

Neonatal thymectomy in children—accelerating the immunologic clock?

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The thymus is critical for central tolerance and diverse T-lymphocyte repertoire development, to provide lifelong defense against pathogens while maintaining self-tolerance. Peak thymic output occurs *in utero*, during infancy, and in early childhood, diminishing throughout life. Infants with congenital heart disease requiring sternotomy often undergo thymectomy to clear the surgical field. The long-term effects of early thymectomy are just being appreciated. Many patients remain asymptomatic despite immunologic findings mirroring those of immunosenescence. Few develop increased infection or lymphoreticular malignancy risk. When considering the effects of infant thymectomy, patients with partial DiGeorge syndrome or hypomorphic recombination-activating gene (*RAG*) mutations may be instructive. These patients are lymphocytopenic, with increased early-onset infection and autoimmunity risk that is not seen in most patients who underwent thymectomy during infancy. The thymic structure of patients with partial DiGeorge syndrome or hypomorphic *RAG* is abnormal, with disrupted architecture inclining to perturbation of central tolerance. Similar findings may be seen in patients with myasthenia gravis, although disrupted peripheral tolerance may play a greater role in autoimmunity development. In conclusion, thymectomy during infancy may increase future risk of infection or autoimmunity, with premature immunosenescence mediated through disruption of central and peripheral tolerance mechanisms initiated by early cessation or diminution of thymic output. Ideally, some thymic tissue should be preserved at the time of surgery. (*J Allergy Clin Immunol* 2020;■■■:■■■-■■■.)

Key words: Thymus, neonatal thymectomy, immunosenescence, T-lymphocyte, immunodeficiency

The thymus is the primary lymphoid organ supporting development of T-lymphocytes capable of reacting with harmful foreign antigens but recognizing and tolerating harmless self-

Abbreviations used

AChR: Acetylcholine receptor
CHD: Congenital heart disease
iTreg: Peripheral (induced) regulatory T-lymphocyte
MG: Myasthenia gravis
nTreg: Thymic (natural) regulatory T-lymphocyte
PTLD: Posttransplant lymphoproliferative disease
TCR: T-cell receptor
TREC: T-cell receptor excision circle
Treg: Regulatory T-lymphocyte

antigens. Complete or partial thymectomy performed during congenital cardiac surgery procedures in infants and young children improves surgical access and allows visualization and cannulation of great vessels.¹ However, partial or complete removal of the thymus in early life may have significant health impact in adulthood, although the data describing this are mixed.

Several studies have described T-lymphophenotype changes and early immunosenescence occurring after early thymectomy for cardiac surgery, although the findings differ between reports, most of which are limited to small patient cohorts, usually with no appropriate controls. The short-term clinical impact of early thymectomy does not result in severe infectious complications, but the long-term consequences are poorly defined. In other conditions for which postthymectomy long-term follow-up data are available (eg, myasthenia gravis [MG]), significant late-onset autoimmune and lymphoproliferative complications are reported.^{2,3} Similar complications following cardiac surgery may occur in patients who were thymectomized early in life.⁴

We describe current understanding of the clinical and immunologic consequences of thymectomy during cardiac surgery performed during infancy or early childhood and explore the link with immunosenescence.

NORMAL THYMIC STRUCTURE AND FUNCTION

The thymus is critical for T-lymphocyte development, providing an optimal microenvironment in which developing thymocytes undergo proliferation, T-lymphocyte receptor rearrangement, and differentiation into mature T-lymphocytes, which are released into the circulation. Mature T-lymphocytes generate effective responses against pathogens and tumor antigens but are tolerant of self-antigens and harmless environmental antigens.^{1,5-9} A critical thymic function is generation of central tolerance occurring in 2 phases: positive selection in the thymic cortex, with apoptosis of T-lymphocytes that fail to recognize self-MHC molecules, and negative selection in the thymic medulla,⁶

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resulting in the removal of high affinity self-antigen-reactive T-lymphocytes that harbor the potential to elicit autoimmunity.

Negative selection is facilitated through central expression of a wide range of peripheral tissue-restricted self-antigens by medullary thymic epithelial cells, partly controlled by the autoimmune regulator transcription factor⁶ and Fezf2.¹⁰

A proportion of T-lymphocytes with high affinity to self-antigen survive negative selection and develop into regulatory T-lymphocytes (Tregs) related to antiapoptotic signals associated with FOXP3 expression. FOXP3 is expressed mainly by CD4⁺CD8⁻ T-lymphocytes, but expression has been observed in double-positive CD4⁺CD8⁺ T-lymphocytes, so the point at which the fate of these cells is determined remains unclear. Approximately 80% of the Treg cell repertoire originates in the thymus,¹¹ but Treg cells can be generated extrathymically by differentiation in the periphery (peripheral [inducible] Treg [iTreg] cells). As the properties and origin of these Treg cell populations differ, discriminating between them is important to better understand their specific functions in regulating immune homeostasis. Markers to differentiate thymic (natural) Treg (nTreg) cells from iTreg cells include expression of the transcription factor Helios, a member of the *Ikaros* transcription factor family, and the surface antigen neuropilin-1,¹² both of which are apparently highly expressed in nTreg cells. However, recent studies suggest that expression of these 2 markers does not unequivocally identify Treg cell clones of thymic or peripheral origin,¹³ and a specific lineage marker for Treg cells remains to be identified.

Various methods quantify thymic export of naive T-lymphocytes, including enumeration of T-lymphocyte receptor excision circles (TRECs) and specific cell surface markers. TRECs are stable circular episomal DNA segments generated as a by-product of T-cell receptor (TCR) gene rearrangement that is not duplicated during mitosis and is thus diluted with cellular division.^{7,14} Generation of circulating TREC-containing naive T-lymphocytes can persist for many months after thymectomy,^{15,16} and extrathymic TRECs may further complicate interpretation. Despite this, PCR quantification provides an accepted and practical measurement of thymic output by calculating the frequency of TRECs in a defined population of cells or absolute number of TRECs per milliliter of blood.¹⁵ The cell surface markers CD45RA, CD27, and CD62L have also been used to identify naive T-lymphocyte populations, although expansion can occur without loss of these markers. CD31⁺CD4⁺CD45RA⁺ T-lymphocytes have a high TREC content that declines with age, reflecting a decline in thymic function.^{15,17} CD45RA⁺CD27⁺ and CD45RA⁺CD31⁺ T-lymphocytes correlate strongly with TRECs in pediatric patients, suggesting that either panel can be used for enumerating peripheral blood naive T-lymphocytes.^{9,15,17,18}

Another indicator of normal thymopoiesis is a broadly diverse TCR repertoire that is necessary to respond adequately to a wide range of antigens.⁹ Quantification of TCRV β family T-lymphocyte expression is a useful tool to detect repertoire restriction, indicating limited T-lymphocyte pool diversity associated with reduced thymic output.^{7,9}

NORMAL AGE-RELATED THYMIC INVOLUTION AND IMMUNOSENESCENCE

The thymus reaches maximum volume and output during the first year of life, after which there is a rapid decline in thymic function between the ages of 1 and 8 years. This is followed by a

more gradual decline in function with increasing age.^{14,19} Age-related thymic decline is a chronic irreversible process¹⁹ attributed to a reduction in the number of developing intrathymic T-lymphocytes and alterations to the thymic stromal microenvironment, particularly with loss of thymic medullary epithelial cells and replacement by connective and adipose tissue.

The consequences of age-related thymic atrophy include decreased peripheral naive T-lymphocyte export and TCR repertoire contraction.⁶ The decline in thymopoiesis, and the life-long chronic antigenic stimulation result in peripheral memory T-lymphocyte predominance, with preservation of immune responses to “known antigens” but possible failure to respond to new antigens.²⁰ Recent evidence suggests the existence of heterogeneous populations of memory T-lymphocytes. In mice, central memory CD4⁺ T-lymphocytes contain subpopulations with different degrees of turnover. The maintenance of CD4⁺ T-lymphocyte memory subsets relies heavily on this recurrent replenishment of new cells sourced from the naive pool. If these observations are also relevant in humans, early thymectomy is likely to have a significant negative impact on replenishing the CD4⁺ T-lymphocyte memory pool.²¹

Accumulation of memory T-lymphocytes accounts for the shift from T_H1 to T_H2 cytokine profiles, promoting humoral immunity, production of autoantibodies, and potential of development autoimmunity. Other immunologic changes that have been reported include reduced T-lymphocyte proliferation due to a decline in IL-2 and IL-2 receptors.²² nTreg cell production is not impaired with age-related thymus atrophy, as decreased negative selection due to thymic atrophy is compensated by maintained, or increased, nTreg cell production.²³

Progressive thymic function decline, alterations to the peripheral T-lymphocyte compartment, and decline in T-lymphocyte function, as well as a shift in cytokine profiles, is attributed to the “immune risk phenotype,”²⁴ which is responsible for increased susceptibility to infection, cancer, and autoimmunity occurring with advancing age.²⁵ Elderly patients are predominantly at risk of cancers derived from tumor neoantigens, whereas immune responses to shared tumor antigens are well retained.²⁰ Although an increase in autoantibodies is observed in the elderly, this does not always translate into increased autoimmunity,²⁵ demonstrating that the precise mechanisms linking altered aging-related immune responses with development of autoimmune disease remain unknown.²⁴ Some studies suggest that contraction of the TCR repertoire may be not be as dramatic,^{20,26} and the continued, albeit reduced, thymic export of naive T-lymphocytes into the peripheral pool that continues into old age provides sufficient supply to maintain immunologic protection,^{27,28} suggesting involvement of other factors in the clinical consequences of immunosenescence.

IMMUNOLOGIC AND CLINICAL CONSEQUENCES OF INFANT THYMECTOMY

As long-term follow-up data become available, the immunologic impact of early pediatric thymectomy becomes appreciable (Table 1^{1,4,5,7,9,18,29-36}). Gradual CD4⁺, but also CD8⁺, T-lymphocytopenia develops.^{1,7,32} Progressive decline in levels of naive T-lymphocytes and TRECs is observed, with an increased proportion of peripheral memory T-lymphocytes mirroring the immunosenescent phenotype previously described.^{1,7,9} These changes are reversible if thymic regeneration occurs,^{31,32,37} emphasizing the

TABLE I. Summary of studies related to postthymectomy immunologic and clinical consequences

Study	Type of study and sample	Major findings
Eysteindottir et al ⁵ (2004)	Case-control study N = 38 (19 pediatric patients and 19 matched healthy controls)	Thymectomized patients present - Decrease in rate of T-cell production - Decrease in CD4⁺ and CD8⁺ T-cell counts, with a decrease in naive T-cell count and an increase in memory T-cell count - Significant decrease in IgA and IgG1 levels
Mancebo et al ⁷ (2008)	Prospective case-control study N = 23 pediatric patients; follow-up for first 3 y of life	No differences between patients with partial and complete thymic resection Thymectomized patients present - Low T-cell production; low CD4⁺ and CD8⁺ T-cell counts, with a decrease in naive T-cell counts and an increase in memory T-cell count - No impact on immunoglobulin levels - Expansions of effector phenotype in T cells - Reduced TCR Vβ repertoire - Reduced TREC count - Elevation in IL-7/plasma with negative correlation with CD4⁺ counts at 2 y after thymectomy
Prelog et al ¹⁸ (2009)	Case-control study N = 182 (101 pediatric patients and 81 matched healthy controls)	Thymectomized patients present - Decrease in naive T-cell count with a decrease in TRECs - Increase in KI-67 as a sign of peripheral proliferation of naive T-lymphocytes
Duarte et al ²⁹ (2009)	Experimental study nTreg cells transferred to syngeneic Rag2 ^{-/-} recipient mice to analyze stability of Foxp3 expression in these cells	The transferred nTreg cell loss of Foxp3 expression in lymphopenic conditions leading to tissue infiltration and damage converting into pathogenic T _H cells
Sauce et al ³⁰ (2009)	Case-control study N = 115 (29 healthy young adult, 35 middle-aged healthy controls, 26 elderly healthy adults, and 25 young adults [aged 18-26 y] who underwent thymectomy during childhood [15 d after birth])	Thymectomized patients present - Decreased CD4⁺ and CD8⁺ T-cell counts - Decrease in proportions of CD4⁺ and CD8⁺ naive T cells - Number of CD57-expressing cells equivalent between the thymectomized and elderly healthy group - Elevation in levels of the proinflammatory cytokines IL-1β, IL-8, and eotaxin in plasma compared with in age-matched controls - No impairment in development of memory T-cell populations specific for persistent virus
Van Gent et al ³¹ (2011)	Case-control study N = 193 (39 adults who underwent thymectomy during childhood [aged 0-1.5 y], 102 matched healthy children [aged 0-18 y], and 52 healthy volunteers [aged 21-39.7 y])	- During first 5 y of life after thymectomy, thymectomized patients presented a rapid decline in CD4⁺ and CD8⁺ T-cell counts compared with those in age-matched controls on account of a decline in naive cells - 5 y after thymectomy and onward, the CD4⁺ and CD8⁺ T cell total counts, and memory and naive levels, were normalized relative to those in age-matched healthy controls - Level of IL-7 was significantly elevated during the 2.5 y after thymectomy relative to that in healthy controls; 5 y after thymectomy, the percentages of proliferating naive CD4⁺ and CD8⁺ T cells were no longer elevated - TREC count in thymectomized patients declined during the 5 y after thymectomy, but this decline was not observed 5 y after thymectomy, indicating a possible <i>de novo</i> pool of naive T cells - Conclusion: thymectomy seems to clearly affect the T-cell compartment during the 5 y after thymectomy, but these changes seem to not be observed during later life; this restoration could be due to a regeneration of thymic tissue because of evidence of <i>de novo</i> naive T cells
Sauce et al ³² (2012)	Case-control study N = 205 (79 healthy controls [3 groups: old = 25, middle-aged = 34, and young = 20], 25 adults who underwent thymectomy during childhood [15 d after birth], and 101 HIV-infected patients [2 groups: untreated patients = 57 and treated patients aged >3 y = 44])	- Increased cell cycling of human naive T cells in lymphopenic donors (elderly and thymectomized) - Increased in IL-7 plasma levels in lymphopenic donors (elderly and thymectomized), supporting an active homeostatic role of this cytokine - Conclusion: there is evidence of a homeostatic regulation of the naive T-cell compartment in humans; the degree of cell cycling in this compartment is directly related to the frequency of donors' naive T cells; the lower their numbers, the higher their turnover - Strong CMV-specific T-cell response may cause marked alterations of the T-cell compartment in the context of lack of adequate thymic output

(Continued)

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Study	Type of study and sample	Major findings
Schadenberg et al ³³ (2014)	Retrospective study N = 26 pediatric patients	Thymectomized patients present - Decrease in CD3⁺CD4⁺, FOXP3⁺CD4⁺, and CD3⁺CD31⁺FOXP3⁺ cells in thymectomized patients - Increase in the proportion of FOXP3⁺CD4⁺ → compensatory peripheral proliferation - Increase in the proportion of both memory fractions of FoxP3 T cells → CD45RO⁺FOXP3^{high} and CD45RO⁺FOXP3^{low} cells
Elder et al ⁹ (2015)	Prospective case-control study N = 18 (10 adult patients [aged >18 y] with cardiac surgery and sternotomy and 8 adult patients [aged >18 y] with cardiac surgery without sternotomy)	Patients with cardiac surgery and sternotomy present - Low T-cell production; decrease in T-cell (CD4⁺ and CD8⁺) counts - Low naive T-cell count and increase in memory T-cell count - Abnormal/reduced TCR Vβ repertoire - No significant increase in exhaustion and immunosenescence markers
Gudmundsdottir et al ¹ (2016)	Prospective case-control study N = 22 (11 pediatric patients and 11 healthy age-matched controls)	Thymectomized patients present - Low T-cell production - Low CD4⁺ and CD8⁺ T-cell counts with a decrease in naive T-cell count and an increase in memory T-cell count - Abnormal TCR Vβ repertoire - Diminished TREC count
Morsheimer et al ³⁴ (2016)	Retrospective study N = 178 pediatric patients	Patients present - T-cell lymphopenia - Low naive T-cell count and increase in memory T-cell count - Difficulties in clearance of cutaneous viral infections
Ekman-Joelsson et al ⁴ (2017)	Retrospective study N = 71 pediatric patients	Overall incidence of posttransplant lymphoproliferative disease is 14% (10/71) Risk factors associated with lymphoproliferative disease - Sternotomy during first y of life (most commonly in first 2 wk) → one-third of these patients develop lymphoma - Mismatched EBV status - Use of tacrolimus after transplantation
Van den Broek et al ³⁵ (2017)	Prospective case-control study Pediatric patients aged 1-5 y (n = 10-27) and aged >10 y (n = 26), as well as healthy controls aged 1-5 y (n = 10-31) and aged >10 y (n = 11)	Alterations in the autoantibody repertoire early in life after thymectomy
Offor et al ³⁶ (2018)	Retrospective study N = 203 pediatric heart transplant recipients (aged <18 y)	- 15 of 203 patients presented with PTLD - All tumors were EBV positive and of B-cell lineage - Conclusion: early sternotomy results in dysregulated T-cell immunity and impaired response to EBV infection, increasing risk of development of PTLD

importance of sparing some thymic tissue at neonatal thymectomy. The frequency of Ki67-expressing naive T-lymphocytes increases in thymectomized patients compared with in age-matched healthy controls, reflecting increased peripheral replication in response to thymic output decline.¹⁸ Most studies suggest a steady gradual decline in cellular-mediated immunity following thymectomy. A seminal study looked at young adults aged 18 to 26 years who had been thymectomized in the first 2 weeks of life for surgery on transposition of the great arteries. The study subjects had CD4⁺ and CD8⁺ lymphocytopenia with a loss of naive cells, a reduction in diversity, and an increase in oligoclonal memory cells, with a switch to a proinflammatory cytokine profile. A subgroup of these patients had an altered T-lymphocyte profile normally related to cytomegalovirus (CMV) infection and leading to induction of strong CMV-specific T-lymphocyte responses. In the absence of adequate T-lymphocyte renewal from the thymus, these responses may deplete the naive T-lymphocyte pool. This is a pattern seen in elderly individuals, and it is indicative of an immune risk phenotype that is predictive of increased mortality.³⁰

An 18-year follow-up study of thymectomized patients demonstrated a quantitative naive T-lymphocyte decline, TCR

repertoire contraction and skewing reflecting a reduced naive T-lymphocyte peripheral pool with expansion of preexisting T-lymphocyte clones, and a potentially reduced T-lymphocyte proliferative ability indicated by shorter telomere length.¹ However, T-lymphocyte function, proliferative responses, and IFN-γ production were normal.⁷ Higher CD8⁺ T-lymphocyte expression of PD-1 (expressed following T-lymphocyte activation and used as a marker of exhaustion) was demonstrated in patients who had previously undergone sternotomy to repair congenital heart disease (CHD) when younger than 12 months age compared with in similar age-matched patients without sternotomy. This may reflect T-lymphocyte dysfunction, although the clinical significance is undetermined.⁹

An increased proportion of Treg cells within the CD4⁺ T-lymphocyte pool has been observed after thymectomy, possibly because of preferential Treg cell proliferation. A decline in CD4⁺CD25⁺CD127^{low}CD45RA⁺ naive Treg cells and increase in CD4⁺CD25⁺CD127^{low}CD45RO⁺ memory Treg cells with greater suppressing potential have been observed, which may reflect compensatory peripheral tolerance secondary to lack of postthymectomy central tolerance.¹ However, evidence from murine models suggests that in severe lymphopenia, FOXP3⁺ nTreg

cells can lose their suppressive capacity and revert to T_H cells that promote tissue infiltration and damage.²⁹ Whether this has relevance in humans is not clear. One study found an increased percentage of $FOXP3^+CD4^+$ T-lymphocytes, but predominantly comprising $CD45RO^+FOXP3^{high}$ suppressive Treg cells and $CD45RO^+FOXP3^{low}$ Treg cells shown to be nonsuppressive.³³ These data suggest a potential risk factor for the development of autoimmunity in patients thymectomized early in life.

One cohort of 178 patients with single-ventricle CHD and early thymectomy demonstrated difficulty clearing molluscum contagiosum and had a high incidence of human papillomata.³⁴ Generally, however, short-term immunologic impacts after thymectomy appear benign, as other studies with longer patient follow-up do not replicate these findings.

Absence of significant infections following neonatal thymectomy may be explained by the diverse naive T-lymphocyte reservoir present at birth, following normal fetal thymic development and function, which exists before thymic ablation and age-related decline. This may protect against serious infections in the short to medium term. In contrast, in congenital thymic aplasia, such as complete DiGeorge syndrome, an absent or restricted oligoclonal naive T-lymphocyte population at birth predisposes patients to significantly impaired adaptive immunity, presenting in infancy with severe, persistent, and opportunistic infection.

Longer-term follow-up in patients who have undergone cardiac thymectomy demonstrates early-onset immunosenescence predisposing to later-onset complications, including development of EBV-associated posttransplant lymphoproliferative disease (PTLD). A 14% incidence of PTLD was demonstrated in a pediatric cohort after cardiac transplantation, occurring in patients with CHD rather than cardiomyopathy,⁴ possibly reflecting the impact of early cardiac surgery in patients with CHD before transplantation. In another study, almost all patients who developed PTLD underwent surgery during infancy, suggesting that early thymectomy resulted in dysregulated T-lymphocyte immunity and impaired EBV responses.³⁶ Given the evidence that early thymectomy gives rise to an altered CMV-related T-lymphocyte profile in some patients,³⁰ it is possible that a similar disruption in anti-EBV T-lymphocyte immunity, possibly associated with a “second hit” such as prolonged immunosuppression following cardiac transplantation, renders some patients more susceptible to PTLD.

Humoral changes likely reflect changes in T-lymphocyte homeostasis. Immunoglobulin disturbances after thymectomy include reduced IgA and IgG subclass levels; however, alterations in B-lymphocyte subsets have not been studied.^{5,35} One study examined pneumococcal polysaccharide antibody response in pediatric patients who had received a cardiac transplant and were undergoing long-term immunosuppression. Patients who received a transplant before 4 years of age were more likely to have impaired pneumococcal polysaccharide antibody responses and low IgG2 antibody levels when older than were those who received a transplant after 4 years of age.³⁸ B-lymphocyte numbers were normal and immunosuppression regimens were similar in both groups, raising the possibility that altered T-lymphocyte help may be implicated.

The appearance of autoantibodies has been described in patients with early thymectomy, with detection of antinuclear antibodies or antineutrophil cytoplasmic antibodies in half of thymectomized patients in 1 cohort that increased with age,³⁵

although this finding is not replicated in all studies.⁵ Patients with autoantibodies had more memory T-lymphocytes than did those without autoantibodies and healthy controls. An increase in level of IgG autoantibodies, which are associated with autoimmune disease, and a decrease in level of IgM natural autoantibodies, which are associated with maintenance of self-tolerance, were observed, possibly reflecting diminished maintenance of self-tolerance. These studies mirror the aging immune system with thymic involution, suggesting early onset of immunosenescence.

Despite the presence of autoantibodies in thymectomized patients, clinical autoimmune disease is not frequent, possibly owing to preferential proliferation of Treg cells and an increase in their proportion after thymectomy, which suppresses autoreactivity.³⁵ However, as autoantibodies can arise long before clinical symptoms develop, further monitoring is necessary to determine whether clinical autoimmune disease develops later in life.

THE PARTIAL DiGeorge SYNDROME MODEL

DiGeorge syndrome, frequently caused by 22q11.2 deletion, is the most common, well-defined thymic primary immunodeficiency, with a spectrum of presentation ranging from athymia to normal thymic function. Most patients have partial DiGeorge syndrome associated with diminished but not absent thymic output. The immune changes described in these patients can be compared with those observed in thymectomized pediatric patients, but with consideration of the main difference between them, namely, the presence of a healthy pool of naive T-lymphocytes before surgery in thymectomized patients in contrast to the diminished, restricted pool of naive T-lymphocytes in patients with DiGeorge syndrome.

Patients with partial DiGeorge syndrome exhibit diminished $CD4^+$ and $CD8^+$ T-lymphocyte numbers, reduced naive T-lymphocyte and TREC numbers, and normal B-lymphocyte numbers but increased naive B-lymphocytes and perturbed memory B-lymphocytes.³⁹⁻⁴¹ TCR repertoire analysis demonstrates an increase in oligoclonal peaks and $V\beta$ family dropouts compared with controls, indicating TCR repertoire restriction similar to that described in thymectomized patients.⁴² However, patients with DiGeorge syndrome experience more infections and autoimmunity than have been reported in thymectomized patients. Of a large cohort of patients with partial DiGeorge syndrome, 64% had recurrent and/or severe infections and 7.8% had autoimmune disease with onset from 7.8 years of age.⁸ Dysphagia, anatomic palatal alterations, and gastrointestinal reflux were identified as predisposing factors related to upper respiratory tract infections, whereas autoimmune disease was related to $CD4^+$ T-lymphocytopenia and increased memory $CD4^+$ T-lymphocytes.³⁹ Clinical or laboratory immune abnormalities and autoimmunity were related to reduced $CD3^+CD4^+CD45RA^+CD31^+$ T-lymphocyte recent thymic emigrants, supporting the role of deficient thymopoiesis in development of autoimmunity in these patients.^{39,43} In 1 study, an increased risk of noncardiac death attributed to severe infection or proliferation was associated with reduced numbers of naive $CD4^+$ T-lymphocytes and $CD8^+$ cytotoxic T-lymphocytes.⁴⁴

Recent data describing changes in thymic structure and function may help explain, at least, the increased risk of autoimmunity: thymuses from patients with 22q11 deletion demonstrated hypoplasia, diminished maturation of the medulla,

a reduction in the number of autoimmune regulator–positive medullary thymic epithelial cells, and a reduced number of thymocytes.⁴⁵ These findings suggest that compromised thymic negative selection, impairing central tolerance and increasing the risk of autoimmunity, is also likely.

These findings contrast with those regarding patients with “partial FOXP1 deficiency” because of heterozygous mutations in *FOXP1*. Affected infants have low levels of TRECs and T-lymphocytopenia, and some have experienced recurrent infections, including severe infections. Unlike in patients with partial DiGeorge syndrome, autoimmunity does not appear to be a significant feature among infants or adults with heterozygous *FOXP1* deficiency.⁴⁶ This implies that T-lymphocytopenia alone does not predominantly drive autoimmunity, but impaired central tolerance is an important risk factor, as seen in patients with partial DiGeorge syndrome⁴⁵ and also in patients with hypomorphic mutations in *RAG1/2*, where T-lymphocytopenia is also associated with thymic dysfunction and impaired central and peripheral tolerance,^{47,48} as well as with a decrease in T-lymphocyte receptor diversity.^{49,50}

Patients with DiGeorge syndrome may have an increased malignancy risk, including lymphoma, neuroblastoma, acute lymphoblastic leukemia, osteosarcoma, Wilms tumor, thyroid carcinoma, and hepatoblastoma, although whether this is due to deletion of certain genes (eg, *SMARCB1* or *COMT*) related to immunodeficiency or other mechanisms remains unclear.^{51,52} Management recommendations for these patients include immunologic follow-up and indications for antimicrobial prophylaxis, vaccination, and immunoglobulin replacement where appropriate.^{53,54} Similar recommendations in “acquired” thymic deficiency are lacking.

THE MG MODEL

Symptoms of MG include muscle weakness and fatigue; they are due to a decrease in acetylcholine receptors at neuromuscular junctions secondary to autoantibodies directed against the acetylcholine receptors (AChRs) in the majority of patients or against neighboring proteins aiding the clustering of acetylcholine receptors. Around 75% of AChR antibody–positive patients with anti-AChR antibodies have thymic abnormalities, with germinal center formation or hyperplasia in approximately 65% or thymoma in approximately 11%.^{55,56} These thymic abnormalities facilitate the development of T-lymphocytes autoreactive to skeletal muscle proteins through presentation of cross-reactive epitopes by thymic epithelial cells.^{57,58} Thymectomy is second-line therapy for management of MG; the mechanisms by which patients respond to thymectomy are unclear,⁵⁹ although removal of the thymus may eliminate a source of continued antigen presentation, and thus reduction in T-lymphocyte–driven autoreactive B-lymphocyte stimulation. Because MG mainly affects adults, comparing data from this patient group with a pediatric population is challenging because of the differences in immunologic maturity. However, longer follow-up data on the MG population are available compared with the data on pediatric patients after cardiac surgery, providing important insights.

Decreased T-lymphocyte counts, reduced TCR V β repertoire and TREC concentration, increased Treg cell counts, and normal B-lymphocyte counts are observed during long-term follow-up in thymectomized patients with MG unlike in normal controls and in

nonthymectomized patients with MG.^{2,16,60} Severe infections have not been reported in thymectomized patients with MG.⁶¹

There is a high incidence of autoimmune disease after thymectomy,^{59,62} with increased IgG, IgM, anti-cardiolipin, and anti–double-stranded DNA antibodies in thymectomized patients with MG compared with in the other groups studied.² A recent study demonstrated a 6.25-fold increase in incidence of autoimmune diseases (including rheumatoid arthritis, SLE, and Sjögren syndrome) compared with in healthy controls. In contrast, patients with MG treated with plasmapheresis did not develop autoimmunity.³ This finding may support a link between thymectomy and subsequent development of autoimmune disease. However, patients undergoing thymectomy are more likely to have thymic abnormalities, including thymoma or germinal center hyperplasia (which may induce other autoimmunity in addition to MG), as well as to have less severe disease. The use of plasmapheresis in severely affected patients may clear other autoantibodies and mask other autoimmunity. Homeostatic regulation of T-lymphocytopenia may also have an impact on clinical consequences. T-lymphocyte proliferation in a lymphocytopenic environment occurs to rebalance T-lymphocyte homeostasis and try to maintain T-lymphocyte diversity and functionality. At least 2 forms of T-lymphocyte proliferation can be observed in murine models: a rapid spontaneous IL-7–independent proliferation that leads to broad differentiation of memory cells and a slow homeostatic IL-7–dependent proliferation that results in more limited differentiation.⁶³ The predominant method of proliferation in a particular clinical setting may influence the likely development of autoimmunity.

Less is known about the relationship between thymectomy for MG and the risk of developing malignancy; thymic and extra-thymic tumors have been described before,⁶⁴ concomitantly with, and after the diagnosis of MG. This makes it difficult to draw firm conclusions between the relationship between thymectomy for MG and subsequent malignancy risk, which is already increased with advancing age.⁶⁵

CONCLUSIONS

Most children with CHD survive into adulthood, and it is therefore important to understand the long-term consequences of early thymectomy. The immunologic effects may not be apparent until several decades later and may mimic physiologic immunosenescence associated with increased infections, autoimmunity, and malignancies, albeit occurring earlier than expected. The immunosenescent phenotype of thymectomized patients resembles that which occurs with normal aging. In the short and medium term, infection and autoimmunity do not appear to be increased, but the longest published follow-up data available are from 18 years after thymectomy. This population may be at future risk, and further monitoring may determine whether observed immunologic changes translate to clinical problems later in life.

However, the clinical consequences may depend on factors additional to the original surgery, including thymic regeneration following partial thymectomy, intrinsic thymic dysfunction leading to perturbed central and peripheral tolerance, and diminished T-lymphocyte receptor diversity (Fig 1) or slow homeostatic T-lymphocyte proliferation leading to reduced diversity. Additionally, in patients who subsequently undergo cardiac transplantation and long-term immunosuppression, the immunologic consequences of early thymectomy may be enhanced, with poor

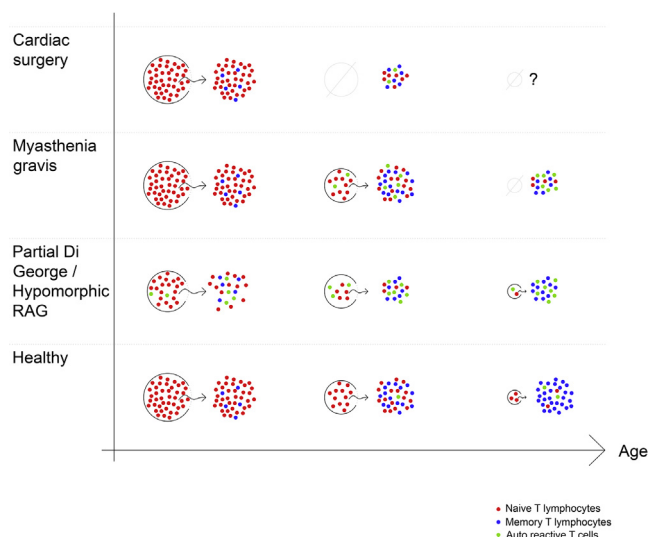


FIG 1. Possible models for development of infection risk or autoimmunity. In patients subject to thymectomy during infancy, early complete disruption of thymopoiesis may lead to peripheral T-lymphocyte expansion, with alterations in natural and induced regulatory T-lymphocyte populations, contraction of repertoire, and skewing to a memory phenotype, with failure to replenish the peripheral T-lymphocyte pool. In MG, onset is much later, leading to a normal peripheral T-lymphocyte pool until adulthood. Abnormal thymic architecture, possibly with alterations in peripheral tolerance may predispose to autoimmunity and infection after thymectomy. Disturbed central tolerance due to disrupted thymic medullary architecture leads to early-onset lymphocytopenia, restricted repertoire, and premature-onset autoimmunity in patients with partial DiGeorge syndrome and holomorphic RAG, whereas in healthy individuals, late-onset physiologic failure of thymopoiesis precedes peripheral T-lymphocyte expansion, enlargement of the T-lymphocyte memory pool, and gradual acquisition of autoreactive T-lymphocytes through homeostatic proliferation.

polysaccharide antibody responses and increased risk of PTLTD. Further studies may determine whether there is an increased risk of autoimmunity and identify the lifelong consequences of thymectomy during the neonatal period or infancy. In the meantime, efforts to preserve at least some thymic tissue during cardiac surgery should be advocated.

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