

# Potential Immunopathogenic Role of the Mucosa–Bone Marrow Axis in IgA Nephropathy: Insights From Animal Models

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**Summary:** Impaired immune regulation along the mucosa–bone marrow axis has been postulated to play an important role in the pathogenesis of IgA nephropathy. Animal models have allowed us to study such changes in detail. Accumulating evidence from a number of animal models suggest that there is dysregulation of innate and cellular immunity in IgA nephropathy, resulting in changes to the mucosal immune system. These changes appear to be linked closely to a disruption of mucosal tolerance, resulting in the abnormal priming and dissemination of cells to sites such as the bone marrow where they are responsible for the synthesis of nephritogenic IgA. These findings suggest that future treatment strategies should focus on manipulating the priming and dissemination of these memory cells to prevent the appearance of nephritogenic IgA in the systemic compartment.

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IgA nephropathy (IgAN) is one of the most frequent forms of glomerulonephritis worldwide, accounting for 25% to 50% of patients with primary glomerulonephritis. Although this condition initially was considered a benign chronic nephropathy, accumulating evidence suggests that 30% to 40% of cases progress to end-stage renal disease by 20 years.<sup>1-3</sup> There remains, however, no clear treatment strategy, principally because of the lack of a comprehensive understanding of the pathogenesis of this disease.

IgAN is defined by deposition of IgA in the glomerular mesangium.<sup>4</sup> Despite a single histopathologic definition, patients with IgAN can have variable clinical and histopathologic features. This heterogeneity may be one of

the major reasons why the pathogenesis of this disease remains unclear. Several clinical studies have identified the importance of IgA or IgA immune complex (IgA-IC) deposition as a fundamental causative factor in IgAN. The observed clinicopathologic heterogeneity may be dependent, at least in part, on the characteristics of the deposited IgA-IC itself or changes in the IgA immune system, including sites of IgA synthesis and stimulation and regulation of immune competent cells involved in the production of IgA.<sup>5</sup> We believe that understanding the mechanisms involved in the generation of nephritogenic IgA will help us explain the clinicopathologic heterogeneity characteristic of IgAN, and also may clarify the molecular mechanisms occurring after IgA deposition, including the expression of IgA receptors by intrinsic renal cells.<sup>6</sup> The recurrence of IgA deposition in renal allografts<sup>7</sup> and the disappearance of IgA deposits from renal allografts taken from donors with undiagnosed IgAN,<sup>8,9</sup> reinforce the importance of systemic abnormalities of the IgA

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immune system in IgAN, arguing against IgAN being a disease limited to intrinsic renal abnormalities.

What are the major changes in the IgA immune system in IgAN? First, there are important changes in the physicochemical properties of the IgA molecule in IgAN (discussed further by Novak, pp. 78-87). High levels of polymeric IgA (pIgA) are present in serum and tonsillar tissues of patients with IgAN. In addition, it generally is accepted that mesangial IgA deposits consist primarily of underglycosylated pIgA1.<sup>5</sup> Second, mucosal vaccination results in impaired mucosal IgA responses in IgAN whereas systemic antigen challenge results in increased titers of circulating pIgA1 antibodies with normal levels in mucosal secretions.<sup>10,11</sup> Moreover, large numbers of pIgA1-positive plasma cells are found in the bone marrow in IgAN.<sup>12</sup> These clinical findings suggest that overproduction of pIgA1 in IgAN seems to be based in systemic immune sites, such as the bone marrow. Finally, many patients with IgAN show episodic macroscopic hematuria, which coincides with mucosal infection, often of the upper respiratory tract,<sup>13,14</sup> and there is evidence that tonsillectomy has a favorable effect on long-term renal survival in IgAN patients.<sup>15</sup> A common feature in all these clinical observations is that abnormal immune responses in both the mucosa and bone marrow with dysregulation of the mucosal immune system may play a key role in the pathogenesis of IgAN.<sup>16</sup>

Almost 20 years ago, Van Es and his colleagues, in a series of elegant studies, identified impaired IgA immune responses in the mucosa-bone marrow axis in IgAN.<sup>11,12,17,18</sup> In the past decade, clinical and experimental studies have revealed continued trafficking of antigen-specific lymphocytes and antigen-presenting cells between the mucosa and bone marrow in human beings.<sup>19</sup> IgA plasma cells primed in mucosal sites, such as salivary glands and tonsils, routinely traffic to the bone marrow and back to the site of antigen encounter.<sup>19</sup> This migration is directed by the local synthesis of specific chemokines and appropriate adhesion/homing-receptor engagement. There is increasing recognition for the presence of a mucosa-bone marrow axis in human beings, and

abnormalities in this axis may play an important role in the development of IgAN.<sup>16</sup>

How then can we study the mucosa-bone marrow axis in IgAN and correlate any changes with the well-documented clinical features of this disease? Undoubtedly there are a number of challenges to the study of this dynamic and complex immune axis in human beings; a number of investigators therefore have used experimental animal models. Although there are interstrain differences in murine IgA immune responses and the ability to induce glomerulonephritis, animal models still serve a useful purpose in investigating the pathogenesis of IgAN. In this review, we describe how rodent animal models have provided a better understanding of the mucosa-bone marrow axis and how these data may be applicable to human IgAN.

#### **DEFINING THE CHARACTERISTICS OF NEPHRITOGENIC IgA AND IgA-IC AND THE POSSIBLE CONTRIBUTION OF ENDOGENOUS ANTIGENS IN IgAN**

Rifai et al<sup>20</sup> described the first animal model of IgAN in 1979. By using murine antidinitrophenole and dinitrophenole-conjugated bovine serum albumin, they generated circulating IgA-IC and showed that these complexes were prone to mesangial deposition. They also found that for mesangial deposition to occur, IgA-IC needed to be either administered repeatedly or to be present persistently in the circulation.<sup>20</sup> In subsequent studies, the same group showed the importance of IgA-IC size in mesangial deposition by studying different pIgA-antigen complexes.<sup>21,22</sup> In the early 1980s, Isaacs et al<sup>23,24</sup> confirmed the importance of circulating IgA-IC in mesangial IgA deposition and initiation of glomerular injury. In these studies, animals were immunized with a bacterial-derived polysaccharide or chemically modified dextran. These studies emphasized both the importance of continual IgA-IC formation as a driver for mesangial IgA deposition and progression of IgAN, and the critical role played by pIgA in the formation of circulating nephritogenic IgA-IC.

It remains unclear whether these nephritogenic circulating and mesangial IgA-ICs contain exogenous antigens. Evidence from several experimental studies suggests a potential patho-

logic role of pIgA complexed with endogenous protein in the induction of IgAN. Studies in patients with IgAN have shown an increased binding of circulating IgA to the Fc $\alpha$  receptor (CD89), despite down-regulation of monocyte CD89 expression (discussed further by Moura, pp. 88-95).<sup>25</sup> Based on these findings, Monteiro et al generated transgenic mice overexpressing human CD89 on monocytes/macrophages and found that these mice developed IgAN in association with the appearance of large IgA-IC in the circulation.<sup>26</sup> These IgA-IC were found to contain IgA complexed with soluble CD89.

Fibronectin (Fn) and collagen co-deposition with IgA have been reported in IgAN. Moreover, high levels of plasma IgA-Fn complexes have been found in 48% to 68% of patients with IgAN.<sup>27</sup> In this regard, it is interesting that 2 independent uteroglobin (UG)-deficient mouse models display a number of features characteristic of human IgAN including high serum IgA-Fn complex levels.<sup>28</sup> Uteroglobin is a steroid-inducible cytokine-like protein with immunomodulatory and anti-inflammatory properties and a high affinity for Fn. In wild-type mice, UG forms Fn-UG heterodimers and thereby prevents both Fn self-aggregation and IgA-Fn association.<sup>29</sup> Coppo et al<sup>30</sup> could not, however, show a decrease in circulating levels of UG in IgAN despite the presence of increased IgA-Fn complexes and the presence of UG in IgA-Fn complexes in IgAN patients.

It is widely accepted that mesangial IgA is predominantly IgA1 and displays abnormal O-glycosylation.<sup>31,32</sup> Although the functional significance of the changes in IgA1 glycosylation remain imprecisely understood, it has been shown that they are associated with the development of autoantibodies against the altered IgA1 hinge region, suggesting that IgA1 itself could be an endogenous antigen in IgAN.<sup>33,34</sup> Furthermore, underglycosylated IgA has a tendency to self-aggregate and is capable of binding to human mesangial cells.<sup>35,36</sup> In this regard, the HIGA (high IgA levels) mouse, which is an inbred strain established by Muso et al<sup>37,38</sup> by selective mating of the spontaneous IgAN-prone ddY mouse, also displays abnormal glycosylation of serum IgA.<sup>39</sup> It also is worth noting that autoimmune-prone (New Zealand White

[NZW]  $\times$  C57BL/6) F1 mice that overexpress human Bcl-2 in B cells display IgA hyperglobulinemia and develop a fatal glomerulonephritis with glomerular IgA deposition.<sup>40</sup> Although circulating immunoglobulin-containing immune complexes were not evaluated, serum IgA purified from this mouse displays reduced levels of galactosylation and sialylation. These experimental findings suggest that aberrant glycosylation of serum IgA may be involved in the induction of IgAN in both mice and human beings. This notion has been reinforced by the recent work of Nishie et al.<sup>41</sup> They showed that mice lacking  $\beta$ -1, 4-galactosyltransferase-I spontaneously developed IgAN and had increased serum polymeric IgA levels. This enzyme transfers galactose to the terminal N-acetylglucosamine of N- and O-linked glycans in a  $\beta$ -1, 4 linkage, and these transgenic mice displayed complete absence of  $\beta$ 4 galactosylation and sialylation of the IgA N-glycans.

#### **DETERMINING THE MAJOR SITE OF NEPHRITOGENIC IgA PRODUCTION AND THE POTENTIAL ROLE OF THE BONE MARROW IN IgAN**

Despite extensive study the major site of pathogenic IgA production in IgAN remains uncertain. Previous clinical studies have reported the presence of large numbers of IgA1 plasma cells in the bone marrow (BM) in IgAN.<sup>12,42</sup> Moreover, BM transplantation or transplantation of peripheral blood stem cells in patients with leukemia and IgAN is associated with cure of both leukemia and IgAN.<sup>43,44</sup> These clinical observations suggest that mucosal-type pIgA1 may be derived from BM, although the origins of the cell(s) responsible for synthesis of this IgA remain unclear. Imasawa et al<sup>45</sup> reported that transfer of BM from wild-type mice attenuated glomerular lesions in HIGA mice, conversely when wild-type mice were transplanted with HIGA BM they developed mesangial IgA deposition and glomerulonephritis. The investigators suggested that IgAN may in part be a disorder of stem cell function.

The ddY mouse is a well-known model of spontaneous IgAN, which first was described by Imai et al.<sup>46</sup> These mice develop glomerulonephritis with striking deposition of IgA in the

mesangium along with IgG, IgM, and C3 co-deposition. Because the ddY mouse is not an inbred strain, studies using this mouse and the HIGA mouse must take into account the wide variability in onset of spontaneous IgAN. Indeed, serum IgA levels do not correlate with the severity of glomerular injury and incidence of the disease in this mouse.<sup>47</sup> We recently reported that ddY mice can be classified into 3 groups, early onset (~20 wk; 35%), late onset (~40 wk; 35%), and quiescent groups (30%) by grading of glomerular lesions and IgA deposition on serial biopsy specimens.<sup>47</sup> Serum levels of IgA were not different in the 3 groups and did not correlate with the degree of glomerular IgA deposition. A genome-wide association analysis of early active and quiescent mice indicated that the susceptibility to murine IgAN is regulated in part by specific loci syntenic to the *IGAN1* gene, a known candidate gene of human familial IgAN.<sup>48</sup> These results suggest that in this model of IgAN disease susceptibility may be regulated, at least in part, by the same genes involved in development of human IgAN and supports the use of this *grouped ddY mice* model for examining the pathogenesis of IgAN.<sup>47</sup>

We have now inbred the early onset mice and have ddY mice that almost universally develop an IgAN phenotype (nearly 100%, our unpublished data). By using this model, we have confirmed that transfer of BM from these early onset mice to wild-type control mice results in the development of IgAN.<sup>49</sup> The early onset ddY mice also display co-deposition of IgA and IgG, and have serum IgA-IgG2a IC levels that correlate with the severity of glomerular lesions. Similar findings are seen in recipient wild-type mice. By contrast, BM transfer from quiescent or wild-type control mice to early onset ddY mice abrogates glomerular injury and mesangial IgA/IgG co-deposition. These findings suggest that the BM may be a reservoir of memory cells capable of synthesizing IgA with a propensity for mesangial deposition and triggering of GN. These BM-located cells appear to be essential for the continuous delivery of pathologic IgA.

To further investigate the role of these BM cells in the development of IgAN, we transplanted BM cells from early onset mice to aly-

phoplasia mice (*aly/aly*). These mice have a point mutation in the nuclear factor  $\kappa$  B-inducing kinase gene and consequently lack Peyer's patches, lymph nodes, and IgA-producing cells and therefore have no serum or fecal IgA.<sup>50</sup> At 12 weeks post-BM transplantation, we observed an increase in serum IgA and an increased number of IgA<sup>+</sup> B220<sup>+</sup>CD138<sup>+</sup> plasma cells in the BM, but no IgA-producing cells in the lamina propria. Both transplanted *aly/aly* and wild-type mice showed mesangial IgA deposition, however, glomerular lesions with IgA/IgG2a co-deposition, high levels of serum IgA/IgG2a immune complex, and helper T cell (Th)1 polarization were detected only in wild-type control mice.<sup>51</sup> These findings suggest that BM cells are capable of synthesizing IgA with a propensity for mesangial deposition independent of homing to mucosal or secondary lymphoid tissues. However, in this model disease progression may require additional priming in secondary lymphoid tissues.

In human beings, mild mesangial IgA deposition is not always accompanied by proteinuria and hematuria. Transient deposition of glomerular IgA perhaps may be considered a physiologic phenomenon.<sup>52,53</sup> Moreover, in secondary IgAN<sup>54</sup> as seen in patients with cirrhosis,<sup>55</sup> portal systemic shunts,<sup>56</sup> dermatitis herpetiformis,<sup>57</sup> celiac disease,<sup>58</sup> and chronic inflammatory disease of the lung,<sup>59</sup> glomerular IgA and C3 deposition without obvious glomerular lesions are observed frequently. Therefore, our findings are consistent with an uncoupling of mesangial IgA deposition and initiation and perpetuation of glomerular injury. In addition, the lack in increase of serum IgA/IgG-IC in transplanted *aly/aly* mice highlights the potential role of IgA as an autoantigen in IgAN and the importance of clearly defining the characteristics of nephritogenic IgA in the development of IgAN.

#### DETERMINING THE IMPORTANCE OF MUCOSAL IMMUNE PRIMING IN THE GENERATION OF NEPHRITOGENIC IgA

B-lineage cells in the BM are one of the major sources of nephritogenic IgA synthesis in IgAN. It remains unclear, however, whether these cells are native BM cells or cells that have en-

countered antigen at other sites and subsequently relocated to the BM. The association of episodic macroscopic hematuria with mucosal infections in IgAN is suggestive of changes to the mucosal immune system, which may include changes in antigen handling, in this disease.<sup>16</sup> The results of immunization studies in IgAN support this notion. Mucosal immunization with neoantigen results in impaired mucosal and systemic IgA responses but normal IgG and IgM responses,<sup>60,61</sup> suggesting that in IgAN there is mucosal hyporesponsiveness to mucosal neoantigens. By contrast, systemic and mucosal immunization with recall antigens results in exaggerated systemic IgA responses with increased and prolonged production of specific IgA.<sup>10,11,62,63</sup> These results suggest that patients with IgAN respond excessively to recall antigens. From these clinical observations we hypothesize that in IgAN there may be impaired elimination of mucosal antigens owing to aberrant local mucosal IgA responses. This results in accumulation of antigen and enhanced antigenic stimulation of B cells with an increase in immunologic memory for IgA1 production in the mucosa and perhaps the BM or other lymphoid tissues. This increase in IgA1-committed memory cells may explain the excessive IgA responses reported after exposure to recall antigens and be responsible for the synthesis of nephritogenic IgA1 in IgAN.<sup>16</sup>

If this hypothesis is correct then there must be continuous antigenic challenge and activation of the IgA immune system driving the synthesis of nephritogenic IgA and the development of IgAN. It is likely that common microbial and food or food-borne antigens play a role in this process. In 1983, Emanipator et al<sup>64</sup> elegantly showed a pathogenic relationship between prolonged mucosal antigenic exposure, the formation of circulating IgA-IC, and the development of glomerulonephritis. The investigators orally immunized Balb/c mice with protein antigens (ovalbumin, bovine gamma-globulins, or ferritin) and found a significant increase in specific-IgA-producing plasma cells in the lamina propria of bronchial and intestinal mucosae coupled with an increase in circulating antigen-specific IgA and mesangial deposits of IgA and J chain. Coppo et

al<sup>65</sup> argued that altered mucosal processing of food antigens such as gliadin, a lectin present in gluten, might be involved in the induction of this disease. High serum levels of IgA antigliadin have been reported in patients with IgAN.<sup>66-68</sup> Coppo et al<sup>69</sup> also showed that mice orally immunized with gliadin or ovalbumin developed glomerular injury with intense glomerular IgA deposition including antigliadin IgA antibodies.

In addition to intrinsic food antigens, food-borne microbial contaminants also may provide an antigenic stimulus in IgAN. Pestka's group and others showed that mice fed meal contaminated with deoxynivalenol developed increased levels of serum IgA, circulating IgA-IC, mesangial IgA deposition, and hematuria, all clinical features of human IgAN.<sup>70-73</sup> Deoxynivalenol (or vomitoxin) is a food-borne mycotoxin that belongs to the trichothecene group of mycotoxins and is known to contaminate agricultural products such as rice, wheat, and corn.<sup>70-72</sup>

Koyama et al<sup>74</sup> described a novel form of glomerulonephritis associated with methicillin-resistant *Staphylococcus aureus* infection and suggested that microbial superantigens may play a key role in its pathogenesis. The abnormalities seen in these patients were similar to those seen in IgAN and included mesangial proliferation with glomerular IgA deposition. In addition, Koyama and his group found similar increases in specific T-cell receptor V $\beta$ + subsets in both patients with post-methicillin-resistant *S aureus* infection glomerulonephritis and IgAN<sup>75</sup> and localization of *S aureus* cell envelope antigen in the glomeruli of IgAN patients.<sup>76</sup> Based on these clinical findings an experimental model was developed in which mice were immunized subcutaneously with *S aureus* antigens. These mice developed a glomerulonephritis very similar to IgAN despite the antigen being injected subcutaneously.<sup>77</sup> Other groups also have shown the development of experimental IgAN after oral immunization with *Haemophilus parainfluenzae* antigens<sup>78</sup> and associated with glomerular deposition of outer membranes of *H parainfluenzae* antigens.<sup>79</sup> High serum levels of IgA antibody against outer membranes of *H parainfluenzae* have been described in patients with IgAN and Henoch-Schönlein nephritis.<sup>80</sup> There

is a high incidence of mesangial IgA deposition in Aleutian disease, a disease of minks associated with persistent parvovirus infection.<sup>81,82</sup> This disease is associated with hypersecretion of immunoglobulins and a plasma cell dyscrasia. Furthermore, Jessen et al<sup>83</sup> experimentally induced murine IgAN by mucosal viral infection with Sendai virus.

Considered together, these studies suggest that exogenous antigens derived from fungi, bacteria, and viruses could play a key role in the development of IgAN. It remains unclear, however, how all of these microbe-related antigens precisely interact with the IgA immune system to trigger disease. Growing evidence from studies of innate immunity may provide a clue. Toll-like receptors (TLRs) are a family of pathogen-recognition molecules that discriminate self from nonself (pathogens) and activate suitable defense mechanisms involving a Th1 immune response.<sup>84</sup> TLRs on antigen-presenting cells also initiate and modulate adaptive immunity during infection.<sup>85</sup> TLR4 binds the gram-negative bacterial cell-wall component lipopolysaccharide and up-regulation at the protein or gene level is associated with development of a lupus-like autoimmune disease in mice.<sup>86</sup> In addition, TLR2 agonists have been shown to exacerbate accelerated nephrotoxic nephritis.<sup>87</sup> TLR9 binds unmethylated CpG dinucleotides (CpG DNA), which are expressed frequently by bacteria and viruses. We recently found increased numbers of IL-5-positive B-1 cells and overexpression of TLR9, mainly on plasmacytoid dendritic cells, in the tonsils of some patients with IgAN (Kano et al, unpublished observations). The same patient group underwent a rapid and successful therapeutic response to combined tonsillectomy and steroid pulse therapy, suggesting that tonsillar TLR9 activation in plasmacytoid dendritic cells may be an important factor in the pathogenesis of IgAN. To investigate this further we maintained our previously described *grouped ddy mice*<sup>47</sup> under conventional conditions and specific pathogen-free conditions. Interestingly, ddY mice reared under conventional conditions developed more marked mesangial IgA deposition and severe glomerular injury, higher serum IgA levels, and enhanced splenic TLR9 expres-

sion. Splenic TLR9 expression correlated with the severity of glomerulonephritis. Nasal challenge with CpG DNA worsened glomerular injury in these mice and was associated with greater mesangial IgA deposition, higher serum IgA levels, and strong Th1 polarization.<sup>88</sup> Our data suggest that TLR9 binding may represent a final common pathway of immune activation for those exogenous antigens (including bacterial, viral, and fungal) that have been shown previously to be involved in the pathogenesis of IgAN. CpG DNA alone may enhance serum levels of IgA/IgG-IC and thereby promote mesangial IgA deposition and glomerular injury. It also is conceivable that TLR activation might trigger modification of the IgA molecule in addition to enhancing production of antigen-specific IgA antibodies.

#### **DETERMINING THE EXTENT OF ABERRANT MUCOSAL PRIMING AND DISRUPTION OF IMMUNE TOLERANCE IN IgAN**

If mucosal priming by common food and microbial antigens is important in driving the development of IgAN then what are the factors responsible for the excessive IgA responses seen in IgAN? These same factors probably underlie the excessive systemic IgA responses seen with recall antigens.<sup>16</sup> The hygiene hypothesis may provide a possible explanation. This hypothesis proposes that early and frequent exposure to bacterial and other antigens, especially at mucosal surfaces, results in the development of a Th1 (cell mediated) immune phenotype.<sup>89</sup> However, with improved public hygiene (as seen in industrialized nations) there is a reduction in antigen exposure with the persistence of a Th2 (antibody-mediated) immune phenotype. This Th2 immune phenotype is associated with an increased risk of developing various allergies. Recent reports have suggested that the prevalence of IgAN correlates with socioeconomic status and the high incidence of IgAN in Western countries could be explained by the hygiene hypothesis.<sup>90,91</sup> The investigators hypothesized that less frequent exposure to infections may lead to a Th2-dominant immune phenotype and increased risk of IgAN. This hypothesis is related closely

**Table 1. IgA immune System Abnormalities in IgAN Identified by Clinical Evidence and Animal Models**

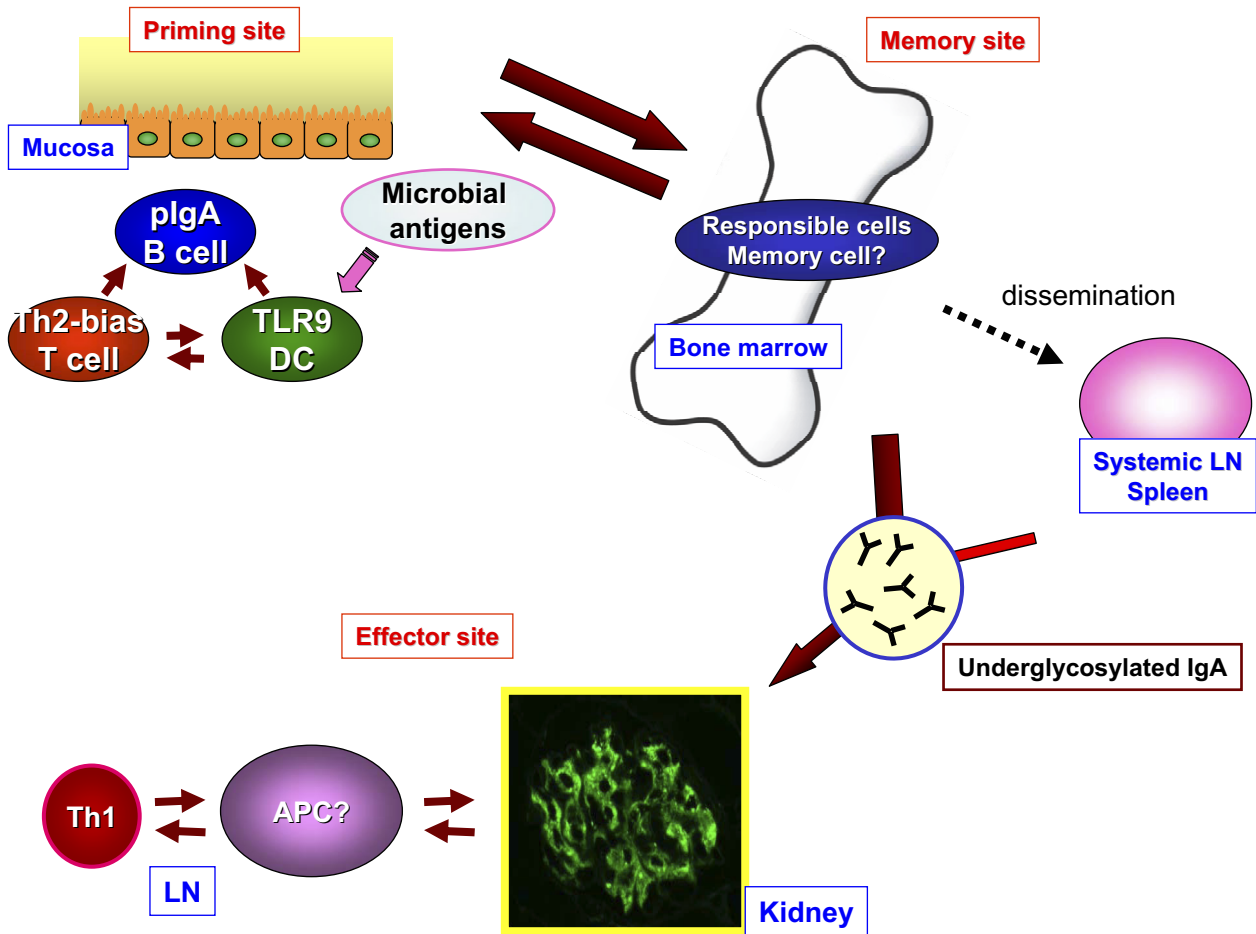
	References	
	Clinical Evidence	Corresponding Animal Models
Characteristics of nephritogenic IgA/IgA-IC		
Affinity for mesangial IgA deposition		20-22 (passive); 23,24 (active)
Endogenous antigens of IgA-IC		
CD89	25	26
Fibronectin	27,30	28
Aberrantly glycosylated IgA	31-33,35	39-41
Production site of nephritogenic IgA		
BM	10-12,42-44	45,49,51
Abnormal mucosal/systemic immune responses to various antigens		
Microbial/microbial-related antigens	10,11,60-63	64
	75,76	77
	79	78
	81,82 (mink)	83
		88
Food/food-borne antigens	65,66,68	69,70-73
Aberrant cellular immunity/immune tolerance	90,91	92,95,96,49,77
Spontaneous IgAN model		37,38,46
	48	47

to immune regulation in mucosal tolerance. Oral tolerance is an important immune mechanism to avoid excessive immune responses to common oral antigens including food antigens and common microorganisms. In 1990, Gesualdo et al<sup>92</sup> provided experimental data suggesting the possible involvement of impaired oral tolerance in the pathogenesis of IgAN. Inhibition of oral tolerance in orally immunized mice by parenteral administration of cyclophosphamide or estradiol given alone or in combination aggravated the nephritis and was associated with enhanced production of systemic IgG and IgM antibodies.

Which factors might be involved in the disruption of mucosal tolerance in IgAN? It is likely that cellular immunity plays a significant role in this process. Signaling through the lymphotoxin and LIGHT (lymphotoxin-like inducible protein that competes with glycoprotein D for binding herpesvirus entry mediator on T cells)

pathways play critical roles in regulating gene expression crucial for innate and adoptive defenses against pathogens, and may contribute to the development of immune tolerance.<sup>93</sup> Transgenic studies have shown that dysregulation of LIGHT signaling results in disturbance of T-cell homeostasis and ultimately the breakdown of peripheral tolerance.<sup>94</sup> LIGHT transgenic mice develop T-cell-mediated intestinal inflammation and profound dysregulation of pIgA production, transportation, and clearance. These changes are accompanied by dominant mesangial IgA deposition and glomerular lesions. This mouse model highlights the direct contribution of T-cell-mediated mucosal immunity to the development of IgAN.<sup>95</sup>

A number of studies have suggested that IgAN is a Th2-biased disease, however, there also is evidence for a Th1 bias in IgAN. Clinical studies to investigate this in human subjects are problematic because of difficulties with the timings of sample



**Figure 1.** Abnormalities in mucosa–bone marrow axis in IgAN as indicated by experimental models. DC, dendritic cell; LN, lymph node; APC, antigen-presenting cell.

collection. In light of this we studied GATA3 transgenic mice. GATA3 is a transcription factor that specifically regulates the Th2 immune response. We crossed GATA3 transgenic mice with mice transgenic for the ovalbumin (OVA)-specific T-cell receptor gene. This double-transgenic mouse then was exposed to repeated mucosal or parenteral OVA challenges. Only repeated mucosal antigen challenge in Th2-biased mice resulted in mesangial deposition of, presumably underglycosylated, IgA and glomerular lesions. Indeed, only mucosally immunized GATA3/OVA T-cell receptor, double-transgenic mice had high serum levels of OVA-specific IgA. Analysis of cytokine expression by splenic cells and Peyer’s patch cells suggested that mesangial IgA deposition was linked to disruption of systemic Th1 tolerance with dysregulation of CD4+CD25+FoxP3+ regula-

tory T cells in the Th2-biased mucosa.<sup>96</sup> These findings suggest that mucosal antigen challenge in a Th2-biased host may induce dysregulation of systemic tolerance, followed by excessive systemic IgA responses, mesangial IgA deposition, and glomerular injury. In this regard, it is intriguing that experimental *S aureus* antigen-associated murine IgAN can be induced in Th2 prone Balb/c mice, but not Th1-prone C57BL/6.<sup>77</sup>

**CONCLUSIONS**

We have discussed how experimental models of IgAN can reflect the clinical features seen in human beings with this disease. The IgA immune system abnormalities identified in IgAN from clinical evidence and corresponding animal models are summarized in Table 1.



As with human IgAN, findings from these experimental models emphasize the pathologic and clinical heterogeneity of this disease. Insights from these animal studies suggest that patients with IgAN have an impaired mucosal immune system with dysregulation of both innate and cellular immunity (Fig. 1). These changes may be linked closely to a disruption of mucosal tolerance. In this setting, repeated exposure to common environmental antigens may induce abnormal priming of cells responsible for the production of nephritogenic IgA production and subsequent dissemination of these cells to sites such as BM. These findings suggest that future treatment strategies should focus on manipulating the priming and dissemination of these memory cells to prevent the appearance of nephritogenic IgA in the systemic compartment.

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