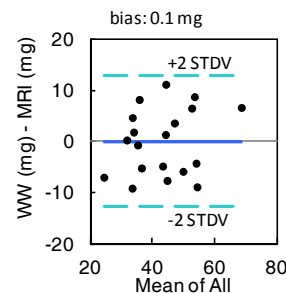


Fig. 3

Discussion/Conclusion

In this study we demonstrate for the first time in vivo the natural growth and involution pattern of the thymus in a group of C57BL/6J male and female mice. Male thymus involution started before mice reached 5 weeks of age and persisted up to 8 weeks. Females from a different litter size showed a different initial growth pattern. Interestingly, both groups reached the highest thymus volume when mice were about 6-7 weeks old. Involution occurred at the same rate. The results of this study provide us with a basic data set for evaluating the effect of compounds on thymus weight and to design studies in light of the observed involution pattern.

References

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