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Sulphilimine cross-links in Goodpasture's disease

R. Vanacore,* V. Pedchenko,* G. Bhave* and B. G. Hudson*†‡
**Center for Matrix Biology and Division of Nephrology & Hypertension, Department of Medicine, and Departments of †Pathology and ‡Biochemistry, Vanderbilt University Medical Center, Nashville, TN, USA*

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Correspondence: R. Vanacore, Division of Nephrology and Hypertension, Department of Medicine, Vanderbilt University Medical Center, S-3221 MCN, 1161 21st Avenue South, Nashville, TN 37232-2372, USA.
E-mail: roberto.vanacore@vanderbilt.edu

Summary

The sulphilimine cross-link of the Goodpasture (GP) autoantigen is a novel molecular mechanism (structural constraint) for conferring immune privilege to a site which otherwise is susceptible to structural changes that induce an immunogenic and pathogenic conformation. Perturbation of the assembly or cleavage of the sulphilimine cross-links could be a key factor in the aetiology of Goodpasture's disease in susceptible individuals.

Keywords: Goodpasture disease, autoantigen, Sulphilimine cross-links

Introduction

Goodpasture's (GP) disease is an autoimmune disease mediated by autoantibodies that attack the glomerular and alveolar basement membranes. GP autoantibodies target the non-collagenous domain (NC1) of the α3 and α5 chain of collagen IV, both of which are part of the α345 NC1 hexamer, the GP autoantigen [1]. A three-dimensional model of the GP autoantigen has been built by homology modelling using the crystal structure of the α121 NC1 hexamer (Fig. 1) [2,3]. The hexamer is formed by the interaction of the trimeric NC1 domains of two adjoining triple-helical collagen IV molecules, and is stabilized by novel sulphilimine bonds that cross-link the interface of the trimeric NC1 domains [3,4]. Understanding the structure of the autoantigen and its epitopes is of pivotal importance in deciphering clues about aetiology of GP disease.

Sulphilimine cross-links and Goodpasture's (GP) disease epitopes

The α345 NC1 hexamer requires dissociation into subunits for epitope formation [1,5,6]. Cross-linked hexamer is inert to binding of GP antibody, but upon dissociation into dimer and monomer subunits by low pH or denaturants, the subunits bind antibodies. In contrast, un-cross-linked hexamer can also be dissociated by GP antibody into monomer subunits that bind the antibody, revealing that sulphilimine cross-links provide a constraint against dissociation (Fig. 2). Hexamer dissociation involves conformational changes that unlock domain-swapping interactions between subunits and exposes residues sequestered by neighbouring subunits [1,7,8]. Concomitantly, hexamer dissociation transforms the non-pathogenic Ea and Eb regions of the α3 and α5 NC1 subunits into pathogenic conformations (epitopes) that confer antibody binding within the dissociated subunits. The sulphilimine cross-links, together with other quaternary interactions, represent a novel molecular mechanism (structural constraints) for conferring immune privilege to sites which otherwise form an immunogenic and pathogenic conformation.

Sulphilimine cross-links and B and T cell epitopes

GP disease is presumed to result from activation of 'ignorant' lymphocytes by an unknown stimulant coupled with epitope presentation and kidney and/or lung susceptibility. B cells are activated upon binding of the B cell receptors (BCR) to the soluble antigen, with assistance from T cells. BCRs presumably do not bind to the native form of the GP autoantigen, the cross-linked hexamer, but bind subunits upon hexamer dissociation into subunits. The un-cross-linked hexamer is more susceptible to dissociation; therefore, the loss of sulphilimine cross-links may be a predisposing factor for binding of BCR to hexamer subunits.

The sulphilimine cross-links may also influence the generation of T cell epitopes. Unlike B cells, T helper cells ($CD4^+$) recognize GP antigens in the form of linear peptides bound to human leucocyte antigen (HLA) class II molecules on the surface of antigen-presenting cells (APC). GP disease

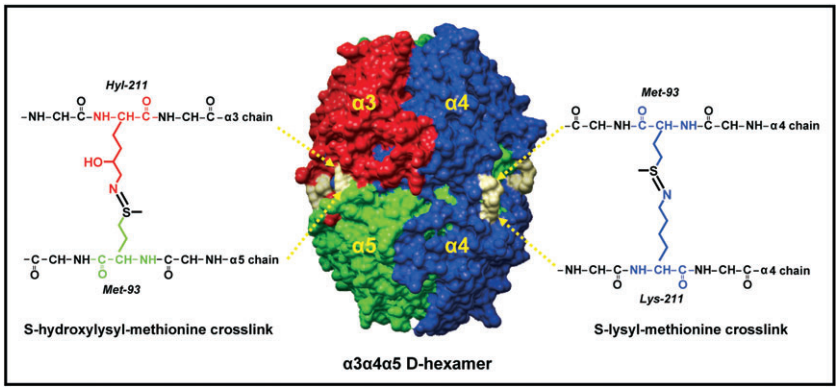

Fig. 1. Sulphilimine bonds reinforce the structure of the α345 non-collagenous domain (NC1) hexamer, the Goodpasture (GP) autoantigen. The illustration shows a space-filling representation of the three-dimensional structure of the α345 NC1 hexamer, the GP autoantigen. It is composed of two trimeric caps, each composed of α3 (red), α4 (blue) and α5 (green) NC1 domains. Residues Met^{93} and Lys^{211} (or its hydroxylated form Hyl^{211}) are connected through a sulphilimine bond, shown in grey, forming two different types of cross-links: s-hydroxyl-methionine (sHM, left) and s-lysyl-methionine (sLM, right).

has strong positive and negative major histocompatibility complex (MHC)-II associations [9]. Previous studies on the processing of GP antigen by feeding human B cells homozygous for HLA-DR15 with recombinant α3NC1 monomer have determined that enzymatic processing, rather than affinity of peptides for HLA molecules, drive GP antigen presentation [10]. The α3NC1 peptides were presented as two core sequences with medium affinity for HLA molecules and their presentation was enhanced by inhibition of the lysosomal protease asparagine endopeptidase.

As a natural mechanism to prevent the activation of T cells, sulphilimine bonds may interfere with the generation of α3NC1 sequences at different stages of proteolytic processing, peptide binding to MHC class II molecules and/or the presentation of peptides–MHC complexes for recognition by $CD4^+$ T cells. However, it is plausible that under pathophysiological conditions that favour the absence of sulphilimine cross-links, antigen processing and presentation may be enhanced, and therefore contribute to the aetiology of GP disease.

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Disclosure

None.

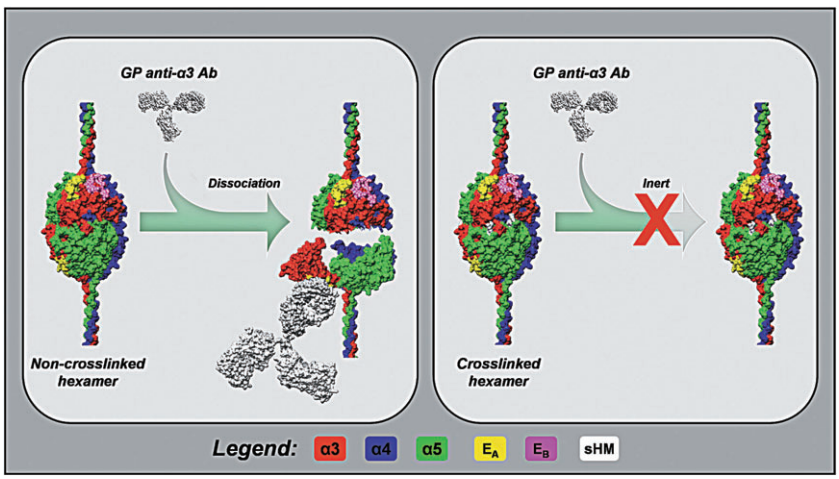

Fig. 2. Sulphilimine cross-links provide a structural constraint against Goodpasture (GP) antibody binding. The α345 non-collagenous domain (NC1) hexamer is shown in the un-cross-linked (left panel) and cross-linked (right panel) forms. Ea and Eb regions are shown in yellow and magenta, respectively, whereas the sulphilimine cross-links are shown in white. The left panel shows an immunoglobulin G (IgG) molecule (white) binding the α3NC1 domain inducing hexamer dissociation and transitioning the Ea region into an epitope. Experimental evidence suggests that hexamer dissociation involves a conformational change within the NC1 domains, which unlocks domain-swapping interactions between subunits. The sulphilimine cross-link provides a constraint against dissociation, rendering the hexamer inert to antibody dissociation and binding.

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