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Autoimmunity in Hyper-IgM Syndrome

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Abstract
Introduction Immunodeficiency with hyper-IgM (HIGM) results from genetic defects in the CD40–CD40 ligand (CD40L) pathway or in the enzymes required for immunoglobulin class switch recombination and somatic hypermutation. HIGM can thus be associated with an impairment of both B-cell and T-cell activation.
Results and discussions There are seven main subtypes of HIGM and the most frequent is X-linked HIGM, resulting from *CD40L* mutations. In addition to the susceptibility to recurrent and opportunistic infections, these patients are prone to autoimmune manifestations, especially hematologic abnormalities, arthritis, and inflammatory bowel disease. Furthermore, organ-specific autoantibodies are commonly found in HIGM patients.
Conclusions The mechanisms by which HIGM associates to autoimmunity are not completely elucidated but a defective development of regulatory T cells, the presence of IgM autoantibodies and an impaired peripheral B-cell tolerance checkpoint have been implicated. This article reviews the main subtypes of HIGM syndrome, the clinical autoimmune manifestations found in these patients, and the possible mechanisms that would explain this association.

Keywords Autoimmunity · CD40 · CD40L · hyper IgM · inflammatory bowel disease

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Immunodeficiency with hyper-IgM (HIGM) was first described in 1960 and refers to a group of disorders characterized by normal to increased serum IgM and very low or undetectable IgG, IgA, and IgE [1]. This immunological phenotype is mainly due to the failure of B cells to complete their maturation through immunoglobulin-isotype class switch recombination (CSR) and somatic hypermutation (SHM). Among primary hyper-IgM syndromes, genetic heterogeneity is supported by the existence of X-linked, autosomal recessive, and autosomal dominant inheritance [2]. HIGM can be caused by defects in CD40L–CD40 pathway, which is essential for B-cell activation, or by defects involving the enzymes required for CSR and SHM [3] (Table 1). The binding of CD40L to CD40 induces CD40 cytoplasmic domain binding to members of TNF receptor-associated-factor (TRAF) proteins. Further signaling by TRAF proteins is mediated by nuclear factor (NF)-kB, and activates pathways leading to immunoglobulin gene switching. Figure 1 shows possible mechanisms involved in HIGM [1, 4].

The X-linked variant of hyper-IgM syndrome (XHIM or HIGM1) is the most frequent subtype of HIGM (65–70%) and results from defects of the *CD40L* gene, which encodes for the CD40 ligand (CD154) molecule expressed transiently on the surface of activated T cells [3, 5]. XHIM patients have a normal number of circulating B lymphocytes which expresses IgM and IgD but not other isotypes at cell surface. About 50% of the XHIM have normal IgM levels at diagnosis, although the majority develops increased IgM

Table I Characteristics of the Main Hyper-IgM Syndromes

Hyper-IgM syndrome	Defective gene	Inheritance	Frequency (%)	Autoimmune manifestations
Type 1	*CD40L*	XL	70	+
Type 2	AID	AR or AD	<1	++
Type 3	*CD40*	AR	10	+
Type 5	*UNG*	AR	5	−
Type 6/HIGM-ED	*NEMO*	XL	1–2	++

CD40L CD40 ligand, *HIGM-ED* hyper-IgM with ectodermal dysplasia, *AID* activation-induced cytidine deaminase gene, *UNG* uracil DNA glycosylase gene, *NEMO* NF-κB essential modulator, *XL* X-linked, *AR* autosomal recessive, *AD* autosomal dominant, *single positive symbol* (+) mild association with autoimmunity, *double positive symbol* (++) moderate association with autoimmunity, *negative symbol* (−) no association with autoimmunity

during follow-up [1, 6]. In addition to defective isotype switching, XHIM is also characterized by low affinity antibody production in response to T-dependent antigens and lack of memory B-cell generation. Lymphnodes of these patients contain primary follicles, but lack germinal centers [1, 7, 8].

CD40L is a member of the tumor necrosis factor (TNF) family of cytokines composed of 261 amino acids and three functional domains (intracellular, transmembrane, and extracellular) [9]. The human *CD40L* gene includes five exons and mutations are more common in the TNF-homology domain encoded by exon 5 [10]. Recently, the first mutation in the promoter for CD40L gene was identified [11]. Its natural receptor is CD40, which is expressed on antigen presenting cells, including B cells, dendritic cells, and macrophages. Nonsense, missense, insertion, deletion, and splice-site mutations have been reported in XHIM patients, and some of these mutations have been shown to prevent binding of CD40L to CD40, while others affect folding of the CD40L monomer or trimer formation [12].

All major CD4 T-cell subsets express substantial amounts of CD154 after activation, including the naïve and memory CD4 T cells, and Th0, Th1, and Th2 cells. Activated CD4 T cells expressing CD154 engage CD40 on B cells, directing B-cell growth and differentiation, formation of germinal centers, upregulation of immunoglobulin heavy-chain transcription, and isotype switching [12]. CD40–CD40L interactions also provide a costimulatory signal for T cells, leading to T-cell activation and inducing CD8 T-cells effector functions, cytokine production by CD4 T cells, and humoral immunity to T-cell-dependent antigens [3, 13].

XHIM patients have an increased susceptibility to infections caused by bacteria, viruses, fungi, and parasites, reflecting their defect in T-lymphocyte function, as well as their hypogammaglobulinemia. These patients are uniquely susceptible to interstitial pneumonia caused by *Pneumocystis carinii* (20–40%) and present protracted chronic diarrhea caused by *Cryptosporidium* sp (30%). Liver disease as a whole is common in XHIM patients, which

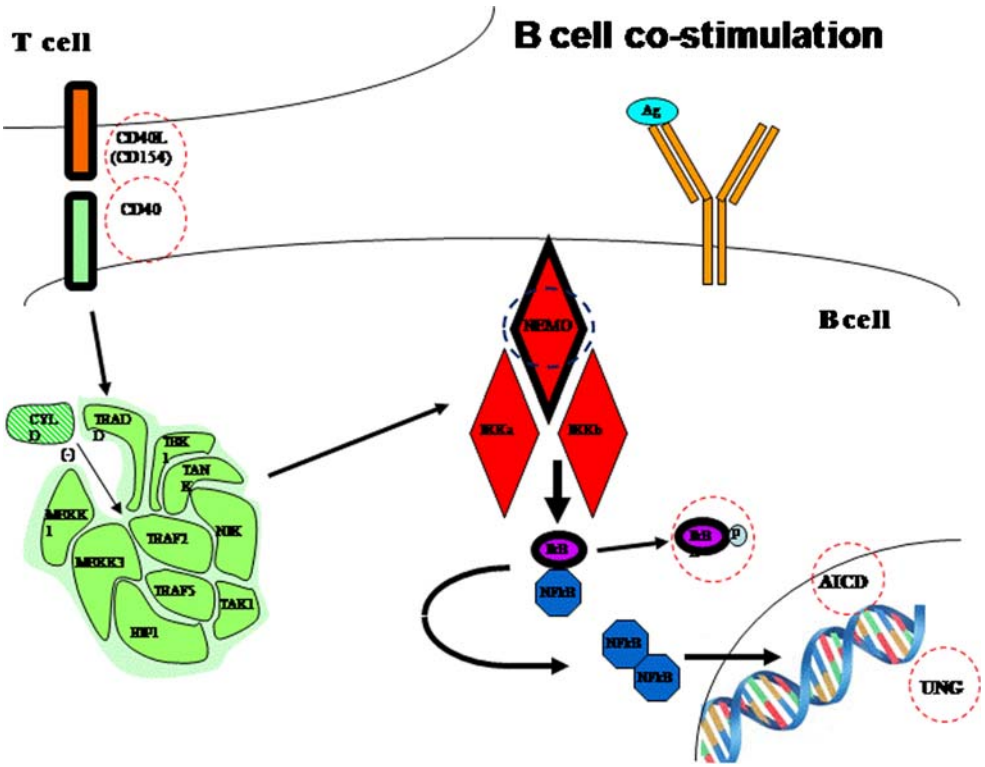

Fig. 1 Schematic illustration of the molecular defects leading to HIGM syndrome phenotype. When the interaction between activated T cell and B cells is impaired (CD40L and Cd40 deficiencies), B cells are unable to undergo class-switch recombination and affinity maturation. Mutations in downstream signaling molecules (NEMO and IkB) or intrinsic B cell enzymes (AICD and UNG) may also result in HIGM. *TRAF* TNF receptor-associated factor; *NF-κB* nuclear factor-κB; *IKKγ/NEMO* inhibitor of κB-kinase/NF-κB essential modulator; *AICD* activation-induced cytidine deaminase; *UNG* uracil DNA glycosylase; *IκBα* α inhibitor of NF-κB (adapted from Orange J, with permission)

frequently present sclerosing cholangitis (related to *Cryptosporidium parvum* infections), chronic hepatitis (due to hepatitis B virus, hepatitis C virus, and cytomegalovirus), and liver and biliary tract tumors. Other characteristic features are recurrent oral ulcers and proctitis, usually associated to neutropenia, observed in about 50% of the patients. Additionally, they may also develop autoimmune or inflammatory disorders and malignancies [1, 6, 14].

HIGM2 and HIGM3 are autosomal recessive variants of HIGM and are caused by mutations in the activation-induced cytidine deaminase gene, *AICDA* (AID, CDA2) and CD40 gene, *TNFRSF5*, respectively. Patients with HIGM3 are clinically undistinguishable from subjects carrying genetic defects of CD40L, while patients with HIGM2 present with a clinical phenotype characterized by enlargement of tonsils and lymphnodes and recurrent bacterial sinopulmonary infections, without increased susceptibility to opportunistic pathogens [2, 15].

HIGM4 is the least characterized subset, with patients clinically resembling a milder form of HIGM2 with residual IgG production. B cells from HIGM4 patients present defective CSR with normal SHM, and no genetic cause has been identified so far [14].

Mutations in *UNG* (uracil DNA glycosylase gene) have been found in a few patients clinically similar to HIGM2. This condition is termed HIGM5 and has an autosomal recessive inheritance. Mutations in *AICDA* and *UNG* are considered intrinsic B-cell defects, as they encode for enzymes involved in CSR and SHM [2].

Another type of HIGM affects males and is characterized by the association of hypogammaglobulinemia with hypohydrotic ectodermal dysplasia (HIGM-ED/HIGM6). This condition is caused by hypomorphic mutations in the NF-κB essential modulator (*NEMO*/*IKKG*) gene. More recently, a defect in another molecule of the same pathway, IκBα, was associated to Hyper IgM syndrome, and the disease named HIGM7. Both mutations result in a defective NF-κB translocation to the nucleus leading to an abnormal expression of multiple enzymes such as AID and UNG [2, 16].

Approximately 25% of HIGM patients have normal CD40L, CD40, AID, and UNG genes and some of these patients have been identified as having a defect in CSR [2, 13].

Autoimmune Manifestations in HIGM

In addition to the susceptibility to opportunistic and bacterial infections, HIGM patients are prone to develop autoimmune diseases such as immune thrombocytopenia, Coombs positive hemolytic anemia and nephritis, suggesting that tolerance is not correctly maintained in these patients [3]. It has been reported that sera from CD40L-deficient patients contain specific antibodies against self-antigens (antierythrocyte, antierythropoietin, antiplatelet, antismooth muscle, anticardiolipin, anti-Ro, anti-RNP, antinuclear, and antithyroid). Other autoimmune manifestations seen in these patients include inflammatory bowel disease, autoimmune hepatitis, seronegative arthritis, hypothyroidism, and discoid lupus erythematosus [2, 17–19].

Most data related to the frequency of autoimmune manifestations comes from XHIM patients. In the cohort of 56 patients with XHIM published by Levy et al. [6], inflammatory bowel disease was seen in 6% of the patients with protracted or recurrent diarrhea, seronegative arthritis in 11%, chronic neutropenia of unknown origin in 44.6%, and Coombs positive hemolytic anemia in 1 patient. Among 79 patients with XHIM registered by Winkelstein et al. [20], 60% presented neutropenia, while anemia and thrombocytopenia occurred in 15% and 4%, respectively. Parvovirus B19 infection was detected in only three patients (4%), raising the possibility of an autoimmune etiology for most of the hematologic manifestations. According to Webster et al. [21], from 62 XHIM patients registered at the European CD40L Defect Database until 1999, 11 presented arthritis as a manifestation, including a man with an aggressive form of polyarticular arthritis complicated by subcutaneous nodules and periarticular cysts. Taken together, these data suggest that autoimmune disease is a common and important feature of X-linked HIGM syndrome.

Regarding intrinsic B-cell defects, autoimmune manifestations occur in 25% of AID deficient patients, mainly hemolytic anemia, thrombocytopenia, and autoimmune hepatitis, and autoantibodies of the IgM isotype have also been described [14]. Also, Melegari et al. [22] described a woman with HIGM not related to CD40 or CD40L mutations, presenting systemic lupus erythematosus manifestations and autoantibodies against double strand-DNA and ribonucleoprotein (RNP).

Among 29 HIGM patients due to AID deficiency reported by Quartier et al., six presented autoimmune disorders, including a 21-year-old male with marked splenomegaly, oral ulcers, thrombocytopenia, and diabetes mellitus. Another patient presented a chronic, destructive, bilateral, and symmetrical polyarthritis. Other autoimmune manifestations observed in this study were autoimmune hepatitis (AIH), hemolytic anemia, and thrombocytopenia in a patient who presented several autoantibodies of IgM isotype (antihepatocyte membrane, antiliver–kidney microsome, antismooth muscle, anticardiolipin, antierythrocyte, and antiplatelets). Chronic hepatitis was observed in another patient and autoimmune etiology in this case was supported by liver histological findings, negative infection screening, and by the efficacy of corticosteroids and immunosuppressive therapy, since AIH autoantibodies were not detected. Other interesting manifestations were inflam-

matory bowel disease mimicking Crohn's disease in one patient and bilateral chronic uveitis in other [23].

Autoimmune manifestations were also reported in patients with HIGM6, related to NEMO defects. Orange et al. [24] described 13 NEMO patients presenting autoimmune manifestations, including inflammatory bowel disease in 10, arthritis in two and autoimmune hemolytic anemia in one.

Thus, it appears that autoimmunity occurs in all types of HIGM syndrome, with variable presentation according to the underlying genetic defect.

Mechanisms of Autoimmunity in HIGM

CD40–CD40L interactions may be involved in the selection of T-cell repertoire, since autoimmune diseases (thyroiditis, syaloadenitis, pancreatitis, oophoritis, and adrenalitis) can be elicited by transferring T cells from CD40-deficient mice into athymic nude mice that lack T cells but have B cells and other APCs expressing CD40 [24]. It was also observed a severe reduction of CD25+CD45RB^{lo} CD4+ T-cell numbers and impaired differentiation of Tr1 cells in CD40-deficienct mice, indicating that defective development of regulatory T cells may result in increased T-cell autoreactivity in the absence of CD40–CD40L interactions [3, 25].

The use of CD40L blocking antibodies in experimental animal models has also contributed to the comprehension of XHIM. These antibodies have demonstrated to be beneficial in murine collagen arthritis, thyroiditis, and experimental allergic encephalomyelitis [10]. Clonal deletion of thymocytes bearing T-cell receptors of low affinity is also altered by the functional blockage of CD40L by anti-CD40L antibodies [10].

Lacroix-Desmazes et al. [18] demonstrated that the IgM reactivity repertoire is skewed toward self-antigens in 19 HIGM patients as compared to normal subjects, while the repertoire of reactivity against foreign antigens did not differ between the two groups. Additionally, IgG antibodies of HIGM patients lacked reactivity with self antigens, in contrast with IgG of healthy controls.

Although the molecular mechanisms by which CD40L induces B-cell proliferation and differentiation are well described, its potential functions in counterselecting human autoreactive B cells are poorly understood. Transgenic mouse models have suggested that CD4 + T cells play an important role in the elimination of peripheral autoreactive B cells, and that CD40–CD40L and MHC class II T-cell receptor are required for the counterselection of transgenic autoreactive B cells [3].

In humans, auto-reactive B cells are removed during central and peripheral B-cell tolerance checkpoints. Whereas central B-cell tolerance involves B-cell receptor signaling pathways, the mechanisms involved in peripheral B-cell tolerance are not well characterized [3, 26]. Hervé et al. tried to determine the impact of CD40L and MHC class II expression on human B-cell tolerance through cloning antibodies from CD40L or MHC class II deficient patients. This study concluded that polyreactive B cells are properly counterselected in the bone marrow of CD40L-deficient patients, revealing a functional central B-cell tolerance checkpoint in the absence of CD40L expression [3]. In contrast, the peripheral B-cell tolerance mechanism seems to be defective in CD40L-deficient patients, resulting in an increase of circulating polyreactive B cells [26]. The same study also concluded that MHC class II is essential in the development of class-switched memory B cells in humans, since MHC class II deficient patients (bare lymphocyte syndrome-BLS) lacked an IgM^{-}CD27^{+} class-switched B-cell population [3].

A third conclusion was a significant decrease in the proportion of CD25^{+}Foxp3^{+}T reg cells in CD40L-deficient patients as compared to the normal controls. Finally, serum BAFF (a potent B-cell survival factor) levels were significantly increased in CD40L-deficient and BLS patients [3].

The CD40 molecule has also been implicated in the autoimmunity found in HIGM syndrome. Since CD40 is expressed not only on B cells, but also on all other APCs, the CD40–CD154 interaction may be important in T-cell-dependent macrophage-mediated immune response. Ligation of CD40 on the surface of monocytes induces the secretion of several cytokines, such as IL-1, IL-6, IL-8, IL-10, IL-12, and TNF-α. These cytokines promote the maturation of dendritic cells, enhancing their ability as APCs. Moreover, as DCs are the main producers of cytokines that regulate T-cell activation like IL-10 and IL-12, CD40-deficient patients may present an imbalance in the production of these cytokines [2].

CD40–CD40L System and Systemic Lupus Erythematosus

Although CD4 T cells do not play a direct role in systemic lupus erythematosus (SLE) tissue damage, they appear to be required for the production of pathogenic autoantibodies. An increased CD154 expression on T cells of lupus-prone mice has been reported, and treatment of these mice with a neutralizing anti-CD154 has been shown to delay and reduce the incidence of glomerulonephritis [12, 27]. In patients with SLE, it has been shown that CD40 expression is markedly increased in kidney and is associated with the presence of infiltrating CD154-expressing mononuclear cells [12]. Desai-Mehta et al. demonstrated increased frequencies of CD154 expressing CD4 T cells among patients with active SLE as compared to those in remission or normal controls. It was also shown that an antibody against CD154 significantly blocked the ability of lymphocytes from active SLE patients

to produce pathogenic antinuclear antibodies in vitro. Soluble CD154 (sCD154) has been also detected in SLE patients and levels of sCD154 correlated with disease activity and anti-dsDNA titers [28].

Dysregulated expression of CD154 has been correlated with other autoimmune diseases such as inflammatory bowel disease (IBD), rheumatoid arthritis (RA), and systemic sclerosis (SS). CD154 has been shown to be expressed by T cells in the joints and peripheral blood of RA patients and in the sera of RA-associated vasculitis patients. Hyperexpression of CD154 by CD4 T cells in IBD intestinal lesions may contribute to pathogenic cytokine production in this disease. However, CD154 expression appears to be normal in patients with idiopathic thrombocytopenic purpura (ITP), suggesting that CD154 is not always overexpressed in autoimmune disorders [12].

Conclusions

HIGM syndrome can be caused by several defects involving the CD40L–CD40 pathway, NEMO, AID, UNG, and other unknown defects, all of them involved in a proper immunoglobulin production by B cells. In addition to the immunodeficiency state leading to recurrent or opportunistic infections, these patients are susceptible to autoimmune manifestations. The association between autoimmunity and X-linked HIGM could be explained by presence of IgM autoantibodies, decreased peripheral control of B lymphocytes, impaired development of T reg cells and increased levels of BAFF (also implicated in peripheral tolerance), as shown by recent studies.

References

1. Notarangelo LD, Hayward AR. X-linked immunodeficiency with hyper-IgM (XHIM). Clin Exp Immunol 2000;120:399–405.
2. Lougaris V, Badolato R, Ferrari S, Plebani A. Hyper immunoglobulin M syndrome due to CD40 deficiency: clinical, molecular, and immunological features. Immunol Rev 2005;203:48–66.
3. Hervé M, Isnardi I, Ng YS, Bussel JB, Ochs HD, Cunningham-Rundles C, et al. CD40 ligand and MHC class II expression are essential for human peripheral B cell tolerance. J Exp Med 2007;204:1583–93.
4. Hess S, Engelmann H. A novel function of CD40: induction of cell death in transformed cells. J Exp Med 1996;183:159–67.
5. Etzioni A, Ochs HD. The hyper IgM syndrome—an evolving story. Pediatr Res 2004;56:519–25.
6. Levy J, Espanol-Boren T, Thomas C, Fischer A, Tovo P, Bordigoni P, et al. Clinical spectrum of X-linked hyper-IgM syndrome. J Pediatr 1997;131:47–54.
7. Facchetti F, Appiani C, Salvi L, Levy J, Notarangelo LD. Immunohistologic analysis of ineffective CD40–CD40 ligand interaction in lymphoid tissues from patients with X-linked immunodeficiency with hyper-IgM. Abortive germinal center cell reaction and severe depletion of follicular dendritic cells. J Immunol 1995;154:6624–33.
8. Seyama K, Nonoyama S, Gangsaas I, Hollenbaugh D, Pabst HF, Aruffo A, et al. Mutations of the CD40 ligand gene and its effect on CD40 ligand expression in patients with X-linked hyper IgM syndrome. Blood 1998;92:2421–34.
9. Notarangelo LD, Lanzi G, Peron S, Durandy A. Defects of class-switch recombination. J Allergy Clin Immunol 2006;117:855–64.
10. van Kooten C, Banchereau J. CD40–CD40 ligand. J Leukoc Biol 2000;67:2–17.
11. Van Hoeyveld E, Zhang PX, De Boeck K, Fuleihan R, Bossuyt X. Hyper-immunoglobulin M syndrome caused by a mutation in the promotor for CD40L. Immunology 2007;120:497–501.
12. Cron RQ. CD154 transcriptional regulation in primary human CD4 T cells. Immunol Res 2003;27:185–202.
13. Durandy A, Peron S, Fischer A. Hyper-IgM syndromes. Curr Opin Rheumatol 2006;18:369–76.
14. Durandy A, Revy P, Imai K, Fischer A. Hyper-immunoglobulin M syndromes caused by intrinsic B-lymphocyte defects. Immunol Rev 2005;203:67–79.
15. Revy P, Muto T, Levy Y, Geissmann F, Plebani A, Sanal O, et al. Activation-induced cytidine deaminase (AID) deficiency causes the autosomal recessive form of the Hyper-IgM syndrome (HIGM2). Cell 2000;102:565–75.
16. Jain A, Ma CA, Liu S, Brown M, Cohen J, Strober W. Specific missense mutations in NEMO result in hyper-IgM syndrome with hypohydrotic ectodermal dysplasia. Nat Immunol 2001;2:223–8.
17. Gulino AV, Notarangelo LD. Hyper IgM syndromes. Curr Opin Rheumatol 2003;15:422–9.
18. Lacroix-Desmazes S, Resnick I, Stahl D, Mouthon L, Espanol T, Levy J, et al. Defective self-reactive antibody repertoire of serum IgM in patients with hyper-IgM syndrome. J Immunol 1999;162:5601–8.
19. Seyama K, Kobayashi R, Hasle H, Apter AJ, Rutledge JC, Rosen D, et al. Parvovirus B19-induced anemia as the presenting manifestation of X-linked hyper-IgM syndrome. J Infect Dis 1998;178:318–24.
20. Winkelstein JA, Marino MC, Ochs H, Fuleihan R, Scholl PR, Geha R, et al. The X-linked hyper-IgM syndrome: clinical and immunologic features of 79 patients. Medicine (Baltimore) 2003;82:373–84.
21. Webster EA, Khakoo AY, Mackus WJ, Karpusas M, Thomas DW, Davidson A, et al. An aggressive form of polyarticular arthritis in a man with CD154 mutation (X-linked hyper-IgM syndrome). Arthritis Rheum 1999;42:1291–6.
22. Melegari A, Mascia MT, Sandri G, Carbonieri A. Immunodeficiency and autoimmune phenomena in female hyper-IgM syndrome. Ann N Y Acad Sci 2007;1109:106–8.
23. Quartier P, Bustamante J, Sanal O, Plebani A, Debré M, Deville A, et al. Clinical, immunologic and genetic analysis of 29 patients with autosomal recessive hyper-IgM syndrome due to activation-induced cytidine deaminase deficiency. Clin Immunol 2004;110:22–9. Erratum in: Clin Immunol. 113:220, 2004.
24. Orange JS, Levy O, Geha RS. Human disease resulting from gene mutations that interfere with appropriate nuclear factor-kappaB activation. Immunol Rev 2005;203:21–37.
25. Kumanogoh A, Wang X, Lee I, Watanabe C, Kamanaka M, Shi W, et al. Increased T cell autoreactivity in the absence of CD40–CD40 ligand interactions: a role of CD40 in regulatory T cell development. J Immunol 2001;166:353–60.
26. Rathmell JC, Townsend SE, Xu JC, Flavell RA, Goodnow CC. Expansion or elimination of B cells in vivo: dual roles for CD40- and Fas (CD95)-ligands modulated by the B cell antigen receptor. Cell 1996;18(87):319–29.
27. Datta SK, Kalled SL. CD40–CD40 ligand interaction in autoimmune disease. Arthritis Rheum 1997;40:1735–45.
28. Desai-Mehta A, Lu L, Ramsey-Goldman R, Datta SK. Hyperexpression of CD40 ligand by B and T cells in human lupus and its role in pathogenic autoantibody production. J Clin Invest 1996;97:2063–73.