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Invited critical review

# Free immunoglobulin light chain: Its biology and implications in diseases

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ABSTRACT

Immunoglobulin light chain (IgLC) is a component of antibodies, but its free form is observed in the circulation, which originates from 10 to 40% excess synthesis over heavy chain in B cells. Complete antibodies function as a defined tetramer structure unit, $H_2L_2$; thus, separation of heavy and light chains results in considerable or complete loss of antigen-binding ability. Free IgLC has been considered as an inconsequential spillover during antibody assembly because, unlike heavy chain, neither effector functions such as complement activation nor specific-receptor binding has been identified in IgLCs. Free IgLC in sera and cerebrospinal fluids increases in inflammatory diseases such as autoimmune diseases and infections, presumably as a result of B-cell activation. This may be just a concomitant event during elevated disease activity, but recent findings suggest that free IgLC is involved in a wide range of immunological phenomena as a signaling effector or an anti-inflammatory molecule. These effects are likely to be intrinsic to IgLC. In this review, we attempt to give a comprehensive view about the biological roles of free IgLC together with the gene expression, secretion, antigen-binding ability, and its metabolic characteristics.


**Contents**



*Abbreviations:* Ag, Antigen; BCR, B-cell receptor; pre-B cells, Precursor B-cells; CDR, Complementarity determining region; $C_H$, Heavy chain constant domain; $C_L$, Light chain constant domain; ER, Endoplasmic reticulum; FLC, Free immunoglobulin light chain; Ig, Immunoglobulin; κ, Kappa; λ, Lambda; CSF, Cerebrospinal fluid; FLC κ/λ ratio, Ratio of FLC κ to FLC λ concentration; TNP, Trinitrophenol; $V_H$, Heavy chain variable domain; $V_L$, Light chain variable domain.

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## 1. Introduction

Immunoglobulin (Ig), or antibody, is composed of two identical heavy chain polypeptides and two identical light chain polypeptides (approximately 22 kDa). In mammalian immune systems, there are two distinct immunologic types of light chains, κ and λ [1] (Fig. 1). The complete Ig molecule (IgG, IgA, IgM, IgD, and IgE) contains only one type of light chain. The ratio of κ to λ light chains secreted as intact Igs differs among species; for example, the two are almost equal in humans, while κ-type is dominant in mice. The C-terminal half of light chain is the constant region ($C_L$), and the N-terminal half is the variable region ($V_L$), in which the amino acid sequence is quite heterogenic as a result of V–J recombination and somatic mutations in *Igl* genes [2].

Heavy and light chains are asynchronously synthesized on the different ribosomes. Light chain is produced as 10–40% excess over heavy chain in B cells [3–5]. Excess light chains are secreted as the free form into the circulation (Fig. 1). The circulating level of FLC is about 1000-fold lower than that of intact Ig due to rapid clearance in the kidney. In healthy subjects, the free form of the light chain (FLC) κ:λ ratio is about 2:1. The Fc portion of heavy chain fulfills the effector function of Igs, whereas the $C_L$ domain of light chain has no known biological function [6]. Thus, FLC has been generally considered as an inconsequential spillover during antibody production. This view has been changing with the finding that FLC functions as an immunomodulatory or anti-inflammatory molecule.

This review intends to give a comprehensive view of the biology of light chains, especially its free form. We will discuss the biological functions of light chains reported so far, considering their gene expression, secretion, and antigen (Ag)-binding ability. We will also briefly address the association of increased FLC levels with certain diseases and the relevancy. Increased FLC in patients with plasma cell proliferative disorders and its diagnostic usefulness will not be addressed here (for reviews, see Refs. [7,8]).

## 2. Light chain expression

Diversity is essential to ensure that the immune system can recognize non-experienced pathogens. To produce a diverse antibody repertoire, antibody genes are assembled by random recombination of Ig variable (V), diversity (D), and joining (J) gene segments for heavy chain and V and J segments for light chain [2]. Two light chain isotypes, κ and λ, which are encoded by different chromosomes, have been found in all mammals studied [1]. Each locus contains V gene segments that differ in number among species; *e.g.*, there are approximately 140 Vκ and 3 Vλ in mice and 35 Vκ and 30 Vλ in humans.

Mature B cells exhibit only a single class light chain, either κ or λ, *i.e.*, isotype exclusion. Rearrangement of the κ locus precedes that of the λ locus. If the κ locus arrangement is productive, rearrangement of the λ locus will be inhibited; otherwise, λ locus rearrangement will be achieved. Although DNA recombination in the κ or λ light chain locus occurs in a similar manner as in the Ig heavy chain locus [9], the regulatory mechanism of Ig light chain isotype exclusion is not well defined yet [10]. Antibody selection is initiated upon light chain gene expression in early immature B cells [11].

## 3. Production and secretion of Ig light chain and free Ig light chain

### 3.1. Pre-B cell receptor assembly

*Igh* gene rearrangements occur before *Igl* gene rearrangements. Rearranged μ-heavy chains undergo the "quality control" criteria test by pairing with the invariant Ig-like surrogate light chain. The surrogate

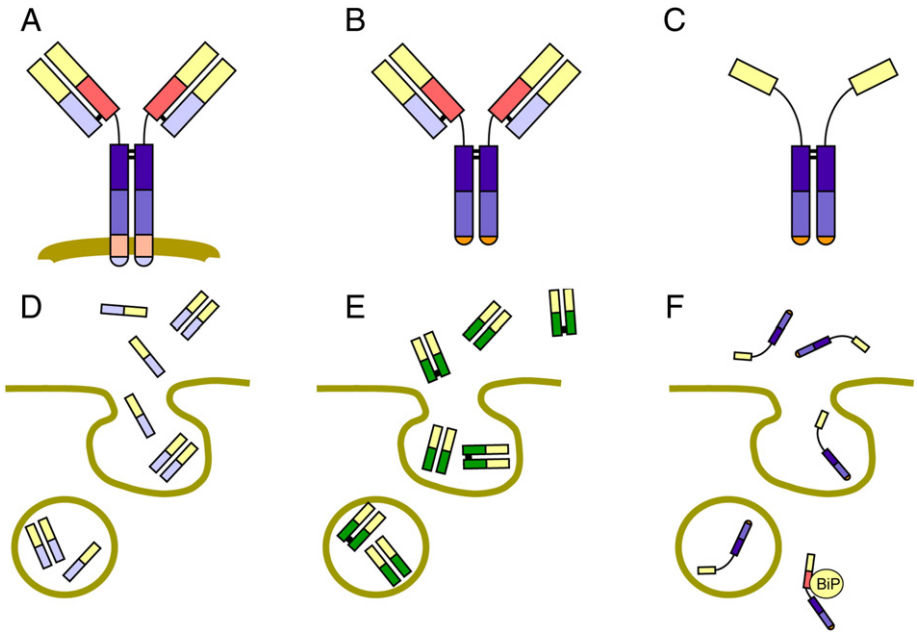


**Fig. 1.** Structure and secretory-competency of immunoglobulin molecules. A, Membrane-bound immunoglobulin; B, Secretory form of immunoglobulin. No membrane binding domain; C, Heavy-chain antibody in camels or mice lacking light chain gene. $C_H1$ domain is lacking; D, FLC λ secretion as monomer or non-covalent dimer; E, FLC λ secretion as non-covalent or covalent dimer. The covalent dimerizations are shown as black junctions; F, Secretion of heavy chain lacking $C_H1$ domain, observed in patients with heavy chain diseases. Heavy chains with $C_H1$ domain undergo proteasomal degradation by binding BiP. $C_H1$ domains in red; $V_H$ and $V_L$ domains in yellow; $C_L$κ in light blue; $C_L$λ in green.

light chain composed of VpreB and λ5 [12] is selectively expressed in progenitor and precursor B (pre-B) cells [13]. This protein associates with a μ-heavy chain to form an Ig-like heterodimer (pre-B cell receptor; pre-BCR). Successful μ-heavy chain production is confirmed by movement of the complex to the surface of the cells; otherwise, the pre-B cells die. Once pre-BCR is expressed, pre-B cells undergo further maturation and differentiation, and thereby the surrogate light chain expression is turned off [13]. Pre-B cells then stop proliferating and begin to rearrange the light chain loci.

### 3.2. Light chain production and B-cell receptor assembly

Heavy chain binding protein, BiP, is a chaperon that is homologous to the 70 kDa heat-shock protein and strongly binds to the $C_H1$ domain to hold heavy chains until the heavy chain is matched up with light chains [14]. $C_L$ domain, which is the cognate partner of the $C_H1$ domain in the complete Ig molecule, displaces BiP and stabilizes the $C_H1$ domain [14]. Unpaired heavy chains are retained in the endoplasmic reticulum (ER) and eventually degraded by the proteasomes [15]. After successful *Igl* rearrangement, heavy and light chains are assembled into a defined $H_2L_2$ quaternary structure. The completely assembled antibody is transferred to the plasma membrane to form B-cell receptor, and then the B cells are subjected to selection by self-antigen presentation in the environment of bone marrow and the periphery [13].

### 3.3. Secretion of free Ig light chains

Heavy and light chains are synthesized asynchronously and assembled in the ER to form complete Ig in B cells at any stage of development beyond pre-B cells [16,17]. Because of the excess production of light chains over heavy chains and their secretion competency, excess light chain is secreted as a free form [3–5]. Degradation of normal Igs does not supply FLC in the circulation [18].

The excess production of light chain over heavy chain maintains a constant intracellular pool of light chains [19], which is located in the perinuclear space [20]. This pool of light chains mediates the release of the relatively insoluble heavy chains from their ribosomes and hampers the formation of toxic heavy chain aggregates [21,22].

FLCs exist in the form of monomers, non-covalent, or covalent dimers in biological fluids: FLC κ consists of nearly equal amounts of monomer and noncovalent dimer; FLC λ is present as covalent dimers [23–25] (Fig. 1).

### 3.4. Clearance and catabolism of free Ig light chains

The kidney is the major site of FLC catabolism. Secreted FLCs readily pass through the glomerular filtration barrier with a serum half-life of 2–6 h [26], whereas the serum half-lives of intact Igs are 20–25 d for IgG, 6 d for IgA, 3 d for IgD, 2 d for IgE, and 5 d for IgM [27]. Most FLCs filtered are rapidly catabolized or reabsorbed by proximal renal tubular cells [4,28,29]. Like other microproteins, FLCs are readily endocytosed through the brush-border membranes and degraded into oligopeptides and amino acid residues inside the lysosomes. In that process, megalin, a 600 kDa glycoprotein receptor, and cubilin, a peripheral membrane protein anchored to megalin, are the major mediators of light chain entry [30–32]. The kidney can absorb 10–30 g of FLC per day [4]; therefore, only small amounts of FLCs are observed in serum and urine under normal conditions [33,34].

In healthy subjects, the serum reference value ranges from 1.2 to 43.5 mg/L for FLC κ and from 3.8 to 55.2 mg/L for FLC λ, depending on the assay method used [35–37]. The mean serum FLC κ/λ ratio is around 0.5 to 12.5. The concentrations of FLC are approximately 1000-fold lower than those of intact Igs [38].

## 4. Light chain in Ig antigen-binding

### 4.1. Contribution of light chain to antigen–antibody interaction

The structural integrity of Ig, including the constant regions, is necessary for Ag specificity, affinity, and plasticity [39]. As such, light chains separated from Ag-specific polyclonal antibodies display little or no Ag-binding activity [40,41]. On the other hand, although separated heavy chains considerably lose Ag-binding ability, the Ag-binding ability of heavy chain has been recovered by refolding with unrelated light chain with affinities approaching the original antibodies [40,42]. Light chain reconstituted with nonspecific heavy chain did not recover Ag-binding ability [43]. These observations have suggested that heavy chain plays a rather dominant role in Ag binding. Further, antibodies without any light chain are found in camels and sharks, implying that light chains may not be necessary for effective Ag binding (Fig. 1). Heavy chain antibodies in camels function at least as well as conventional antibodies [44,45]. In addition, it is possible to obtain the desired $V_H$ domains that possess specific Ag-binding activity from diverse libraries of $V_H$ genes [46].

### 4.2. Autoantibodies

Light chain appears to play a relatively important role in modulating antibody specificity [47–49]. The majority (55 to 75%) of all antibodies expressed by early immature B cells display self-reactivity, including polyreactive and anti-nuclear specificities [50]. Such naturally-occurring autoantibodies typically have a positively charged long heavy chain CDR3 with a pattern favoring self-reactivity and less specificity [51–53]. B cells with self-reactivity silence the self-reactivity of the antibodies by receptor editing, *i.e.*, light chain exchange [50,54,55]. Thus, light chain can shape or skew the unfavorable antibody reactivity, and thereby, convert dangerous self-reactive antibodies into innocuous antibodies.

## 5. Biological function of free Ig light chains

### 5.1. FLC as a mediator of hypersensitivity-like responses in allergy

Activation of mast cells through various pathways is the central mechanism in allergic disorders. IgE is the major mediator of the activation by crosslinking Ags to FcεRI on mast cells [56]; however, a considerable number of patients (*e.g.*, about 40% of adult asthmatics and individuals allergic to cow's milk [57,58]) display allergic symptoms without increased and/or Ag-specific IgE.

In 2002, Redegeld et al. [59] revealed that light chains separated from monoclonal antibodies specific for trinitrophenol (TNP) or oxazolone mediate a mast-cell-dependent hypersensitivity-like response in mice challenged with the respective Ags. They excluded the involvement of endogenous Igs or Fc receptors, such as FcεRI and FcγRIII, in FLC-mediated reactions. They suggested that the reaction was elicited by FLC, because F991, a 9-mer polypeptide derived from the light chain binding domain in Tamm-Horsfall glycoprotein, specifically inhibited the FLC-mediated allergic response. However, Tamm-Horsfall glycoprotein binds to the CDR3 region of light chain [60,61], which is exposed on the surface of the antibody in both free and bound light chain forms [62]. Thus, F991 is likely to bind not only FLC but also Igs. The specificity of F991 to the 'free' form of light chain has not been confirmed at the molecular level. The phenomenon by light chain is expected to explain non-IgE allergic responses as described above [63–66], although it is still unknown how FLCs activate mast cells to elicit allergic responses. Possible light chain receptors have been implicated [59], but not yet found.

Earlier works on light chains separated from Ag-specific Igs [67] showed that they had little or no Ag-binding ability, raising a question about the Ag-binding ability and specificity of endogenous FLC.

**Table 1**
Clinical relevancy to the increases of free immunoglobulin light chains and abnormal FLC κ/λ ratio in biological fluids.

| Diseases | Findings | Major results/conclusions | Year, (Ref no.) |
|---|---|---|---|
| Autoimmune diseases | | | |
| SLE | Serial urinary FLC measurement serves as a valuable guide to disease management | Urinary FLC increase preceded the onset of active disease conditions, while successful treatment was associated with decreasing urinary FLC level | 1974, [85] |
| | Increased, decreased FLCs, and/or abnormal FLC κ/λ ratio in urine of patients with SLE | Abnormal FLC levels and/or FLC κ/λ ratio, 12/18 (67%) | 1998, [86] |
| | Urinary FLC increase might be a marker of B-cell stimulation | A significant rise in urinary FLCs was associated with a subsequent clinical relapse of disease | 1989, [87] |
| | Urinary FLC levels were increased in SLE patients in active state | Mean value of serial FLC measurements was 2.7–7.2-fold greater in active than stable states (5/5) | 2000, [88] |
| RA and SLE | Increased FLCs in the sera of patients with SLE and in the synovial fluid in patients with RA | Increased in the sera of 38/40 (95%) with SLE and in the synovial fluid of 12/13 (92%) with RA | 1966, [89] |
| | A significant rise in urinary FLCs was always associated with a subsequent clinical relapse of the disease. | $\beta_2$-microglobulin and lysozyme did not increase during monitoring, indicating that the FLC increase was not due to tubular dysfunction | 1989, [87] |
| RA and primary Sjögren's syndrome | Serum FLC levels are increased and correlated with disease activity in RA and primary Sjögren's syndrome | Serum mean FLC levels of patients with RA (n50) and primary Sjögren's syndrome (n139) were approximately 1.5–2-fold greater than those in controls (n80) ($p<0.001$–$0.0001$) | 2006, [90] |
| Multiple sclerosis (MS) | CSF FLC κ level was a specific marker for MS | Increased CSF FLC κ in 33/39 (84%) MS; 1.4 ± 1.21 mg/L (Mean ± SD), but no detectable CSF FLC κ in all (except one) patients with inflammatory and infectious diseases | 1986, [91] |
| | | FLC κ was concentrated 71–120-fold in the CSF of patients with MS compared to reference proteins | 1986, [91] |
| | CSF FLC κ was increased in MS and CSF FLC λ was increased in infections | Higher levels of CSF FLC κ, FLC κ/λ ratio, FLC κ/Alb were characteristics of MS; Levels of FLC κ and FLC λ were correlated with IgG level in MS ($r=0.79$) and infections ($r=0.81$), respectively | 1987, [92] |
| | FLC κ and FLC λ indices [(CSF FLC/serum FLC):(CSF Alb/serum Alb)] distinguish MS from infections in the CNS | Increased FLC κ and FLC λ indices in 86% (123/143) and 85% (86/101), respectively, with MS, 40% (30/75) and 23% (15/64), respectively, with infections | 1988, [93] |
| | FLC κ was more accurate for the diagnosis of MS than other clinical parameters | Areas under curves with FLC κ, CSF IgG, and IgG index were 0.91, 0.49, 0.71, respectively, by the ROC curve analysis | 1989, [94] |
| | Increased CSF FLCs in early MS patients | 19/33 (58%) patients with possible and probable MS had increased FLC levels | 1990, [95] |
| | Ratio of urinary FLC κ to creatinine was useful for monitoring disease activity | Increased of FLC κ/creatinine corresponded to worsening of disease. | 1991, [96] |
| | CSF FLC κ level predicted subsequent physical deterioration | Patients with CSF κ levels in the upper quartile had a higher risk of progression | 1995, [97] |
| | FLC κ in CSF may predict disease progression to MS in patients with isolated optic neuritis | Only elevated FLC κ in CSF observed in 5 of 8 patients was correlated with disseminated disease among the clinical tests tested | 1986, [98] |
| Heart failure | FLCs are increased in the sera of patients with heart failure | The mean serum FLC κ and FLC λ concentrations of the patients were 1.47-fold and 2.14-fold, respectively, those of the controls | 2005, [97] |
| | | FLC κ and FLC λ concentrations were correlated with serum NT-proBNP ($r=0.71$ with FLC κ, $r=0.90$ with FLC λ) | 2005, [99] |
| | NT-proBNP decreased corresponding to decreased FLC in serum of patients with cardiac dysfunction in AL amyloidosis (n52) | Reduced concentration of circulating amyloidogenic precursor (FLC) may be associated with improvement of cardiac dysfunction | 2006, [100] |
| Diabetes mellitus | Greater excretion of FLC κ into urine in IDDM patients with normal albumin excretion | FLC κ/albumin × 10 (Mean ± SEM): 10.5 ± 4.0 in IDDM patients with proliferative retinopathy; 32.4 ± 7.9 in IDDM patients without proliferative retinopathy; 1.1 ± 0.3 in non-IDDM patients with proteinuria | 1985, [101] |
| | Duration of IDDM is associated with excretion of FLC κ | mg/GFR (Mean ± SEM): 44 ± 5 in long-standing IDDM; 6 ± 1 in newly diagnosed IDDM; 2 ± 1 in controls | 1990, [102] |
| | Higher urinary FLC κ levels in patients with NIDDM | mg/GFR; 7 ± 2 in NIDDM, 2 ± 1 in controls (Mean ± SEM) | 1990, [102] |
| Renal failure | | | |
| Renal insufficiency | The concentration of FLCs was inversely correlated with glomerular filtration rate | FLC κ vs. GFR, $r=-0.88$, $p<0.001$; FLC λ vs. GFR, $r=-0.85$, $p<0.001$ (n29) | 1976, [103] |
| Uremia | Abnormal FLC κ/λ ratio in serum and inconsistency of FLC κ/λ ratio with total light chain κ/λ ratio | Lower FLC κ/λ ratio in 6/8 with uremia than those in controls | 1974, [104] |
| Chronic renal failure and nephropathy | Increased FLC λ and decreased FLC κ/λ ratios in urine | Both FLCs increased in the urine of 13/31 (42%) patients and increased FLC λ in 5/31 (16%) | 1998, [86] |
| Proteinuria | Both increased FLC κ and λ in urine (polyclonal increase) | 45(3.8–211, range) mg/24 h of FLC κ; FLC κ/λ ratio, 1.27 (0.23–3.4, range) | 1982, [105] |
| Hemodialysis | Increased FLC λ in patients with chronic hemodialysis | Serum FLC κ and FLC λ in patients on hemodialysis were increased 4.0-fold and 1.8-fold, respectively, over the controls | 1991, [73] |
| Inflammation in CNS | The serum polyclonal FLC levels were approximately double in patients with chronic active sarcoidosis | Serum FLC κ and FLC λ in the patients were increased 2.4-fold and 1.7-fold, respectively, over the controls | 1981, [106] |
| | Intrathecal immunoglobulin synthesis was identified by FLC κ and FLC κ CSF/serum | The areas under the ROC curves were 0.991 for FLC κ only and 0.978 for FLC κ CSF/serum | 2004, [107] |

**Table 1** (*continued*)

| Diseases | Findings | Major results/conclusions | Year, (Ref no.) |
|---|---|---|---|
| HIV infection | Increased FLC levels in the CSF and sera of patients with HIV-1 infection | CSF and serum FLC levels in patients with HIV-1 infection were approximately 10-fold and 3 to 5-fold, respectively, greater than those in controls; These levels were also slight and approximately 2-fold greater than those in patients with MS | 1991, [108] |
| | The presence of CSF FLCs, especially λ-type FLC, without oligoclonal IgG or increased intrathecal IgG synthesis | CSF FLCs with restricted heterogeneity in the absence of oligoclonal IgG (14/18) and normal intrathecal IgG synthesis (8/11) | 1990, [109] |

Alb, albumin; CSF, cerebrospinal fluid; FLC, free immunoglobulin light chain; GFR, glomerular filtration ratio; HIV, human immunodeficiency virus; IDDM, insulin-dependent diabetes mellitus; NIDDM, non-insulin-dependent diabetes mellitus; NT-proBNP, N-terminal natriuretic peptide type B (NT-proBNP); RA, rheumatoid arthritis; ROC, receiver operating characteristic; SLE, systemic lupus erythematosus; MS, multiple sclerosis.

Redegeld et al. [59] detected a light chain portion in the sera of immunized mice that binds to the hapten-conjugated column, whereas we did not detect Ag-binding activity of purified FLC fractions in sera from patients with allergy [68]. If FLCs have Ag-binding ability comparable to the parent antibodies, such FLC-antibodies may be nonspecific and thus could be self-reactive or induce unnecessary immunological responses (see Section 4). It is possible that antigen specificity is not necessary in light chain-mediated hypersensitivity-like responses, as observed in non-specific IgE-mediated immune sensitization [69].

### 5.2. Other possible immunological roles of FLC

Neutrophils are the main cells of the first line of the non-specific immune defense system during bacterial infections, which are still the main cause of increased morbidity and mortality among uremic patients [70], mainly as a result of the altered functions of neutrophils [71]. Mechanisms of altered neutrophil function in uremia include malnutrition, iron overload, increased intracellular calcium, dialysis treatment, and circulating plasma proteins [70]. Neutrophils isolated from uremic patients have disrupted carbohydrate metabolism and reduced chemotactic activity, thereby demonstrating disturbed intracellular killing ability.

In patients suffering severely impaired kidney function, serum FLC was increased about 5-fold [72]. Interestingly, Wakasugi et al. [73] observed an increase of FLC κ (about 2-fold) and FLC λ (about 4-fold) in patients receiving hemodialysis. Cohen et al. [74,75] revealed that FLCs irreversibly inhibited the chemotactic movement of neutrophils and glucose uptake by the cells. Further, in multiple myeloma, in which patients often display light chain monoclonal gammopathy, an increased risk of bacterial infections as a result of decreased neutrophil functions has been indicated [76]. This suggests that the modulatory effect of light chain on neutrophils is associated with a bacterial infection-prone feature of uremia and multiple myeloma.

FLC also attenuated the coordinated apoptotic cell death of neutrophils, which was abolished by light chain antibodies [71]. This may be associated with the chronic inflammatory status observed in end-stage renal disease patients. Hutchison et al. [77,78] found that removal of FLC by hemodialysis increased the rate of renal recovery in multiple myeloma patients with severe renal failure. Matsumori et al. [79] demonstrated that FLC greatly improved survival in viral myocarditis, partly via the anti-inflammatory effect. This suggests that FLC is involved in a wide range of immunological regulatory systems. FLC could exert this effect directly, or the hypothetical idiotypic network of circulating FLC might be involved in such modulatory effects [80,81].

Wall et al. [82] showed that light chain binds not only B-cells but also synthetic phospholipid membranes. Moreover, it has been demonstrated that light chain binds mast cells. The direct inhibition of viral replication by nonspecific light chains [82] indicates the interaction of light chains with human amnion cells used as the host, or the virus particles. These findings suggest that light chains interact with plasma membranes to induce stimulatory effects on the cells. In fact, the cell surface is positively charged and Igs, as well as light chains, are anionic. Further, light chains have been shown to stimulate tyrosine phosphorylation [83], enhance tumor forming factor-β secretion of mesangial cells [84], and increase interleukin-10 secretion in the heart [79].

## 6. Increased FLC and diseases

### 6.1. FLC in diseases

Increases of FLC κ, FLC λ, or both have been observed in various diseases (Table 1; FLC increases in plasma cell proliferative disorders are not included). Urinary FLC levels increase when tubular reabsorption of FLC is impaired and/or renal FLC filtration overwhelms the kidney's ability to catabolize [85–88,101–105]. Serum FLC levels increase when FLC production is elevated, presumably as a result of B-cell activation in inflammatory diseases [99,106,107], such as autoimmune diseases [87,90,96] and infections [93,108,109].

### 6.2. Multiple sclerosis

Intrathecal Ig light chain synthesis is commonly observed in inflammatory disorders, including multiple sclerosis [91–95,97,98,107]. Several studies have shown the increase of FLC κ, but not FLC λ, in the cerebrospinal fluid (CSF) and urine of patients with multiple sclerosis, suggesting its clinical usefulness for measurement [91,94]. On the other hand, isoelectric focusing analysis to identify multiple bands (oligoclonal pattern of FLC) in CSF is more sensitive to detect Ig synthesis within the brain and is more informative than FLC quantification [110]. Thus, detection of the “finger pattern” by isoelectric focusing analysis is recommended as a diagnostic method for multiple sclerosis [111]. The advantage of quantitative assay is the ability to monitor the therapeutic efficacy and predict prognosis [97].

### 6.3. Diabetes mellitus

Microalbuminuria is the primary clinical marker of early glomerular damage in patients with diabetes mellitus; however, several tubular proteins and enzymes can be detected before the onset of microalbuminuria, indicating that tubular dysfunction precedes glomerular damage [112]. Teppo and Groop [101] showed increased excretion of FLC κ in the urine of patients with diabetes but not with nondiabetic proteinuria, suggesting the specific increase of FLC κ in diabetic patients. They also found increased FLC κ in the early stage of diabetes when no abnormality was detected by measuring urinary albumin and $\beta_2$-microglobulin, a tubular damage marker [102]. Thus, urinary FLC κ can be used as a marker of early diabetic nephropathy.

## 7. Conclusions

Light chain contributes to form Ag–antibody complex by composing the binding site. Separated light chains do not have comparable Ag-binding ability. It is likely that light chain is initially required to confer Igs secretory competency and allow greater antibody diversity. Excess light chains spill over as FLCs into the circulation. In the initial

development of the antibody system, spilled light chains might be just inconsequential remnants.

However, such spillovers can be used as signaling molecules among the cells. For example, degraded peptide fragments of hormone precursors, such as pro-opiomelanocortin [113], proglucagon [114], and the linker of insulin, C-peptide [115], function as signaling molecules. Moreover, ATP filling synapse as a counterpart anion against positively charged acetylcholine is used as a neurotransmitter. ATP spilled over from apoptotic cells functions as a "find me" signal so that dying cells are removed [116]. Moreover, living cells leak trace ATP that probably forms microclimate $[ATP]_e$ in the peripheral spaces, which may be involved in establishing the basal level of activation [117]. It is possible to think that such fragments were initially remnants and then receptors for them developed later, implying that FLC is utilized as a signaling molecule after emergence into the circulation. Moreover, B cells can enhance FLC secretion irrespective of their antibody production rate [5], indicating the presence of an independent regulatory system for light chain production from the antibody production rate. To further elucidate the biological roles of FLC, identification of the responsible mechanism or receptors is necessary.

It is still unknown how FLC mediates its biological functions. The existence of receptors for FLCs is suggested. Another possibility is the binding of FLC to the cell surface or the endocytosis of light chains. Data from separate light chain analysis suggest that the requirement of antigen-binding ability for FLCs is unlikely [69]. Indeed, in anti-inflammatory or modulating effects of neutrophil functions by FLC, antigenic specificity and binding ability are unnecessary. The usually hidden region of $C_L$ domain in intact Ig might function in these effects.

The circulating level of FLC increases in association with disease activity or when the clearance is disturbed. In the former, light chain may play a role in the pathogenesis of the disease while, in the latter, increased FLC could induce unnecessary reactions, such as toxicity to the kidney. Whichever mechanism is used, the measurement of FLC in biological fluids may provide a useful parameter to understand the role of FLC in diseases and to monitor disease progression for diagnostic purposes.

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