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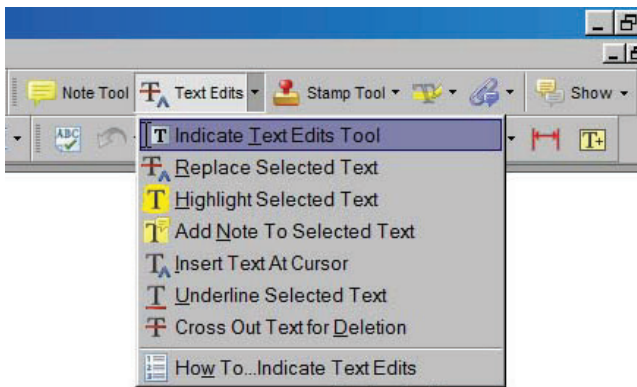


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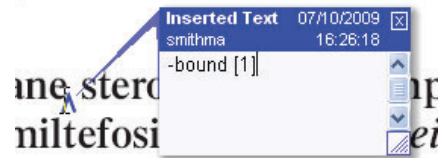


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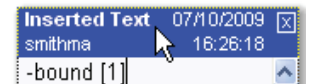
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Full Review

Interleukin-6 in renal disease and therapy

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ABSTRACT

Interleukin (IL)-6 has become a major target for clinical intervention in various autoimmune conditions. Here, drugs including the humanized anti-IL-6 receptor (IL-6R) antibody Tocilizumab emphasize the clinical importance of IL-6 in driving disease and poor patient outcomes. During the course of this review we will outline the biology surrounding IL-6 and will discuss the impact of IL-6 in renal disease and the clinical complications associated with renal replacement therapies and transplantation. We will also consider the merit of IL-6 measurement as a prognostic indicator and provide a clinical perspective on IL-6 blocking therapies in renal disease.

Keywords: cytokines, interleukin-6, renal disease, therapy

INTRODUCTION

Cytokines perform pivotal roles during infection, trauma, cancer and inflammation where they control cellular proliferation, differentiation, survival or death and cytokine specific gene expression. Here, cytokine-driven communication between immune cells and stromal non-haematopoietic cells enables resolution of the condition and is part of the healing process [1]. However during chronic inflammatory conditions, appropriate regulation of the immune response is lost and drives disease progression. Under these circumstances, cytokines affect the development of autoimmunity, chronic inflammation and deleterious tissue damage [1]. This has ultimately led to the design of biologic drug agents that target specific cytokines to prevent the rapid clinical progression of disease. For example, tumour necrosis factor- α (TNF α) blockers (e.g. the neutralizing anti-TNF α antibodies Infliximab, Adalimumab, Golimumab, Certolizumab or the soluble TNF-R2 Fc-fusion protein

Etanercept) are broadly used to treat various autoimmune conditions, while interleukin (IL)-1 β inhibitors (e.g. the IL-1 receptor antagonist, Anakinra) have shown robust efficacy in auto-inflammatory conditions [2]. Although these agents are effective treatments for many diseases, not all biologics work in all patients, and not all biologics work in all inflammatory conditions [1, 2]. Such observations are significantly influencing the way researchers consider cytokine involvement in disease. Emphasis is now placed on identifying alternative cytokine targets and strategies for therapeutic intervention, while a greater attention to cytokine biology and signalling is providing opportunities to stratify patients with chronic disease for more appropriate treatment. Here, research is leading to a detailed understanding of how the cytokine network becomes distorted to drive chronic inflammation instead of competent host defense. During the course of this review, we discuss the impact of IL-6 in renal disease and will describe aspects of its biology that affect disease onset and progression, prognosis and treatment decisions.

INTERLEUKIN-6 AS A CLINICAL TARGET

Interleukin-6 was first described as interferon β 2, hepatocyte stimulating factor, cytotoxic T-cell differentiation factor, B-cell differentiation factor and B-cell stimulatory factor-2, which reflects its capacity to regulate lymphocyte activation and the acute phase response [3]. While these activities are markedly impaired in IL-6-deficient mice, it is important to remember that IL-6 also controls various homeostatic functions including glucose metabolism, the hypothalamic-pituitary-adrenal (HPA) axis, affecting mood, fatigue and depression and haematopoiesis [3,4]. In this regard, systemic elevations in IL-6 cause hyperthermia and leads to a general loss of activity and appetite [5]. As an inflammatory cytokine, IL-6 is one of the

most highly regulated mediators of inflammation (increasing from 1–5 pg/mL to several µg/mL in certain conditions) and performs central roles in infection, autoimmunity and cancer [6–9]. While traditionally viewed as a downstream target of TNFα and IL-1β activity, various other inflammatory stimuli induce IL-6 expression, and IL-6 forms part of an integrated cytokine network that controls innate and adaptive immunity [3, 8, 9]. As a consequence, IL-6 is a major target for therapeutic intervention and the complex nature of its biology has led to development of various therapies that target either the cytokine directly (e.g. Olokizumab, Clazakizumab) or the α-subunit of its receptor (e.g. Tocilizumab, Sarilumab) [9]. A full list of blocking IL-6 strategies and the current status of their clinical development is shown in Table 1. This information covers specific IL-6 targeting agents and small molecule inhibitors that block intracellular proteins associated with (but not necessary exclusive to) IL-6 receptor signalling. Given that cytokines, such as IL-6 contribute to the progression of renal disease and associated complications (e.g. vascular calcification, wasting, fatigue and cardiovascular risk), the potential applications of anti-cytokine targeted intervention deserves closer attention [16, 17]. For additional information on IL-6 targeted therapies the reader is directed elsewhere [2, 9, 18].

The IL-6 receptor complex consists of an 80 kDa cognate receptor (IL-6R, CD126) and a 130 kDa signal-transducing element (gp130, CD130) [8, 9]. Although IL-6R is largely confined to hepatocytes, certain leukocytes and some epithelial lining cells, IL-6 activity is also controlled by a naturally occurring soluble IL-6R (sIL-6R). The sIL-6R is a key regulator of IL-6 responses and forms a sIL-6R/IL-6 complex capable of activating cells via the ubiquitously expressed gp130 [3, 7–9, 19]. This process is called IL-6 trans-signalling, and activates IL-6-type responses in cells lacking IL-6R (e.g. vascular endothelial cells, peritoneal mesothelial cells and synovial fibroblasts). Interleukin-6 responses *in vivo* are therefore mediated by IL-6 activation of a membrane-bound IL-6R (classical IL-6R signalling) or via its soluble receptor (Figure 1). In both cases, IL-6 activates gp130 associated cytoplasmic tyrosine kinases (Janus kinases; Jak1, Jak2 and Tyk2), which control the latent transcription factors STAT1 and STAT3, and signalling through the Ras–Raf cascade. For a more detailed overview of IL-6 signalling the reader is directed elsewhere [20].

From a clinical perspective, measurements of IL-6, its soluble receptor and indices of IL-6 bioactivity (e.g. STAT3 responses) are increasingly viewed as surrogate markers of inflammation, disease severity and valuable predictors of disease progression. During the course of this review we will consider the impact of IL-6 biology in various aspects of renal disease, transplantation and therapy and detail potential avenues for future clinical translation (Table 1).

CONTROL OF IL-6 EXPRESSION BY INFLAMMATION, GENETIC VARIATION AND MICRORNAS

In chronic inflammation and autoimmunity IL-6 plays roles in both local (e.g. control of chemokine-directed leukocyte

recruitment) and systemic inflammation (e.g. activation of the acute phase response). Here, IL-6 transcription is regulated by various pro-inflammatory cytokines and growth factors (e.g. IL-1, TNFα and platelet derived growth factor; PDGF), increases in intracellular cyclic AMP and certain pattern recognition receptors following activation by microbial or endogenous ligands [1]. Interleukin-6 is rapidly expressed in a highly transient manner during inflammation. Here, circulating sIL-6R concentrations act as a buffering system that helps to maintain the circulating half-life of IL-6. Thus, sIL-6R is a central regulator of IL-6 bioactivity and must be tightly controlled to limit overt IL-6 signalling (Figure 1). While circulating levels of soluble gp130 (sgp130) selectively antagonize IL-6 trans-signalling, intracellular regulators of IL-6R-gp130 signalling (e.g. suppressor of cytokine signalling; SOCS proteins) prevent a prolonged activation by IL-6. Of equal importance are genetic factors, which substantially influence *IL6* expression. This is best illustrated by the identification of functional polymorphisms within the *IL6* promoter. For example, a G > C (rs1800795) mutation found at position –174 bp upstream from the transcriptional start site leads to enhanced IL-6 expression [21]. Similar genetic variations up to 6 kb from the start of transcription also correlate with serum and constitutive IL-6 levels. Patients displaying the rs1800975 mutation often show increased susceptibility to coronary artery disease, juvenile idiopathic arthritis and many other conditions typified by chronic inflammation or autoimmunity. Similar genetic mutations also occur in the *IL6R* loci, with the rs2228145 variant affecting the proteolytic release of sIL-6R from cells. Significantly, the rs2228145 mutation in various ethnic populations has been linked to insulin resistance, an increase in body mass index, Type-II diabetes and diabetic nephropathy [22–28].

Recent data show that microRNAs also govern IL-6 expression and activity. MicroRNAs are short endogenous RNA regulators of gene expression, the first of which was identified in the nematode *Caenorhabditis elegans* in 1993 [29]. MicroRNAs are present in all human cells, and each represses the expression of a specific set of genes. The let-7 family contains the first microRNAs to be identified in mammals [30]. Let-7 microRNAs, most notably let-7a, target and repress synthesis of IL-6 [31]. Let-7 microRNAs are themselves down-regulated by Lin28B [31], which is induced following activation of nuclear factor-κB (NF-κB) to ensure optimal *Il6* expression and cellular transformation [32]. Also, microRNA-23a targets and represses *Il6r* [33], meaning that cellular capacity to respond to IL-6 is also microRNA-regulated. MicroRNAs are also important effectors of IL-6 responses. For example, microRNAs-21 and microRNAs-181b-1 are induced by IL-6 activation of Stat3. These increase NF-κB activity via repression of their targets phosphatase and tensin homologue (PTEN) and CYLD (the human gene associated with cylindromatosis) [34]. Finally, emerging data show that microRNAs may be released by cells, and serve as a mechanism for intercellular communication. A subset of these extracellular microRNAs can induce pro-inflammatory responses, including IL-6 secretion, by binding to and activating Toll-Like Receptors [35].

Table 1. Targeting IL-6 in disease

Targeting Strategy	Compound	Company	Specificity	Disease	Phase	ClinicalTrials.gov
Global IL-6 signaling Targeting soluble and membrane-bound Interleukin 6 receptor	Tocilizumab (ACTEMRA, RoACTEMRA)	Chugai, Roche	Humanized IL-6 receptor-specific mAb	Castleman's disease	2005, Japan	Approved
				Juvenile idiopathic arthritis	2008, Japan	Approved
				Systemic-onset juvenile idiopathic arthritis	2008, Japan	Approved
				Rheumatoid Arthritis	2009, EMEA; 2010, FDA	Approved
				Adult's still disease	Phase II; 2009–11	NCT01002781
				Graves' ophthalmopathy	Phase III; 2011–13	NCT01297699
				Relapsing polychondritis	Phase II; 2010–12	NCT01104480
				Ankylosing spondylitis	Phase III; 2010–14	NCT01209702
				Type II diabetes and exercise	2010–12	NCT01073826
				Rheumatoid arthritis	Phase II; 2010–12	NCT01217814
Global IL-6 signaling Targeting Interleukin 6	ALX-0061 CDP6038 (Olokizumab)	Regeneron/ Sanofi-Aventis	Fully human IL-6 receptor-specific mAb	Rheumatoid arthritis	Phase II + III; 2010–13	NCT01061736
				Ankylosing spondylitis	Phase III; 2010–15	NCT01146652
					Phase II; 2010–11	NCT01284569
					Phase III; 2010–15	NCT01118728
				Rheumatoid arthritis	Phase I + II; 2011–12	NCT01242488
				Rheumatoid arthritis	Phase I + II; 2009–10	NCT01009242
					Phase II; 2010–12	NCT01242488
					Phase II; 2011–14	NCT01296711
				Rheumatoid arthritis	Phase II; 2008–11	NCT00718718
				Castleman disease	Phase II; 2010–12	NCT01024036
IL-6 Trans-signaling Targeting IL-6/sIL-6R Downstream signaling Targeting janus kinases (JAKs) downstream of gp130/IL6ST	CNTO136 (Sirukumab) CNTO328 (cCLB, Siltuximab)	Centocor, Inc., Centocor, Inc.,	Fully human IL-6-specific mAb Chimeric IL-6-specific mAb	Multiple myeloma [125]	Phase II; 2006–12	NCT00402181
					Phase II; 2009–14	NCT00911859
					Phase III; 2010–14	NCT01266811
				Prostate cancer	Phase I; 2005–10	NCT00401765
					Phase II; 2006–008	NCT00385827
					Phase II; 2007–09	NCT00433446
				Solid tumors	Phase I + II; 2009–11	NCT00841191
				Metastatic renal cell carcinoma	Phase I + II; 2005–10	NCT00265135
				Metastatic kidney cancer	Phase II; 2006–10	NCT00311545
				Rheumatoid arthritis	Phase II; 2008–09	NCT00867516
IL-6 Trans-signaling Targeting IL-6/sIL-6R Downstream signaling Targeting janus kinases (JAKs) downstream of gp130/IL6ST	ALD518 (BMS-945429)	Alder Biopharm./ Bristol-Myers Squibb	Humanized IL-6-specific mAb, aglycosylated	Non-small cell lung-cancer related fatigue and cachexia	Phase II; 2008–09	NCT00866970
				Crohn's disease	Phase I; 2006–07	NCT00353756
				Crohn's disease	Preclinical	
				Rheumatoid arthritis	Phase III; 2013	NCT00661661
				Kidney transplant	Phase II; 2006	NCT00106639
				Psoriasis	Phase I-III; 2004–14	NCT01519089
				Rheumatoid arthritis	Phase I; 2012–13	NCT01741493
				Multiple myeloma	Phase II; 2010	NCT00639002
				Myelofibrosis	Phase II; 2012–13	NCT01340651
				Plaque psoriasis	Phase II; 2009	NCT00820950
IL-6 Trans-signaling Targeting IL-6/sIL-6R Downstream signaling Targeting janus kinases (JAKs) downstream of gp130/IL6ST	Baricitinib (INCB-28050) GLPG-0634	Eli Lilly/Incyte Abbot/Galapagos	JAK1 and 2 JAK1	Rheumatoid arthritis	Phase II; 2010	NCT00902486
				Rheumatoid arthritis	Phase I+ II; 2011–12	NCT01384422
						NCT01668641
						NCT01287858
IL-6 Trans-signaling Targeting IL-6/sIL-6R Downstream signaling Targeting janus kinases (JAKs) downstream of gp130/IL6ST	AC430	Ambit Biosciences	JAK2	Rheumatoid arthritis	Phase I, 2011	NCT01287858

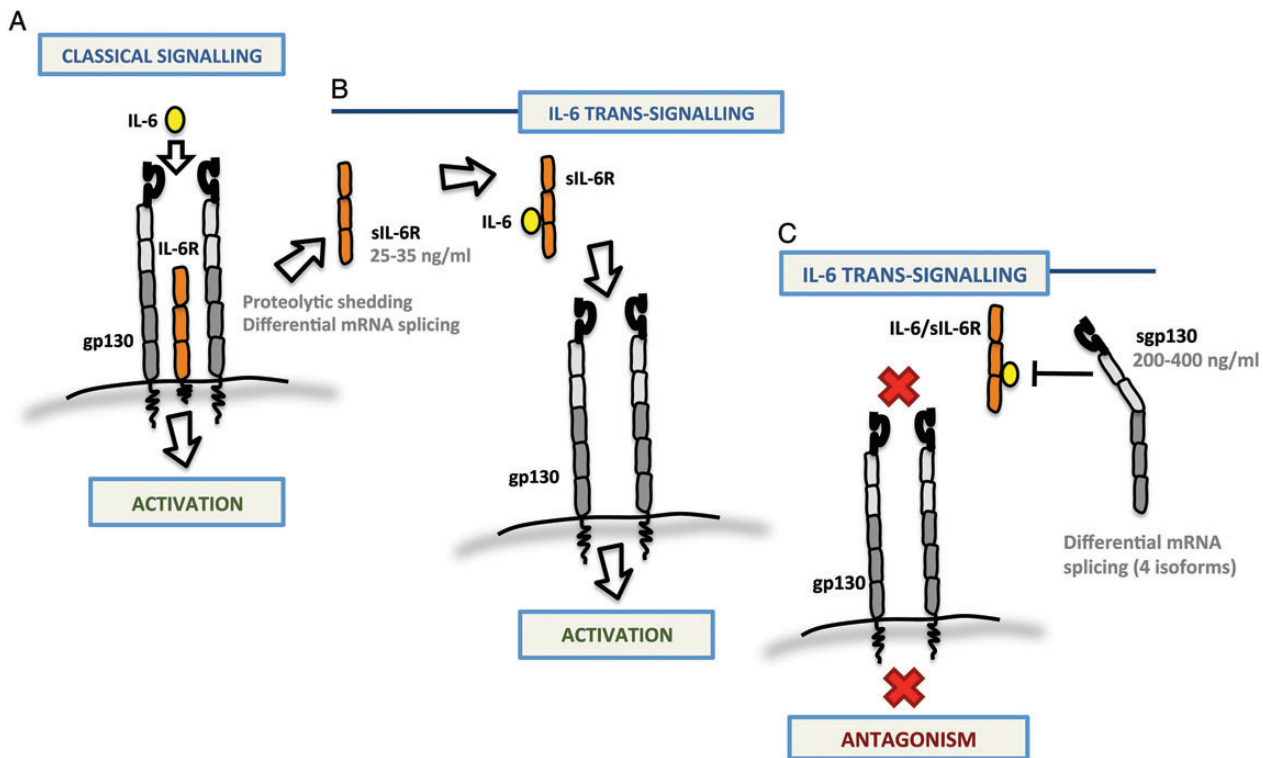


FIGURE 1: Interleukin-6 and mechanisms for receptor signalling. Interleukin-6 activates cells through two distinct mechanisms termed classical IL-6R signalling and IL-6 trans-signalling. The receptor complex responsible for regulating IL-6 responses consists of a non-signalling cognate receptor (IL-6R, CD126), which binds IL-6 and dimerizes with the signal-transducing receptor subunit gp130. (A) Classical IL-6R signalling occurs in cell types that inherently express both IL-6R and gp130. (B) While expression of IL-6R has a restricted cellular expression (hepatocytes, leukocytes and certain epithelial cells), these cells also generate a soluble form of the IL-6R (sIL-6R) that retains cytokine-binding properties and mediates IL-6 responses in cell types that lack IL-6R, but expression gp130 (IL-6 trans-signalling). (C) In many instances, IL-6 trans-signalling regulates various inflammatory activities and must be tightly regulated. Here, a soluble form of gp130, which circulates at high serum concentrations acts as an antagonist of IL-6 trans-signalling and binds IL-6 only when bound to sIL-6R.

IL-6 IN GLOMERULONEPHRITIS, CHRONIC KIDNEY DISEASE AND ACUTE KIDNEY INJURY

185 Clinical and experimental studies suggest that IL-6 contribute to renal injury in glomerulonephritis and other forms of renal disease. For example, elevated IL-6 expression in kidneys and urine of patients with mesangial proliferative glomerulonephritis is often associated with poor outcome [36]. In this context, IL-6 induces mesangial cell proliferation [36]. In murine models of lupus nephritis IL-6 activities promote tissue damage and disease severity [37–39], while transgenic mice displaying elevated levels of circulating IL-6 develop proteinuric nephropathy that culminates in death from renal failure [40].

190 Ultimately, the role of IL-6 in these conditions may relate to IL-6 involvement in fibrosis and tissue damage. In models of angiotensin-II induced renal disease, infusion of angiotensin-II induces IL-6 expression and renal fibrosis. This response is IL-6 dependent and IL-6-deficient mice remain resistant to renal injury [41]. Mechanistically, IL-6 may contribute to

200 renal injury by enhancing the signalling response of tubular epithelial cells to pro-fibrotic cytokines such as transforming growth factor- β (TGF β) [42]. However, data from other experimental models of renal injury show that IL-6 is not always

a key facet of progressive kidney damage [43–45]. What factors contribute to these differences in disease outcome remain to be established, but may reflect differences in disease induction methods or protocols using cytokine-deficient animals versus direct manipulation of cytokine activity in wild type strains.

In patients with acute kidney injury (AKI), high circulating levels of IL-6 are predictive of increased mortality [10]. This outcome is also seen in murine models of ischaemia reperfusion injury- and nephrotoxin-induced models of AKI [11, 46, 47]. In mercuric chloride-induced AKI, IL-6 deficient mice exhibit less kidney-associated inflammation, and have improved outcome [47]. In the same study, IL-6 trans-signalling in tubular epithelial cells ameliorated injury and led to preservation of renal function. This led the authors to conclude that IL-6 simultaneously promotes an injurious inflammatory response and, through a mechanism involving IL-6 trans-signalling, protects the kidney from further injury [47]. These studies are akin to the role of IL-6 and IL-6 trans-signalling in regulating homeostatic gut epithelia remodelling versus colitis-like inflammation [9]. Significantly, tocilizumab is not prescribed in patients with a history of diverticulitis [2, 9].

Biologics against IL-6 are highly effective in autoimmune conditions including rheumatoid arthritis [1, 2]. To date, there is little data relating to IL-6 blockade in patients with renal

230 disease. Case reports of tocilizumab use in patients with condi-
tions where renal complications is an associated co-morbidity
show promising improvements in urinary sediment, proteinu-
ria and stabilization of renal function. These include the lym-
phoproliferative disorder multicentric Casteman's disease [48,
49], AA Amyloidosis [50, 51] and a subset of five patients with
235 renal dysfunction in a Phase I study of SLE [52]. While these
studies endorse the potential clinical application of IL-6 tar-
geted interventions in acute or chronic renal disease, definitive
randomized controlled trials are lacking.

240 INTERLEUKIN-6 IN RENAL TRANSPLANTATION

Renal transplantation is considered to be the 'gold standard'
treatment for most patients with end stage renal failure. Bene-
fits include improved patient survival, quality of life and
healthcare cost [53]. Recent advances in treatment means that
245 the rate of acute clinical rejection (AR) has fallen. However,
AR remains a determining factor for the development of
chronic rejection and long-term allograft survival [53, 54].

Interleukin-6 has long been highlighted as a pro-inflammatory
cytokine associated with renal allograft rejection. While IL-6
250 levels are low in the serum and urine of healthy individuals,
renal transplant recipients display high serum and urinary
IL-6 levels immediately post transplantation and during AR
[55, 56]. For example, increased IL-6 mRNA transcripts have
been identified in renal biopsies from patients undergoing AR
255 [57]. Notably, while AR episodes have been associated with
increased serum and urine IL-6 levels, preventative rejection
treatments stabilize IL-6 expression and return them to
baseline [12, 58].

Most studies have underlined the greater sensitivity of urinary
260 IL-6, over serum measurements, as potential indicators of
rejection. For example urinary actin, IL-6 and CXC-chemokine
ligand 8 (CXCL8) have been proposed as biomarkers of sus-
tained acute renal failure in allograft recipients [59]. Kwon
et al. [59] observed elevated urinary IL-6 excretion in patients
265 displaying sustained acute renal failure compared with those
that went on to recover. For patients showing sustained failure,
urinary IL-6 was increased on the day of transplant and also
remained higher at postoperative Day 5. In a recent analysis of
90 transplant patients, stable allograft recipients showed
270 similar levels of serum IL-6 to healthy individuals. However
patients undergoing allograft rejection displayed significant in-
creases in circulating IL-6 [60]. Notably, higher IL-6 levels
were observed in individuals undergoing chronic allograft re-
jection compared with patients in AR [60]. Interestingly, while
275 increases in serum and urinary IL-6 are associated with AR,
sIL-6R levels do not correlate with rejection [12]. While the
mechanisms affecting these outcomes are far from clear, they
may relate to associated genetic factors. Meta-analysis of trans-
plant patients shows that alterations in AR risk are associated
280 with individuals bearing the rs1800795 'high' IL-6 producing
loci [61, 62]. Notably, in allogenic haematopoietic cell trans-
plantation, the rs1800795 variant is linked with increased risk
of developing acute graft-versus-host disease [62]. Therefore,

IL-6 donor genotype may be more important in graft rejection
than recipient genotype. Here, IL-6 may serve as an immune 285
'danger signal' thereby disrupting allograft tolerance.

Mechanisms underpinning the role of IL-6 in allograft
transplant rejection may hinge on its partnership with TGF β ,
which together balance the induction of T-cell tolerance
versus pro-inflammatory effector responses. Regulatory T-cells
290 (T_{reg}) provide tolerance by suppression of allo- and auto-
immune responses [63–65]. These cells are defined by their ex-
pression of transcription factor FoxP3, and are either thymus
derived (natural T_{reg}; nT_{reg}) or can differentiate from naïve
CD4⁺ T-cells activated in the presence of TGF β (induced T_{reg};
295 iT_{reg}) [66]. Such has been the impact of T_{reg} cells on allograft
tolerance in experimental animal models, that recent studies
have proposed FoxP3 as a prognostic marker for renal allograft
outcome (see review [67]). Interestingly, IL-6 inhibits the
TGF β -mediated differentiation of iT_{reg} cells, instead favouring
300 the development of IL-17-producing CD4⁺ T-cells (T_h17 cells)
[68, 69]. While T_{reg} cells provide tolerance, there is mounting
evidence of a role for T_h17 cells in allograft rejection. For
example, allograft infiltrating T_h17 cells were associated with
hallmarks of chronic rejection including exacerbated vasculo-
305 pathology and fibrosis in models of cardiac allograft rejection [70,
71]. In renal allografts, elevated IL-17 mRNA and protein has
been demonstrated during AR in both clinical patients and ex-
perimental models [72–74]. Owing to the bias of IL-6 for in-
ducing effector T_h17 cells rather than regulatory T_{reg}
310 populations with tolerogenic properties in allogenic grafts,
therapeutically targeting the IL-6 signalling pathway may
prove beneficial to renal transplant outcomes in patients
undergoing acute and chronic rejection.

315 THE BIOLOGY OF INTERLEUKIN-6 IN RENAL REPLACEMENT THERAPY

Haemodialysis (HD) and peritoneal dialysis (PD) represent
the two major renal replacement therapies available for pa-
tients with end-stage renal disease (ESRD). Both IL-6 and sIL-
6R are considered important prognostic markers of clinical
320 outcome in ESRD patients. ESRD patients have elevated
serum IL-6 levels prior to treatment [75, 76]. Impaired excre-
tion due to reduced kidney function has been suggested as one
reason for this elevation [77], although IL-6 mRNA is in-
creased in the peripheral blood mononuclear cells of ESRD pa-
325 tients [78]. PD treatment itself leads to increases in systemic
and intraperitoneal IL-6 and sIL-6R levels [79]. Here, systemic
elevations in circulating levels may reflect a consequence of
persistent or episodic bouts of inflammation, patient co-
morbidities, genetic factors, obesity, alterations in metabolism,
330 infection incidence or other immunological events [80, 81].
However, raised serum IL-6 and sIL-6R levels at the beginning
of treatment remain powerful predictors of mortality in both
HD and PD patients [17, 75, 82, 83]. These changes may
reflect the systemic inflammatory status of a patient, and often
335 corresponds to elevations in C-reactive protein [83, 84]. For
example, high IL-6 levels contribute to dialysis associated mal-
nutrition [13, 14] and are prognostic of cardiovascular risk

240
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[85], which is an adverse outcome of haemodialysis [15]. Here, systemic elevations in IL-6 likely arise from the liver, muscle and the inflammatory activation of stromal tissues or myeloid cells (Table 2).

Patients receiving PD often experience a number of clinical complications. These include: (i) susceptibility to recurrent episodes of peritonitis and (ii) changes in the structure of the peritoneal membrane resulting in loss of ultrafiltration capacity and treatment failure. PD-associated peritonitis is caused predominantly by Gram-positive *Staphylococcus* species (most commonly by *Staphylococcus epidermis* and *Staphylococcus aureus*), but also by Gram-negative bacteria (e.g. *Escherichia coli*) and fungi (e.g. *Candida albicans*) [86]. Here, IL-6 is essential for the appropriate control of acute inflammation and promotes bacterial clearance. Murine models of peritonitis have shown IL-6/sIL-6R signalling via STAT3 regulates leukocyte recruitment and activation [87–91], and IL-6-deficient mice are less able to clear a number of bacterial species [4, 92] (unpublished data).

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Interestingly, individuals with defects in IL-6 or STAT3 display an impaired immune defense against *Staphylococcal* infection [93, 94], implying IL-6 is an essential part of the immune response to *Staphylococcus* species. Under these conditions, IL-6 is highly expressed by resident peritoneal leukocytes and mesothelial cells following microbial sensing by pattern recognition receptors [95–98]. Indeed elevated concentrations of IL-6 and sIL-6R are present in peritoneal dialysis effluent of patients during acute episodes of bacterial peritonitis [88].

Extended PD therapy is associated with functional and morphologic alterations to the peritoneal membrane and result in PD treatment failure [99]. These changes may be induced by uraemia, hyperglycaemia, prolonged exposure to bio-incompatible peritoneal dialysis fluids, age and recurrent episodes of peritonitis. Here, vascular alterations contribute to increased peritoneal solute transport [100]. IL-6 levels are also an important determinate of solute transport in PD [101]. Intraperitoneal IL-6 and sIL-6R levels significantly correlate with the baseline peritoneal solute transport (PSTR) observed in PD patients and are predictive of pro-angiogenic factors present in the dialysate (CCL2; CC-chemokine ligand 2 and VEGF; vascular endothelial growth factor) [102]. In this regard, plasma levels of IL-6 and VEGF are associated with a high PSTR [17]. Again, the rs1800795 genetic loci relate to intraperitoneal IL-6 levels and baseline PSTR [103], and represent an independent risk factor for mortality and treatment failure [104]. Regarding the role of IL-6 in fibrotic changes,

IL-6 plays a clear role in normal wound healing [105] and fibrogenesis in various organs (e.g. lung, skin and kidney) [41, 106,107]. Recently, we have found that IL-6 is essential for the development of peritoneal fibrosis following recurrent peritonitis in a murine model [108]. IL-6-dependent changes in peritoneal Th1 responses and IFN- γ and STAT1 activation within the peritoneal membrane lead to alterations in the balance of matrix metalloproteinase to tissue inhibitors of matrix metalloproteinases. Collectively, these studies demonstrate a fundamental aspect of IL-6 involvement in inflammation and specifically emphasize its importance in governing the balance between provision of competent host defense and inflammation-induced tissue injury.

INTERLEUKIN-6, COMORBIDITIES AND URAEMIA IN END-STAGE RENAL DISEASE

Considerable emphasis has been placed on the relationship between IL-6 (and to a lesser extent sIL-6R) and C-reactive protein, cardiovascular risk and patient outcomes such as fatigue. However IL-6 also functions as a homeostatic regulator of catabolism, iron uptake and muscle wasting. The IL-6 control of these processes has a major bearing on patients with ESRD and affects the incidence of anaemia, protein-energy wasting and muscle atrophy [16, 109–113]. For example, the hepatic control of hepcidin expression in response to systemic elevations in IL-6 disrupts iron homeostasis and leads to iron-restricted erythropoiesis and anaemia [114–116]. In patients with rheumatoid arthritis, treatment with the blocking anti-IL-6R monoclonal antibody tocilizumab rapidly improves anaemia by reducing serum hepcidin [117]. In ESRD many of these processes are ultimately influenced by underlining alterations in uraemia. Studies show that uraemia is a contributing factor in the control of increased plasma IL-6 concentrations [76, 118]. Thus, therapeutic control of IL-6 with selective anti-cytokine interventions in combination with a standard treatment for uraemia is likely to improve many compounding complications associated with the clinical management of ESRD [112, 119, 120].

INTERLEUKIN-6 AND CLINICAL OUTCOMES

It is widely acknowledged that cytokines play an integral role in determining the course of disease and IL-6 is increasing

Table 2. Role of the IL-6 pathway in renal disease, transplantation and renal replacement therapy

Factor	Condition	Associated outcome	Reference
IL-6	AKI	Mortality	[10]
	Ischaemia reperfusion injury	Unclear	[11]
	Chronic allograft rejection	Acute rejection	[12]
	Haemodialysis and peritoneal dialysis	Mortality	Pecoits-Filho <i>et al.</i> (2002) [17]
		Malnutrition	[13, 14]
IL-6R	Haemodialysis and peritoneal dialysis	Vascular changes	[15]
		Mortality	Pecoits-Filho <i>et al.</i> (2002) [17]

This table summarizes the published clinical and experimental studies linking the IL-6 pathway to renal disease, transplantation and renal replacement therapy, and its associated outcomes.

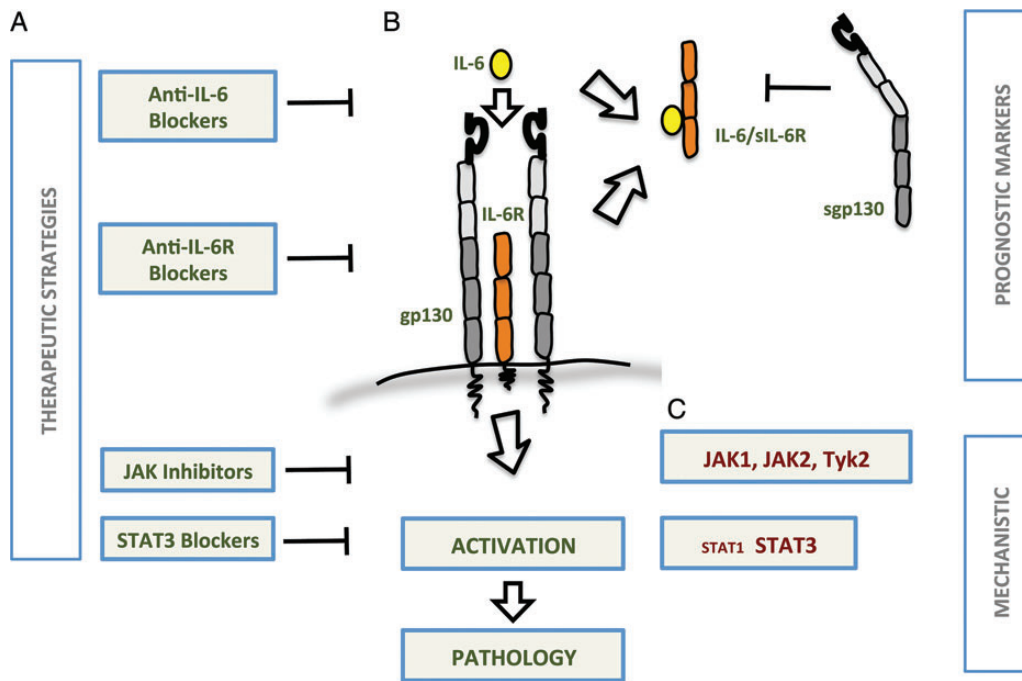


FIGURE 2: Clinical implications for IL-6 research. (A) Various therapeutic strategies are either in standard clinical practice or emerging through pre-clinical studies and early stage clinical trials. These include specific cytokine and cytokine receptor blockers and less selective inhibitors of signal transduction components. (B) Measurement of IL-6, sIL-6R and sgp130 are increasingly being viewed as prognostic indicators and inform clinical decisions ranging from the overall severity of inflammation to co-morbidities including cardiovascular risk. (C) An increased understanding of IL-6 signalling is identifying novel mechanisms for the involvement of IL-6 in autoimmunity, disease and cancer. Such research has also identified crosstalk between IL-6 signalling and other cytokines or immune sensing mechanisms.

viewed as major drug targets for therapy (Figure 2). The application of biologics in conditions such as rheumatoid arthritis emphasize that early therapeutic intervention is essential to ensure appropriate management of the condition and potential disease remission [2]. While certain biologics (e.g. bevacizumab, a humanized monoclonal antibody to VEGF-A) have shown therapeutic efficacy in forms kidney cancer, their application in chronic renal diseases appears to have minimal appeal. This in part may reflect the success of renal transplantation. Instead, clinical assessments of cytokine expression or activities may be considered important prognostic or predictive biomarkers that forecast the course of disease and aid treatment decisions. For example, the monitoring of IL-6 in PD patients is providing valuable information on the loss of ultra-filtration capacity, fibrosis onset, the control of infection and treatment failures [17, 77, 79, 82, 83, 102]. Here, IL-6 activities not only predict local disease processes, but also provide valuable information on systemic inflammatory events and patient co-morbidities (Figure 2). For example, detection of serum IL-6 in PD patients significantly increases with time on dialysis and, as seen in other chronic diseases, correlates with indices of cardiovascular risk [121]. Such findings offer a valuable addition to the standard measurement of C-reactive protein and provide additional information on the potential efficiency of treatment. While the clinical assessment of C-reactive protein is used to reflect the degree of systemic inflammation and potential cardiovascular risk associated with a patient's disease, the role of C-reactive protein in determining the underlining pathology is unclear [122]. Interleukin-6 is the principle driver

of C-reactive protein expression and may be viewed as a surrogate marker of IL-6 bioactivity. In this context, the clinical assessment of IL-6 may provide a more powerful prediction of inflammation burden in ESRD [123, 124].

The impact of chronic kidney disease on healthcare systems around the world is increasing. In response, it is essential for new clinical assessments to be applied to provide a more personalized approach to patient stratification, and improvements in treatment decisions. Studies emphasize that IL-6 and associated downstream signalling events may represent one such marker (Figure 2). However to identify the pathways contributing to chronic disease progression in patients with varying degrees of renal disease, we must understand how cytokines like IL-6 govern acute resolving inflammation and how their activities become distorted to drive chronic inflammation.



REFERENCES

- McInnes IB, Schett G. Cytokines in the pathogenesis of rheumatoid arthritis. *Nat Rev Immunol* 2007; 7: 429–442
- Choy EH, Kavanaugh AF, Jones SA. The problem of choice: current biologic agents and future prospects in RA. *Nat Rev Rheumatol* 2013; 9: 154–163
- Kishimoto T. IL-6: from its discovery to clinical applications. *Int Immunol* 2010; 22: 347–352
- Kopf M *et al.* Impaired immune and acute-phase responses in interleukin-6-deficient mice. *Nature* 1994; 368: 339–342
- Schobitz B *et al.* Soluble interleukin-6 (IL-6) receptor augments central effects of IL-6 in vivo. *FASEB J* 1995; 9: 659–664

6. Waage A, Brandtzaeg P, Halstensen A *et al.* The complex pattern of cytokines in serum from patients with meningococcal septic shock. Association between interleukin 6, interleukin 1, and fatal outcome. *J Exp Med* 1989; 169: 333–338
- 485 7. Silver JS, Hunter CA. gp130 at the nexus of inflammation, autoimmunity, and cancer. *J Leukoc Biol* 2010; 88: 1145–1156
8. Jones SA. Directing transition from innate to acquired immunity: defining a role for IL-6. *J Immunol* 2005; 175: 3463–3468
9. Jones SA, Scheller J, Rose-John S. Therapeutic strategies for the clinical blockade of IL-6/gp130 signaling. *J Clin Invest* 2011; 121: 3375–3383
- 490 10. Simmons EM *et al.* Plasma cytokine levels predict mortality in patients with acute renal failure. *Kidney Int* 2004; 65: 1357–1365
11. Lemay S, Rabb H, Postler G *et al.* Prominent and sustained up-regulation of gp130-signaling cytokines and the chemokine MIP-2 in murine renal ischemia-reperfusion injury. *Transplantation* 2000; 69: 959–963
- 495 12. Waiser J *et al.* Interleukin-6 expression after renal transplantation. *Nephrol Dial Transplant* 1997; 12: 753–759
13. Kaizu Y *et al.* Interleukin-6 may mediate malnutrition in chronic hemodialysis patients. *Am J Kidney Dis* 1998; 31: 93–100
- 500 14. Beberashvili I *et al.* IL-6 levels, nutritional status, and mortality in prevalent hemodialysis patients. *Clin J Am Soc Nephrol* 2011; 6: 2253–2263
15. Stenvinkel P, Heimbürger O, Jogestrand T. Elevated interleukin-6 predicts progressive carotid artery atherosclerosis in dialysis patients: association with *Chlamydia pneumoniae* seropositivity. *Am J Kidney Dis* 2002; 39: 274–282
- 505 16. Stenvinkel P *et al.* IL-10, IL-6, and TNF- α : central factors in the altered cytokine network of uremia—the good, the bad, and the ugly. *Kidney Int* 2005; 67: 1216–1233
17. Pecoits-Filho R *et al.* Plasma and dialysate IL-6 and VEGF concentrations are associated with high peritoneal solute transport rate. *Nephrol Dial Transplant* 2002; 17: 1480–1486
- 510 18. O’Shea JJ, Laurence A, McInnes IB. Back to the future: oral targeted therapy for RA and other autoimmune diseases. *Nat Rev Rheumatol* 2013; 9: 173–182
19. Rose-John S, Heinrich PC. Soluble receptors for cytokines and growth factors: generation and biological function. *Biochem J* 1994; 300: 281–290
- 515 20. Heinrich PC *et al.* Principles of interleukin (IL)-6-type cytokine signaling and its regulation. *Biochem J* 2003; 374: 1–20
21. Fishman D *et al.* The effect of novel polymorphisms in the interleukin-6 (IL-6) gene on IL-6 transcription and plasma IL-6 levels, and an association with systemic-onset juvenile chronic arthritis. *J Clin Invest* 1998; 102: 1369–1376
- 520 22. Esteve E *et al.* Polymorphisms in the interleukin-6 receptor gene are associated with body mass index and with characteristics of the metabolic syndrome. *Clin Endocrinol* 2006; 65: 88–91
- 525 23. Hamid YH *et al.* Variation in the interleukin-6 receptor gene associates with type 2 diabetes in Danish whites. *Diabetes* 2004; 53: 3342–3345
24. Hsieh CH *et al.* Interleukin-6 receptor gene 48892 A/C polymorphism is associated with metabolic syndrome in female Taiwanese adolescents. *Genet Test Mol Biomark* 2012; 16: 1376–1381
- 530 25. Qi L, Rifai N, Hu FB. Interleukin-6 receptor gene variations, plasma interleukin-6 levels, and type 2 diabetes in U.S. Women. *Diabetes* 2007; 56: 3075–3081
26. Qi L, Zhang C, van Dam RM *et al.* Interleukin-6 genetic variability and adiposity: associations in two prospective cohorts and systematic review in 26,944 individuals. *J Clin Endocrinol Metab* 2007; 92: 3618–3625
- 535 27. Song Y *et al.* The interaction between the interleukin 6 receptor gene genotype and dietary energy intake on abdominal obesity in Japanese men. *Metab Clin Exp* 2007; 56: 925–930
28. Wolford JK, Colligan PB, Gruber JD *et al.* Variants in the interleukin 6 receptor gene are associated with obesity in Pima Indians. *Mol Genet Metab* 2003; 80: 338–343
- 540 29. Lee RC, Feinbaum RL, Ambros V. The *C. elegans* heterochronic gene lin-4 encodes small RNAs with antisense complementarity to lin-14. *Cell* 1993; 75: 843–854
- 545 30. Pasquinelli AE *et al.* Conservation of the sequence and temporal expression of let-7 heterochronic regulatory RNA. *Nature* 2000; 408: 86–89
31. Viswanathan SR *et al.* Lin28 promotes transformation and is associated with advanced human malignancies. *Nat Genet* 2009; 41: 843–848
32. Iliopoulos D, Hirsch HA, Struhl K. An epigenetic switch involving NF-kappaB, Lin28, Let-7 MicroRNA, and IL6 links inflammation to cell transformation. *Cell* 2009; 139: 693–706
33. Zhu LH *et al.* MicroRNA-23a promotes the growth of gastric adenocarcinoma cell line MGC803 and downregulates interleukin-6 receptor. *FEBS J* 2010; 277: 3726–3734
- 555 34. Iliopoulos D, Jaeger SA, Hirsch HA *et al.* STAT3 activation of miR-21 and miR-181b-1 via PTEN and CYLD are part of the epigenetic switch linking inflammation to cancer. *Mol Cell* 2010; 39: 493–506
35. Fabbri M *et al.* MicroRNAs bind to Toll-like receptors to induce prometastatic inflammatory response. *Proc Natl Acad Sci U S A* 2012; 109: E2110–E2116
- 560 36. Horii Y *et al.* Involvement of IL-6 in mesangial proliferative glomerulonephritis. *J Immunol* 1989; 143: 3949–3955
37. Kiberd BA. Interleukin-6 receptor blockage ameliorates murine lupus nephritis. *J Am Soc Nephrol* 1993; 4: 58–61
38. Liang B, Gardner DB, Griswold DE *et al.* Anti-interleukin-6 monoclonal antibody inhibits autoimmune responses in a murine model of systemic lupus erythematosus. *Immunology* 2006; 119: 296–305
39. Ryffel B *et al.* Interleukin-6 exacerbates glomerulonephritis in (NZB \times NZW) F1 mice. *Am J Pathol* 1994; 144: 927–937
40. Fattori E *et al.* Development of progressive kidney damage and myeloma kidney in interleukin-6 transgenic mice. *Blood* 1994; 83: 2570–2579
- 570 41. Zhang W *et al.* Interleukin 6 underlies angiotensin ii-induced hypertension and chronic renal damage. *Hypertension* 2012; 59: 136–144
42. Zhang XL, Topley N, Ito T *et al.* Interleukin-6 regulation of transforming growth factor (TGF)- β receptor compartmentalization and turnover enhances TGF- β 1 signaling. *J Biol Chem* 2005; 280: 12239–12245
43. Eitner F *et al.* Role of interleukin-6 in mediating mesangial cell proliferation and matrix production in vivo. *Kidney Int* 1997; 51: 69–78
44. Karkar AM, Smith J, Tam FW *et al.* Abrogation of glomerular injury in nephrotoxic nephritis by continuous infusion of interleukin-6. *Kidney Int* 1997; 52: 1313–1320
- 580 45. Yang J *et al.* Effect of interleukin 6 deficiency on renal interstitial fibrosis. *PLoS One* 2012; 7: e52415
46. Vaidya VS, Shankar K, Lock EA *et al.* Molecular mechanisms of renal tissue repair in survival from acute renal tubule necrosis: role of ERK1/2 pathway. *Toxicol Pathol* 2003; 31: 604–618
47. Nechemia-Arbely Y *et al.* IL-6/IL-6R axis plays a critical role in acute kidney injury. *J Am Soc Nephrol* 2008; 19: 1106–1115
48. Komaba H, Nakazawa T, Yamaguchi Y *et al.* Interleukin-6 receptor inhibition with tocilizumab in various renal involvements associated with multicentric Castelman’s disease: a report of three cases. *NDT Plus* 2008; 1: 423–426
- 590 49. Maeshima A *et al.* Efficacy of tocilizumab, a humanized neutralizing antibody against interleukin-6 receptor, in progressive renal injury associated with Castelman’s disease. *CEN Case Rep* 2012; 1: 7–11
- 595 50. Magro-Checa C *et al.* Successful use of tocilizumab in a patient with nephrotic syndrome due to a rapidly progressing AA amyloidosis secondary to latent tuberculosis. *Amyloid* 2011; 18: 235–239
51. Canas-Ventura A, Rodriguez E, Andreu M *et al.* Tocilizumab in amyloidosis-associated kidney disease secondary to inflammatory bowel diseases. *Dig Dis Sci* 2013; 58: 2736–2737
- 600 52. Illei GG *et al.* Tocilizumab in systemic lupus erythematosus: data on safety, preliminary efficacy, and impact on circulating plasma cells from an open-label phase I dosage-escalation study. *Arthritis Rheum* 2010; 62: 542–552
- 605 53. Thiruchelvam PT, Willicombe M, Hakim N *et al.* Renal transplantation. *BMJ* 2011; 343: d7300
54. Goldfarb-Rumyantzev AS, Naiman N. Genetic predictors of acute renal transplant rejection. *Nephrol Dial Transplant* 2010; 25: 1039–1047
55. Van Oers MH, Van der Heyden AA, Aarden LA. Interleukin 6 (IL-6) in serum and urine of renal transplant recipients. *Clin Exp Immunol* 1988; 71: 314–319
- 610 56. Casiraghi F *et al.* Sequential monitoring of urine-soluble interleukin 2 receptor and interleukin 6 predicts acute rejection of human renal allografts before clinical or laboratory signs of renal dysfunction. *Transplantation* 1997; 63: 1508–1514
- 615 57. Vandenbroecke C *et al.* Differential in situ expression of cytokines in renal allograft rejection. *Transplantation* 1991; 51: 602–609

58. Budde K, Waiser J, Neumayer HH. The diagnostic value of GM-CSF and IL-6 determinations in patients after renal transplantation. *Transplant Int* 1994; 7: S97–S101
59. Kwon O, Molitoris BA, Pescovitz M *et al.* Urinary actin, interleukin-6, and interleukin-8 may predict sustained ARF after ischemic injury in renal allografts. *Am J Kidney Dis* 2003; 41: 1074–1087
60. Sonkar GK, Singh S, Sonkar SK *et al.* Evaluation of serum interleukin 6 and tumour necrosis factor alpha levels, and their association with various non-immunological parameters in renal transplant recipients. *Singapore Med J* 2013; 54: 511–515
61. Lv R *et al.* Association between IL-6 -174G/C polymorphism and acute rejection of renal allograft: evidence from a meta-analysis. *Transplant Immunol* 2012; 26: 11–18
62. Chien JW *et al.* Evaluation of published single nucleotide polymorphisms associated with acute GVHD. *Blood* 2012; 119: 5311–5319
63. Kendal AR *et al.* Sustained suppression by Foxp3+ regulatory T cells is vital for infectious transplantation tolerance. *J Exp Med* 2011; 208: 2043–2053
64. Regateiro FS, Howie D, Cobbold SP *et al.* TGF-beta in transplantation tolerance. *Curr Opin Immunol* 2011; 23: 660–669
65. Faust SM, Lu G, Wood SC *et al.* TGFbeta neutralization within cardiac allografts by decorin gene transfer attenuates chronic rejection. *J Immunol* 2009; 183: 7307–7313
66. Afzali B, Lombardi T, Lechler RI *et al.* The role of T helper 17 (Th17) and regulatory T cells (Treg) in human organ transplantation and autoimmune disease. *Clin Exp Immunol* 2007; 148: 32–46
67. Zuber J *et al.* Prognostic significance of graft Foxp3 expression in renal transplant recipients: a critical review and attempt to reconcile discrepancies. *Nephrol Dial Transplant* 2013; 28: 1100–1111
68. Bettelli E *et al.* Reciprocal developmental pathways for the generation of pathogenic effector TH17 and regulatory T cells. *Nature* 2006; 441: 235–238
69. Chen X *et al.* Blockade of interleukin-6 signaling augments regulatory T-cell reconstitution and attenuates the severity of graft-versus-host disease. *Blood* 2009; 114: 891–900
70. Faust SM *et al.* Role of T cell TGFbeta signaling and IL-17 in allograft acceptance and fibrosis associated with chronic rejection. *J Immunol* 2009; 183: 7297–7306
71. Yuan X *et al.* A novel role of CD4 Th17 cells in mediating cardiac allograft rejection and vasculopathy. *J Exp Med* 2008; 205: 3133–3144
72. Loong CC, Hsieh HG, Lui WY *et al.* Evidence for the early involvement of interleukin 17 in human and experimental renal allograft rejection. *J Pathol* 2002; 197: 322–332
73. Van Kooten C *et al.* Interleukin-17 activates human renal epithelial cells in vitro and is expressed during renal allograft rejection. *J Am Soc Nephrol* 1998; 9: 1526–1534
74. Hsieh HG, Loong CC, Lui WY *et al.* IL-17 expression as a possible predictive parameter for subclinical renal allograft rejection. *Transplant Int* 2001; 14: 287–298
75. Kimmel PL *et al.* Immunologic function and survival in hemodialysis patients. *Kidney Int* 1998; 54: 236–244
76. Herbelin A, Urena P, Nguyen AT *et al.* Elevated circulating levels of interleukin-6 in patients with chronic renal failure. *Kidney Int* 1991; 39: 954–960
77. Pecoits-Filho R, Barany P, Lindholm B *et al.* Interleukin-6 is an independent predictor of mortality in patients starting dialysis treatment. *Nephrol Dial Transplant* 2002; 17: 1684–1688
78. Yamaguchi T *et al.* IL-6 mRNA synthesis by peripheral blood mononuclear cells (PBMC) in patients with chronic renal failure. *Clin Exp Immunol* 1996; 103: 279–284
79. Pecoits-Filho R, Carvalho MJ, Stenvinkel P *et al.* Systemic and intraperitoneal interleukin-6 system during the first year of peritoneal dialysis. *Perit Dial Int* 2006; 26: 53–63
80. Jofre R, Rodriguez-Benitez P, Lopez-Gomez JM *et al.* Inflammatory syndrome in patients on hemodialysis. *J Am Soc Nephrol* 2006; 17: S274–S280
81. Kaysen GA, Eiserich JP. Characteristics and effects of inflammation in end-stage renal disease. *Semin Dial* 2003; 16: 438–446
82. Bologa RM *et al.* Interleukin-6 predicts hypoalbuminemia, hypocholesterolemia, and mortality in hemodialysis patients. *Am J Kidney Dis* 1998; 32: 107–114
83. Lambie M *et al.* Independent effects of systemic and peritoneal inflammation on peritoneal dialysis survival. *J Am Soc Nephrol* 2013
84. Chung SH, Heimbürger O, Stenvinkel P *et al.* Influence of peritoneal transport rate, inflammation, and fluid removal on nutritional status and clinical outcome in prevalent peritoneal dialysis patients. *Perit Dial Int* 2003; 23: 174–183
85. Ridker PM, Rifai N, Stampfer MJ *et al.* Plasma concentration of interleukin-6 and the risk of future myocardial infarction among apparently healthy men. *Circulation* 2000; 101: 1767–1772
86. Williams JD, Coles GA. Gram-positive infections related to CAPD. *J Antimicrob Chemother* 1991; 27: 31–35
87. McLoughlin RM *et al.* Interplay between IFN-gamma and IL-6 signaling governs neutrophil trafficking and apoptosis during acute inflammation. *J Clin Invest* 2003; 112: 598–607
88. Hurst SM *et al.* IL-6 and its soluble receptor orchestrate a temporal switch in the pattern of leukocyte recruitment seen during acute inflammation. *Immunity* 2001; 14: 705–714
89. Fielding CA *et al.* IL-6 regulates neutrophil trafficking during acute inflammation via STAT3. *J Immunol* 2008; 181: 2189–2195
90. McLoughlin RM *et al.* IL-6 trans-signaling via STAT3 directs T cell infiltration in acute inflammation. *Proc Natl Acad Sci U S A* 2005; 102: 9589–9594
91. Jones GW *et al.* Loss of CD4+ T cell IL-6R expression during inflammation underlines a role for IL-6 *trans* signaling in the local maintenance of Th17 cells. *J Immunol* 2010; 184: 2130–2139
92. Onogawa T. Local delivery of soluble interleukin-6 receptors to improve the outcome of alpha-toxin producing *Staphylococcus aureus* infection in mice. *Immunobiology* 2005; 209: 651–660
93. Holland SM *et al.* STAT3 mutations in the hyper-IgE syndrome. *N Engl J Med* 2007; 357: 1608–1619
94. Puel A *et al.* Recurrent staphylococcal cellulitis and subcutaneous abscesses in a child with autoantibodies against IL-6. *J Immunol* 2008; 180: 647–654
95. Colmont CS *et al.* Human peritoneal mesothelial cells respond to bacterial ligands through a specific subset of Toll-like receptors. *Nephrol Dial Transplant* 2011; 26: 4079–4090
96. Park JH *et al.* Nod1/RICK and TLR signaling regulate chemokine and antimicrobial innate immune responses in mesothelial cells. *J Immunol* 2007; 179: 514–521
97. Strunk T *et al.* TLR2 mediates recognition of live *Staphylococcus epidermidis* and clearance of bacteremia. *PLoS One* 2010; 5: e10111
98. Kato S *et al.* Endotoxin-induced chemokine expression in murine peritoneal mesothelial cells: the role of toll-like receptor 4. *J Am Soc Nephrol* 2004; 15: 1289–1299
99. Williams JD *et al.* Morphologic changes in the peritoneal membrane of patients with renal disease. *J Am Soc Nephrol* 2002; 13: 470–479
100. Numata M *et al.* Association between an increased surface area of peritoneal microvessels and a high peritoneal solute transport rate. *Perit Dial Int* 2003; 23: 116–122
101. Cho Y *et al.* Dialysate interleukin-6 predicts increasing peritoneal solute transport rate in incident peritoneal dialysis patients. *BMC Nephrol* 2014; 15: 8
102. Oh KH *et al.* Intra-peritoneal interleukin-6 system is a potent determinant of the baseline peritoneal solute transport in incident peritoneal dialysis patients. *Nephrol Dial Transplant* 2010; 25: 1639–1646
103. Hwang YH *et al.* Effects of interleukin-6 T15A single nucleotide polymorphism on baseline peritoneal solute transport rate in incident peritoneal dialysis patients. *Perit Dial Int* 2009; 29: 81–88
104. Verduijn M *et al.* The -174G/C variant of IL6 as risk factor for mortality and technique failure in a large cohort of peritoneal dialysis patients. *Nephrol Dial Transplant* 2012; 27: 3516–3523
105. Gallucci RM *et al.* Impaired cutaneous wound healing in interleukin-6-deficient and immunosuppressed mice. *FASEB J* 2000; 14: 2525–2531
106. Barnes TC, Anderson ME, Moots RJ. The many faces of interleukin-6: the role of IL-6 in inflammation, vasculopathy, and fibrosis in systemic sclerosis. *Int J Rheumatol* 2011; 2011: 721608



107. O'Donoghue RJ *et al.* Genetic partitioning of interleukin-6 signalling in mice dissociates Stat3 from Smad3-mediated lung fibrosis. *EMBO Mol Biol* 2012; 4: 939–951
- 760 108. Wadding Ceri A, *et al.* Interleukin-6 signaling drives fibrosis in unresolved inflammation. *Immunity* 2014. <http://dx.doi.org/10.1016/j.immuni.2013.10.022>
109. Raj DS. Role of interleukin-6 in the anemia of chronic disease. *Semin Arthritis Rheum* 2009; 38: 382–388
- 765 110. Raj DS, Sun Y, Tzamaloukas AH. Hypercatabolism in dialysis patients. *Curr Opin Nephrol Hypertens* 2008; 17: 589–594
- Q8** 111. Xu H *et al.* Oxidative DNA damage and mortality in hemodialysis and peritoneal dialysis patients. *Perit Dial Int* 2014
- 770 112. Pecoits-Filho R, Lindholm B, Axelsson J *et al.* Update on interleukin-6 and its role in chronic renal failure. *Nephrol Dial Transplant* 2003; 18: 1042–1045
113. Garibotto G *et al.* Peripheral tissue release of interleukin-6 in patients with chronic kidney diseases: effects of end-stage renal disease and microinflammatory state. *Kidney Int* 2006; 70: 384–390
- 775 114. Pietrangelo A *et al.* STAT3 is required for IL-6-gp130-dependent activation of hepcidin in vivo. *Gastroenterology* 2007; 132: 294–300
115. Nemeth E *et al.* IL-6 mediates hypoferremia of inflammation by inducing the synthesis of the iron regulatory hormone hepcidin. *J Clin Investig* 2004; 113: 1271–1276
- 780 116. Wrighting DM, Andrews NC. Interleukin-6 induces hepcidin expression through STAT3. *Blood* 2006; 108: 3204–3209
- 785 117. Garnero P, Thompson E, Woodworth T *et al.* Rapid and sustained improvement in bone and cartilage turnover markers with the anti-interleukin-6 receptor inhibitor tocilizumab plus methotrexate in rheumatoid arthritis patients with an inadequate response to methotrexate: results from a substudy of the multicenter double-blind, placebo-controlled trial of tocilizumab in inadequate responders to methotrexate alone. *Arthritis Rheumat* 2010; 62: 33–43
118. Jacobs P, Glorieux G, Vanholder R. Interleukin/cytokine profiles in haemodialysis and in continuous peritoneal dialysis. *Nephrol Dial Transplant* 2004; 19: V41–V45 790
119. Carrero JJ *et al.* Additive effects of soluble TWEAK and inflammation on mortality in hemodialysis patients. *Clin J Am Soc Nephrol* 2009; 4: 110–118
120. Zhang L *et al.* IL-6 and serum amyloid A synergy mediates angiotensin II-induced muscle wasting. *J Am Soc Nephrol* 2009; 20: 604–612 795
121. Cho Y *et al.* Baseline serum interleukin-6 predicts cardiovascular events in incident peritoneal dialysis patients. *Perit Dial Int* 2014 
122. Meuwese CL, Stenvinkel P, Dekker FW *et al.* Monitoring of inflammation in patients on dialysis: forewarned is forearmed. *Nat Rev Nephrol* 2011; 7: 166–176 800
123. Zoccali C, Tripepi G, Mallamaci F. Dissecting inflammation in ESRD: do cytokines and C-reactive protein have a complementary prognostic value for mortality in dialysis patients? *J Am Soc Nephrol* 17, S169–S173  **Q9**
124. Panichi V *et al.* Interleukin-6 is a stronger predictor of total and cardiovascular mortality than C-reactive protein in haemodialysis patients. *Nephrol Dial Transplant* 2004; 19: 1154–1160 805
125. Voorhees PM, Manges RF, Somlo G *et al.* A phase II multicenter study of CNTO 328, an anti-IL-6 monoclonal antibody, in patients (pts) with relapsed or refractory multiple myeloma (MM). *J Clin Oncol* 2009; 27 (Suppl. Abstr. 8527) 810

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