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ORIGINAL ARTICLE

Normative values of thymus in healthy children; stiffness by shear wave elastography

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PURPOSE
Thymus grows after birth, reaches maximal size after the first few years and involutes by puberty. Because of the postnatal developmental and involutional duration, we aimed to investigate normal stiffness values of mediastinal thymus by shear wave elastography (SWE) in different age groups of children and discuss imaging findings of thymus.

METHODS
We prospectively examined 146 children (90 girls, 56 boys) who underwent a thyroid or neck ultrasound examination. All subjects underwent ultrasound and SWE evaluation of mediastinal thymus by parasternal and suprasternal approach. We grouped the subjects based on age as 0 to 2 months, >2 to 6 months, >6 months to 2 years, >2 to 5 years, >5 to 8 years, and greater than 8 years old. We investigated differences of mean shear wave elasticity (kPa) and shear wave velocity (m/s) values among age groups and the association of SWE values with age, body mass index (BMI), height, and weight of the patients.

RESULTS
Median and range of age, height, weight, and BMI were 24 months (2–84 months), 85 cm (55–120 cm), 12 kg (4.55–22 kg), 15.37 kg/m^2 (13.92–17.51 kg/m2), 11 cc (2.64–23.15 cc), respectively. Mean shear wave elasticity of thymus of all participants was 6.76±1.04 kPa. Differences of mean elasticity values among the age and gender groups were not statistically significant. Thymus elasticity and velocity values showed highly significant negative correlations with age (r= –0.3), height (r= –0.26), weight (r=–0.3) ($P < 0.001$).

CONCLUSION
Quantitative evaluation of the thymus by SWE provides normative stiffness values based on age and gender groups. The thymus elasticity decreases with increased age, height, and weight.

Thymus is a soft, roughly triangular shaped lymphoreticular organ presenting an age dependent appearance and content. Embryologically, the thymus develops bilaterally at approximately the 5th week of gestation from the third and fourth pharyngeal pouches and migrates caudally and medially along with thymopharyngeal duct (1). Ventral wing of the third pharyngeal pouch forms the thymic tissue. Along the descent pathway, thymic tissue can be observed anywhere from the angle of the mandible to the manubrium sterni. An abnormally positioned thymus could be determined either as aberrant if thymus is located along the normal descent pathway, or as ectopic if it is located at different locations such as the pharynx, trachea, posterior neck, or esophagus (2).

Ultrasonographic (US) features of mediastinal thymus have been published in a recent study (3). Thymus is a homogeneous soft tissue hypoechogenic to thyroid gland and includes punctuate echogenicities. The echogenicity is equal to or lower than strap muscles especially in newborns and increases with age. On unenhanced computed tomography (CT), thymus density is lower than thyroid gland due to the internal iodine content of thyroid gland and similar with mediastinal lymph nodes. Therefore, it would be easy to differentiate plunging goiter or ectopic thyroid from thymus on CT due to radiodensity differences. However, it will be difficult to differentiate rebound thymic hyperplasia from mediastinal lymphoreticular malignancy recurrence

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only by density-based CT evaluation. Thymic tissue is hyperintense than salivary glands and isointense to lymph nodes on T2-weighted magnetic resonance imaging (MRI). Along the descent pathway, ectopic or aberrant thymic tissue should be kept in mind among the differential diagnosis of anterior cervical and superior mediastinal lymph nodes depicted on T2-weighted MRI. Despite well-described imaging features, many cases regarding aberrant thymic tissue either located as cervical or intrathyroidal have been published as a diagnostic challenge (4, 5). In such cases an ultrasound-based evaluation that is noninvasive and quantitative, such as SWE, is required as an additional imaging modality.

US is a noninvasive, radiation-free, reproducible, first-choice imaging modality in pediatric patients with anterior mediastinal enlargement, especially in the first few years of life. US is indispensable for newborns with mediastinal widening, because of partial ossification of the sternal segments and costal cartilages and also homogeneous echo texture of an enlarged thymus gland providing an acoustic window. As a descriptive qualitative evaluation, contour lobulation and heterogeneity in the thymus would lead to consideration of neoplasia or parenchymal diseases. In terms of quantitative evaluation, the strain ratio of intrathyroidal ectopic thymic tissue compared to thyroid gland has been investigated by strain elastography revealing semiquantitative data (6). However, there is lack of quantitative criteria for diagnosing thymic tissue by US-based applications such as shear wave elastography (SWE).

Thymus presents a postnatal growth period followed by involution. In this study, we aimed to investigate SWE values as diagnostic quantitative data according to age and gender groups during developmental and involutional processes. Being aware of normative data would provide diagnostic contribution in distinguishing thymic tissue by a radiation-free diagnostic modality.

Methods

Subjects and study design

This prospective study included 146 children, 56 boys and 90 girls, between March and June 2019. The local ethics committee of the institutional review board approved this study (File number: 2019/99). Before parasternal and suprasternal US and SWE examinations, informed consent was obtained from the parents of participants. Due to lack of sufficient number of cases to constitute nomograms for each age, the subjects were divided into several age groups in order to reveal changes in thymus stiffness. Subjects were analyzed in classes A, B and C, consisting of six, four, and two age groups, respectively. The classifications include neonate, infant, preschool age children, school age group, and adolescent age group (Table 1). Demographic data were noted as gender, height, weight, and body mass index (BMI). We included patients who are proposed to be healthy during SWE examination based on physical examinations. Exclusion criteria were rejecting participation in the study, hematological malignancy or chemoradiotherapy history, prematurity for newborns, fever during assessment, weight loss, previous mediastinal surgery, primary immunodeficiency, immunosuppressant medication use, or focal intraglandular lesion.

Ultrasonographic evaluation

All gray-scale and SWE evaluations were performed by using an Aplio^{TM} 500Platinum US device (Canon Medical Systems Co. Ltd.) and a high-frequency linear probe that was set to small parts preset (frequency range, 5–14 MHz). SWE evaluations were performed by a pediatric radiologist with more than seven years of pediatric radiology and three years of SWE experience and a radiologist with a year of pediatric radiology and SWE experience, in consensus. The patients were in the supine position. Once the whole thymus gland was evaluated carefully in terms of absence of a local heterogeneity or contour lobulation (Fig. 1), the best acquisitions demonstrating parallel propagation lines were obtained by consensus of the two radiologists. Propagation map along with SWE window were evaluated when the lines were parallel and smooth on the propagation mode (arrival time contour). For SWE measurements, the entire linear probe surface was covered with ultrasonic gel pad of 5 mm in thickness. Any external pressure was not applied to the probe, and the operator was stationary. The long axis of the probe was positioned such that it was perpendicular to the tissue surface. When the tissue with parallel propagation lines was obtained, five circular regions of interest (ROI) of 3 mm in diameter were selected on the acquisition by consensus of the two radiologists (Fig. 2). The medians of five shear wave elasticity and shear wave velocity values were selected to reveal closest value for each thymus gland. The SWE parameters were measured in kilopascal (kPa) for shear wave elasticity (SWe) and meters/second (m/s) for shear wave velocity (SWv). The elastographic scale was set to 0 to 40 kPa and 0 to 8 m/s with a real-time propagation map. Overall examination regarding the scan of the gland with gray-scale US, obtaining optimal propagation lines, selection of the ROIs by consensus took 5–6 minutes in general, but the examination took 2–3 minutes longer in newborns and infants.

Statistical analysis

All data were processed in Microsoft Office Excel and transferred to SPSS (version 21.0, IBM Corp.) for statistical analysis. The distribution of the data was assessed with

Main points

- Several thymic pathologies such as intrathyroidal ectopic thymus, undescended cervical thymus, or thymic hyperplasia would be confounding entities.
- Being aware of the normal shear wave elasticity and velocity values of thymus according to age groups would limit the differential diagnoses and decrease unnecessary interventions.
- SWE evaluation of the thymus is a noninvasive, quantitative diagnostic tool for distinguishing thymic tissue.

Table 1. Characteristics of the patients and elasticity and velocity values of the thymus

n=145	Min–max	Mean±SD	Median	Interquartile range
Age (months)	1–180	47.5±50.4	24	2–84
Height (cm)	40–168	93.3±36.6	85	55–120
Weight (kg)	3.2–67	16.4±14.2	12	4.55–22
BMI	9–26	15.5±4.0	15.3	13.92–17.51
SWe (kPa)	4.50–10.40	6.76±1.04	6.90	6–7.55
SWv (m/s)	0.61–1.87	1.49±0.13	1.52	1.42–1.59

SD, standard deviation; BMI, body mass index; SWe, shear wave elasticity; SWv, shear wave velocity.

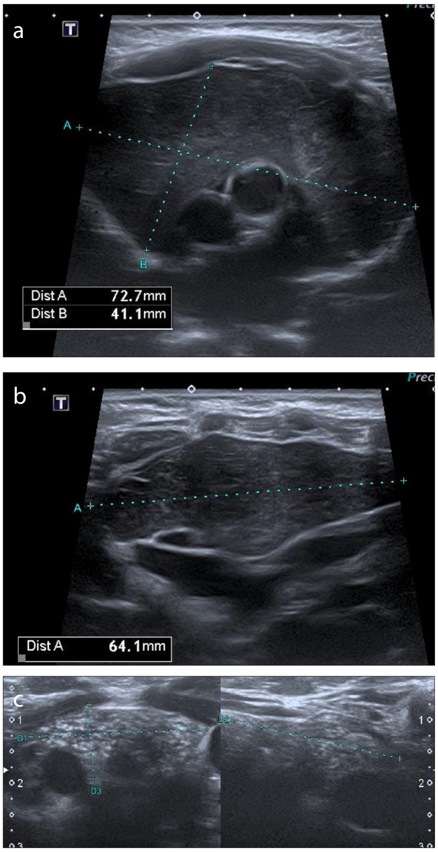

Figure 1. a–c. Gray-scale images **(a, b)** of the diffusely hypoechogenic mediastinal thymus in a male newborn. Gray-scale image **(c)** reveals thymus of a 13-year-old female patient.

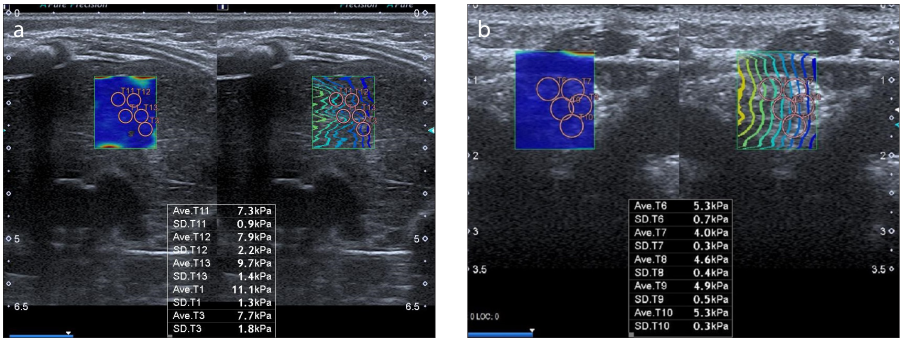

Figure 2. a, b. Shear wave elasticity (SWe) measurements and propagation map on shear wave elastography (SWE) are seen. On SWE image **(a)** of a newborn, mean SWe value was 8.74 kPa for the thymus. B. On SWE image **(b)** of an adolescent girl, mean SWe value was 4.82 kPa for the thymus.

the Kolmogorov-Smirnov test. Differences of SWE parameters among the age and gender groups were compared using the Student t test and ANOVA. The groups demonstrating differences among mean values have been tested with Tukey's HSD (honestly significant difference) test.

Correlation analysis of the SWE parameters with BMI, height, weight, and age was performed using the Spearman ρ (r) correlation coefficient. Quantitative variables were shown as mean with standard deviation, or median with range. Variables were studied at the 95% confidence interval with $P < 0.05$ accepted as statistically significant.

Results

Descriptive statistics of the demographic data including age, height, weight, and BMI of all the participants are given in Table 1. Mean values of thymus SWe and SWv for all participants were 6.76±1.04 kPa and 1.49±0.13m/s, respectively.

Table 2 represents the comparison of the mean SWe and SWv values among the age groups. Among the six consecutive age groups under classification A (0–2 months, n=25; 2 to 6 months, n=30; 6 months to 2 years, n=19; 2 to 5 years, n=24; 5 to 8 years, n=25; >8 years, n= 22), differences of mean SWe and SWv values were not statistically

Table 2. Comparison of gender and age groups according to different group classifications (A, B, C)

				Age		SWe		SWv	
			Age range	Mean±SD	n	Mean±SD	*P*	Mean±SD	*P*
Age groups	A	1	0–2 m	1±0.5	25	7.14±1.21	0.005^{a}	1.53±0.13	0.005^{a}
		2	2–6 m	3.3±1.3	30	7±0.96		1.52±0.12	
		3	>6 m to 2 y	17.4±5.3	19	6.9±1.04		1.47±0.23	
		4	>2 to 5 y	43.5±8.8	24	6.59±0.95		1.49±0.11	
		5	>5–8 y	81±7.9	25	6.43±0.89		1.46±0.09	
		6	>8 y	136±25.7	22	6.31±0.95		1.45±0.11	
	B	1	0–3 m	1.5±0.77	41	7.1±1.14	0.001^{a}	1.53±0.13	0.001^{a}
		2	>3 m to 2 y	17.1±11.5	42	6.96±0.94		1.49±0.17	
		3	>2 to 10 y	66.1±16.3	37	6.35±0.83		1.46±0.01	
		4	>10 y	132.2±27.3	25	6.4±1		1.46±0.11	
	C	1	0–6 y	9.6±11.4	83	7.05±1.04	0.001^{b}	1.51±0.15	0.003^{b}
		2	6–18 y	94.3±39.3	62	6.37±0.90		1.46±0.11	
Gender groups			Male	46.2±50.7	86	6.65±0.97	0.11^{b}	1.5±0.17	0.70^{b}
			Female	49.5±50.2	59	6.93±1.12		1.49±0.11	

Tukey's HSD test results. Group A, SWe; group 1 vs. 5 ($P = 0.022$), group 1 vs. 6 ($P = 0.013$) and SWv; group 1 vs. 5 ($P = 0.037$), group 1 vs. 6 ($P = 0.033$). Group B, SWe; group 1 vs. 3 ($P = 0.003$), group 1 vs. 4 ($P = 0.018$), group 2 vs. 3 ($P = 0.034$) and SWv; group 1 vs. 3 ($P = 0.007$), group 1 vs. 4 ($P = 0.02$), group 2 vs. 3 ($P = 0.04$).
SWe, shear wave elasticity; SWv, shear wave velocity; SD, standard deviation, m, months; y, years.
^{a}*P* values obtained by ANOVA test.
^{b}*P* values obtained by t test.

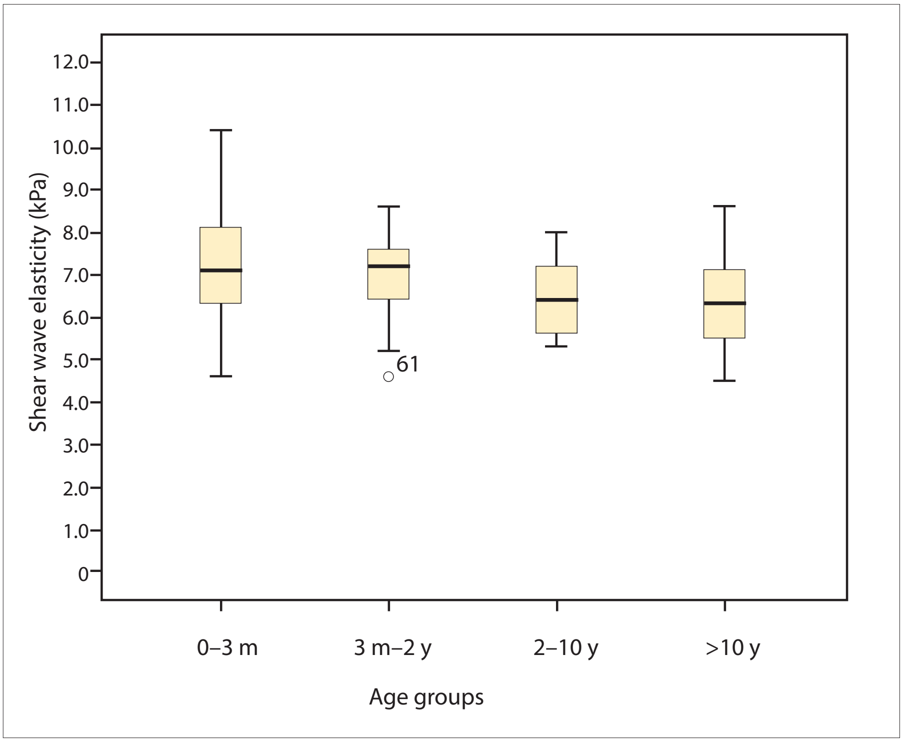

Figure 3. Box plot presents SWe values of thymus in all four age groups (class B).

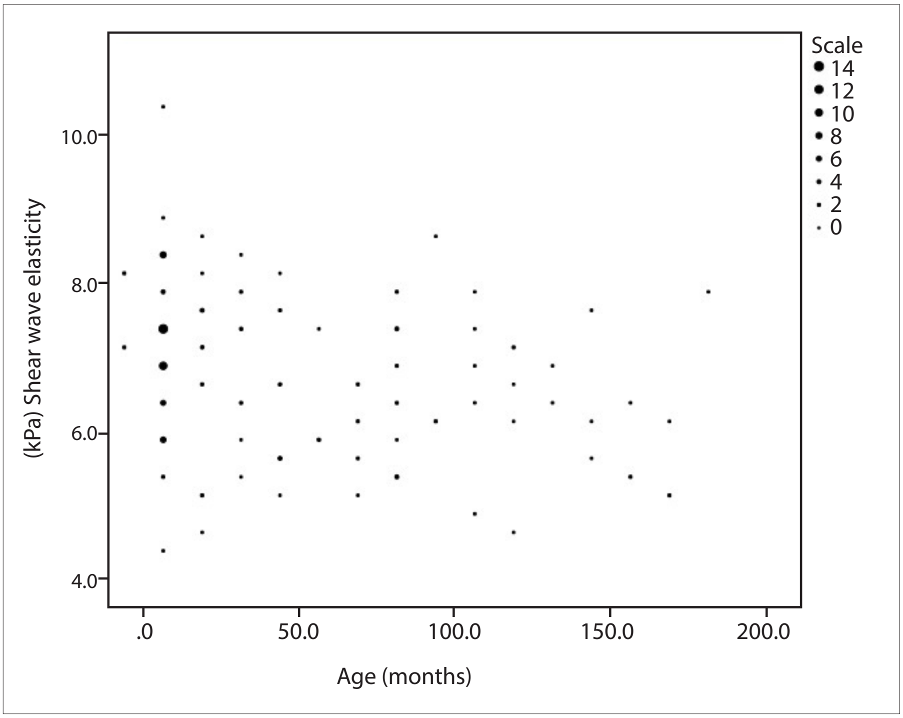

Figure 4. Scatter plot diagram presents the negative correlation of age with SWe of the thymus.

significant. However, we found significant difference within the age groups in classification A (*P* = 0.005). Mean values of SWe of group 1 vs. 5 (*P* = 0.022) and group 1 vs. 6 (*P* = 0.013) were significantly different. Similarly, mean values of SWv of group 1 vs. 5 (*P* = 0.037) and group 1 vs. 6 (*P* = 0.033) were significantly different.

Among the four consecutive age groups under classification B (0–3 months, n=41; 3 months to 2 years, n=42; 2 to 10 years, n=37; > 10 years, n=25), no significant difference was found in terms of mean values of SWe and SWv. However, we found significant difference within the age groups in classification B (*P* = 0.001). Mean values of SWe of group 1 vs. 3 (*P* = 0.003), group 1 vs. 4 (*P* = 0.018), group 2 vs. 3 (*P* = 0.034) were significantly different. Similarly mean values of SWv of group 1 vs. 3 (*P* = 0.007), group 1 vs. 4 (*P* = 0.02), group 2 vs. 3 (*P* = 0.04) were significantly different.

Under classification C, with two age groups of children 0–6 years (n=83) and 6–18 years (n=62), both SWe (*P* < 0.001), and SWv values (*P* < 0.003) decreased after six years and differences of SWe (group 1 vs. 2; *P* = 0.001) and SWv (group 1 vs. 2; *P* = 0.003) values were statistically significant. The mean SWe (*P* = 0.11) and SWv (*P* = 0.7) values did not differ significantly among gender groups.

The SWe values presented statistically significant mild negative correlation with age (*P* = 0.001, r=-0.31) (Fig. 4), height (*P* = 0.003, r=-0.26) and weight (*P* = 0.001, r=-0.30) (Table 3). The SWv values were also negatively correlated with age (*P* = 0.001, r=-0.26) (Fig. 4), height (*P* = 0.027, r=-0.19) and weight (*P* = 0.008, r=0.22).

Discussion

The thymus components involving true thymic epithelial space and the perivascular space are fully developed before birth, and thymus upregulates functions during the early neonatal period (7, 8). Mean thymus volume has been found to be significantly greater in term infants compared to preterm neonates among children younger than two years old (9). Thymus is a common initial consideration in children with a wide superior mediastinum at early childhood. On chest roentgenogram, the wavy contour of an enlarged mediastinum in an asymptomatic newborn reflects the softness of the thymic tissue (10). In addition, such as a typical geographic shape in case of ectopic intrathyroidal location, and pliable properties such as elongation through the carotid space and positioning between the major vessels in either ectopic or eutopic location in absence of mass effect help to differentiate a normal thymus from its mimickers. Beyond these qualitative and descriptive radiological features, there has been only one study investigating relative stiffness of intrathyroidal ectopic thymus with strain elastography compared with thyroid gland, revealing a mean strain ratio value as 1.02 (range, 0.95–1.09) (6). However, no study

Table 3. Association of the demographic data with SWE parameters

	Shear wave elasticity (kPa)		Shear wave velocity (m/s)	
	P	r	*P*	r
Age	0.001	-0.31	0.001	-0.26
Height	0.003	-0.26	0.027	-0.19
Weight	0.001	-0.30	0.008	-0.22
Body mass index	0.18	-0.11	0.31	-0.08

Data represent Spearman correlation analysis results.
SWE, shear wave elastography; r, correlation coefficient.

investigating the normal elasticity range for thymic stiffness via US-based applications has been conducted.

SWe values of several superficial neck organs in children have been published in recent studies (11–15). In these studies, mean±standard deviation / median (interquartile range) for SWe values of thyroid gland, submandibular gland, parotid gland, cervical lymph nodes and palatine tonsils in healthy pediatric population have been reported as 6.38±1.97 kPa (11), 11.8±2.2 kPa (12), 8.37±2.09 kPa (13), 11.19 kPa (5.9–16.6 kPa) (14), 9.38±1.27 kPa (15), respectively. Mean SWe of the thyroid gland has been found as 14.6±3.3 kPa in a recent study (12). The preliminary results of the present study suggest the softness of the thymic tissue by revealing the mean stiffness of the thymus as 6.76±1.04 kPa, which is lower than previously determined stiffness values for parotid gland, submandibular gland, thyroid gland, lymph nodes and palatin tonsils in the pediatric population. We reported SWE results as SWe (kPa) and also as SWv (m/s). We propose that the preliminary results would help to identify thymic tissue, especially in case of aberrant location. An ectopic thymic tissue around the angle of the mandible would be considered in differential diagnosis of palatin tonsils or lymph nodes due to the their hypoechogenic appearance because of having a lymphoreticular origin. Lower SWe values would suggest thymic tissue rather than a lymph node or tonsil. Pathological conditions mimicking an ectopic thymus in children include lymph node metastasis of papillary thyroid carcinoma and neuroblastoma. As the elasticity of the reactive lymph nodes (14) is already higher than the thymus, the lymph node elasticity is expected to increase in the presence of metastasis. In addition, our results would be used in the daily practice for eliminating suspected diffusely infiltrative lymphoreticular disorders of the thymus such as leukemia and lymphoma.

The primordial thymopharyngeal tract remnants can appear anywhere from the piriform sinus to the manubrium along the thyrohyoid membrane and carotid artery (16). In case of a mass detected along the descent pathway of the thymus, presenting with hyperintensity on T2-weighted MRI and homogeneous contrast enhancement, further evaluation with high-resolution US as well as SWE can support the diagnosis of ectopic thymus if lower stiffness is found compared with superficial neck organs; thus, a diagnosis can be reached using a noninvasive, radiation-free diagnostic method without the need for sedation or contrast administration.

Dyshormonogenetic glands in children with congenital hypothyroidism tend to be enlarged, hypoechogenic relative to normal thyroid gland and cricoid cartilage on US, and also associated with increased vascularity relative to healthy thyroid glands on Doppler US examination (17). Patients with congenital hypothyroidism may be misdiagnosed as thyroid agenesia with ectopic cervical thymic tissue, due to the significantly decreased echogenicity. The tiny echogenic reticulations in dyshormonogenetic gland and punctate echogenic foci within the thymic tissue would help to differentiate the origin of the tissue (17). Because there has not been a significant difference in elasticity among dyshormonogenetic and normal thyroid glands in children (median values of SWe, 9.75 kPa vs. 9.5 kPa) (17), and we have depicted the mean SWe of thymus gland as 6.76±1.04 kPa; an ectopic thymus could be distinguished by SWE even when it is located in a dyshormonogenetic thyroid gland. During routine cervical and thyroid examinations, SWE together with gray-scale US can be very helpful for diagnosing ectopic thymic tissue, which has a considerable reported incidence of 0.99% for intrathyroidal ectopy (3), by obtaining SWe values equal to or smaller than 7 kPa. Thus, SWE would help to achieve final diagnosis noninvasively.

Rebound thymic hyperplasia and also ectopic cervical thymus extending into the highest mediastinal lymphatic stations and supraclavicular region would be confusing in patients with lymphoreticular malignancy (18). In addition, positive interim FDG-PET/CT examinations may have poor predictive values for prediction of recurrence of primary mediastinal B-cell lymphoma (19). The Deauville criteria (20) interpret uptake values of the lesions in comparison with mediastinum and liver. In cases with Deauville score 3 (mediastinum< FDG uptake < liver) or more, US and elastography evaluation could be valuable in decision making of residual or recurrent lymphoproliferative disorders by excluding eutopic and ectopic normal thymic tissue along with thymus hyperplasia.

There have been several US based elastography methods. Qualitative and semiquantitative results could be obtained with quasi-static strain imaging and acoustic radiation force impulse (ARFI) imaging by measuring the shape deformation. Transient elastography (TE) and ARFI quantification, as well as point shear wave elastography (p-SWE) could quantitatively calculate tissue stiffness in a target region. However, two-dimensional SWE evaluates viscoelastic tissue properties reflected by shear or Young's modulus quantitatively as a more popular and also more operator-independent method (21, 22). We used two-dimensional SWE to provide easily measurable, quantitative and objective data in the pediatric age group.

There are several limitations to our study. First, we had a limited number of cases per age groups. Nevertheless, our study population for the SWE of the thymus is the largest one to date in a single study. Second, the measurements were made by two operators in consensus but we did not obtain separate SWE values for each participant. Repeated acquisitions and many regions of interest would be performed to reduce inter and intraobserver variability. Third, it is too difficult to stabilize patients especially infants and younger children resulting in suboptimal SWE images and propagation lines. We excluded the data that could not be assessed and asked the mothers to breastfeed or put the infants to sleep in order to stabilize them during acquisitions. We kept the gel warm to avoid waking the infants. Moreover, especially infants and patients with small thymic tissue around vessels are prone to cardiac or vascular pulsation artifacts, which was a limiting factor on calculations. Suprasternal approach and selection of the regions of the interest from the periphery of the gland were used to avoid pulsation artifacts. In addition breath holding in patients who can comply would facilitate achieving optimal acquisitions rapidly.

In conclusion, mean thymus stiffness represents a narrow range in terms of elasticity. Thymus elasticity decreases with age, independently from gray-scale and Doppler imaging characteristics. SWE evaluation of the thymus is a noninvasive, quantitative diagnostic tool for distinguishing the thymic tissue.

Conflict of interest disclosure

The authors declared no conflicts of interest.

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