Spatiotemporal expression of chemokines and chemokine receptors in experimental anti-myeloperoxidase antibody-mediated glomerulonephritis

B. S. van der Veen,* A. H. Petersen,*
J. A. Belperio,[†] S. C. Satchell,[‡]
P. W. Mathieson,[‡] G. Molema* and
P. Heeringa*

*Department of Pathology and Medical Biology, University Medical Center Groningen, University of Groningen, Groningen, the Netherlands, †Division of Pulmonary and Critical Care Medicine, Department of Medicine, David Geffen School of Medicine at UCLA, Los Angeles, CA, USA, and ‡Academic Renal Unit, University of Bristol, Southmead Hospital, Bristol, UK

Accepted for publication 30 June 2009 Correspondence: P. Heeringa, Department of Pathology and Medical Biology (EA11), University Medical Center Groningen, Hanzeplein 1, 9713 GZ Groningen, the Netherlands.

E-mail: p.heeringa@med.umcg.nl

Summary

Myeloperoxidase (MPO)-anti-neutrophil cytoplasmic autoantibody (ANCA)associated necrotizing crescentic glomerulonephritis (NCGN) is characterized by abundant leucocyte infiltration. Chemokines are chemotactic cytokines involved in receptor-mediated recruitment of leucocytes. Our objective was to analyse spatiotemporal gene expression of chemokines and chemokine receptors in anti-MPO-mediated NCGN, to find potential targets for intervening with leucocyte influx. NCGN was induced in mice by co-administration of anti-MPO immunoglobulin (Ig)G and lipopolysaccharide. mRNA expression levels of chemokines and chemokine receptors were analysed in whole kidney lysates as well as in laser microdissected glomeruli and tubulo-interstitial tissue 1 and 7 day(s) after NCGN induction. Several chemokines and chemokine receptors were induced or up-regulated in anti-MPO-mediated NCGN, both on day 1 (chemokines CCL3, 5; CXCL2, 5, 13; receptor CXCR2) and on day 7 (chemokines CCL2, 5, 7, 8, 17, 20; CXCL1, 2, 5, 10; CX₃CL1; receptors CCR2, 8; CX₃CR1). The expression levels of most chemokines and receptors were higher in glomeruli than in the tubulointerstitium. Because of the temporal induction of CXCR2 on day 1, we hypothesized CXCR2 as a potential target for treatment in anti-MPO-induced NCGN. Inhibition of CXCR2 using a goat-anti-CXCR2 serum prior to NCGN induction increased glomerular neutrophil influx but did not affect crescent formation and albuminuria. In conclusion, expression levels of various chemokines and chemokine receptors were increased in anti-MPO NCGN, and expressed particularly in glomeruli. These chemokines and receptors may serve as potential targets for treatment. Inhibition of a single target, CXCR2, did not attenuate anti-MPO NCGN. Combinatorial interventions may be necessary to avoid redundancy.

Keywords: ANCA, chemokines, crescentic glomerulonephritis, CXCR2, MPO

Introduction

Circulating anti-neutrophil cytoplasmic autoantibodies (ANCA) directed against myeloperoxidase (MPO) are associated with systemic small vessel vasculitis, often characterized by necrotizing crescentic glomerulonephritis (NCGN) [1]. In mice, administration of murine anti-MPO antibodies induces an acute glomerular inflammation that progresses to NCGN within days [2]. This process is aggravated severely upon co-administration of lipopolysaccharide (LPS) [3]. Pathologically, the model is characterized by an early glomerular accumulation of neutrophils that progresses to crescentic glomerulonephritis with abundant glomerular and interstitial macrophage infiltration. Moreover, neutrophils are the main effector cells in disease induction, as neutrophil depletion completely prevented disease development [4].

Recruitment of inflammatory cells to sites of inflammation is, to a large extent, regulated by chemokines. Chemokines are small chemotactic cytokines that are secreted by activated or injured cells and can be recognized by specific G-protein coupled receptors expressed on leucocytes [5,6]. Chemokines are classified into four families, -C, CC, CXC and CX₃C-, according to the position of the first two cysteines in the conserved amino acid sequence. Most chemokines belong to the CXC chemokine family, recruiting neutrophils and T and B cells, or the CC chemokine family, recruiting multiple leucocyte subsets including monocytes and T cells but not neutrophils. CXC chemokines containing an ELR+ (glutamic acid-leucine-arginine) motif are particularly powerful chemoattractants for neutrophils. The most potent ELR⁺CXC chemokine is interleukin (IL)-8 (CXCL8), which binds and activates its receptors CXCR1 and CXCR2 with similar affinity. CXCR1 is specific for IL-8, whereas other ELR+CXC chemokines (CXCL1, 2, 3, 5 and 7) can act through CXCR2. MPO-ANCA can activate neutrophils to produce IL-8 [7,8], and IL-8 is present in crescentic lesions of ANCA-associated NCGN patients [7], suggesting a pathogenic role for IL-8 in MPO-ANCA-associated vasculitis. Because a murine orthologue of human IL-8 does not exist, the activities of other ELR+CXC chemokines, e.g. keratinocyte-derived chemokine (KC/CXCL1) and macrophage inflammatory protein-2 (MIP-2/CXCL2), are more prominent in mice. CXCR2 is expressed predominantly on neutrophils, but can also be detected on monocytes/ macrophages and non-inflammatory cells, including microvascular endothelial cells [9-12].

Because of the diversity in chemokine receptors expressed by leucocytes, production of different chemokines during distinct phases of an inflammatory response determines the recruitment of specific leucocyte subsets in time and space. Spatiotemporal analysis of chemokine and chemokine receptor expression patterns during the course of glomerulonephritis may reveal potential targets for intervening in recruitment of specific leucocyte subsets. Chemokine and chemokine receptor expression patterns have been analysed in several glomerulonephritis models, such as nephrotoxic nephritis and immune complex glomerulonephritis (reviewed in [13] and [14]). No studies have been conducted, however, in experimental models of ANCAassociated vasculitis.

In this study, we analysed the renal gene expression levels of chemokines and chemokine receptors in the mouse model of anti-MPO-mediated NCGN, both in time and space, to identify potential targets for intervention. Based upon our initial results, we examined specifically the role of the chemokine receptor CXCR2 in experimental anti-MPO immunoglobulin (Ig)G-induced NCGN using a CXCR2blocking serum.

Materials and methods

Animals

Mpo^{-/-} mice were back-crossed to a C57BL/6 background seven times [15] and bred in-house. Female C57BL/6 wild-type mice were purchased from Harlan (Horst, the Netherlands). All animal experiments were performed according to national guidelines and upon approval of the Animal Care and Use Committee of Groningen University.

Production of polyclonal mouse anti-MPO IgG

Murine MPO was purified from WEHI-3 cells and used for immunization of $Mpo^{-/-}$ mice, as described previously [3]. Total IgG was isolated from pooled sera of immunized $Mpo^{-/-}$ mice and the anti-MPO titre was checked by enzyme-linked immunosorbent assay (ELISA), as reported previously [3].

Induction and evaluation of anti-MPO IgG-induced NCGN

Wild-type C57BL/6 mice (8-10 weeks) received 100 µg/g body weight of anti-MPO IgG intraperitoneally, followed by an intraperitoneal injection with 1500 EU/g (0.5 µg/g) LPS (Escherichia coli, serotype O26:B6; Sigma, St Louis, MO, USA) 1 h later. Mice were killed after 1 or 7 day(s), and kidneys were harvested, cut and partly snap-frozen for gene and protein analyses and partly embedded in paraffin for histopathological evaluation. Plasma and (17-h) urine were collected at both time-points. Urine samples were tested for haematuria (0-4+ score) by Combur-Test® strips (Roche Diagnostics BV, Almere, the Netherlands) and albuminuria by ELISA (Bethyl Laboratories, Montgomery, TX, USA). Periodic acid-Schiff staining was performed on paraffin sections and the number of glomerular crescents was counted in 100 consecutive glomerular cross-sections in a blinded fashion, as described previously [16]. Immunohistochemical staining for neutrophils was performed on acetone-fixed 5 µm cryosections using an anti-rabbit peroxidase-based Envision®+ system (DakoCytomation, Carpinteria, CA, USA), according to the manufacturer's protocol. Sections were incubated for 30 min with 10 µg/ml rat-anti-mouse-Ly6G (clone 1A8; BD Biosciences, Breda, the Netherlands) or isotype control antibody (IgG2a; Antigenix America, Huntington Station, NY, USA) followed by a 30-min incubation with 10 µg/ml unlabelled rabbit-anti-rat secondary antibody (Vector Laboratories, Burlingame, CA, USA). After detection of peroxidase activity with 3-amino-9-ethylcarbazole, sections were counterstained with Mayer's haematoxylin.

Chemokine expression analysis

For chemokine analysis, we studied kidneys from untreated mice (n = 12), mice subjected to LPS for 1 day (n = 9) and 7 days (n = 7) and mice subjected to anti-MPO IgG-induced NCGN for 1 day $(n = 11; 1.88 \pm 0.69$ neutrophils/glomerular cross-section; albuminuria $59.0 \pm 69.6 \ \mu g/16 \ h)$ and 7 days $(n = 14; 19.9 \pm 6.6\%$ crescents; albuminuria $1203 \pm 944 \ \mu g/16 \ h)$. For whole kidney gene expression analysis, RNA was isolated using the RNeasy mini kit (Qiagen Benelux BV, Venlo, the Netherlands) with DNase I treatment on the column. For analysis of microdissected material, $606 \ (range \ 410-873)$ glomeruli (equal to $2.72 \pm 0.29 \times 10^6 \ \mu m^2$) and surrounding tubulo-interstitial tissue $(2.88 \pm 0.25 \times 10^6 \ \mu m^2)$ were dis-

sected using the Laser Robot Microbeam System (PALM Micro Laser Technology, Bernried, Germany), as described previously [17], and RNA was isolated using the RNeasy micro kit (Qiagen). Reverse transcription was carried out using Superscript III reverse transcriptase (Invitrogen, Breda, the Netherlands) and random hexamer primers (Promega, Leiden, the Netherlands). Gene expression was analysed with a chemokine-focused 384-well micro fluidic card, containing primer-probe sets for 48 different genes (Table 1) using the ABI Prism 7900HT Sequence Detection System (Applied Biosystems, Nieuwerkerk a/d IJssel, the Netherlands). Relative mRNA levels were calculated as $2^{\text{-}\Delta CT}$, in which ΔCT is $CT_{gene of interest} - CT_{gapdh}$. CT-values that were beyond detection level were set manually to 40. Plasma levels of CXCL1 and CXCL2 protein were detected on a Luminex 100-based analyser (Luminex Corporation, Austin, TX, USA), using a Fluorokine Mouse MultiAnalyte Profiling Base Kit, containing antibody-coated microparticles and biotin-conjugated detection antibodies (R&D Systems Europe, Abingdon, UK). Protein levels of CXCL1, CXCL2 and CXCL5 in renal homogenates were determined with specific DuoSet ELISA kits (R&D Systems) and corrected for total protein concentration as measured with Bradford protein assay (Bio-Rad Laboratories, Veenendaal, the Netherlands).

Cell culture

Human conditionally immortalized glomerular endothelial cells (CiGEnC) [18] were cultured in endothelial growth medium 2-microvascular (EGM2-MV; Cambrex-Lonza, Breda, the Netherlands) containing fetal calf serum (5%) and growth factors as supplied, without vascular endothelial growth factor (VEGF). CiGEnC up to passage 40 were propagated at 33°C (when cells have a proliferative phenotype), whereas experiments were carried out after 5–7 days of incubation at 37°C (non-proliferative/quiescent phenotype).

Gene expression analysis of human glomerular endothelial cells and neutrophils

CiGEnC were seeded in 12-well plates (90 000 cells/well) and incubated at 33°C for 1 day before thermoswitching to 37°C. Neutrophils were isolated from heparinized venous blood of healthy donors by density gradient centrifugation on Lymphoprep (Axis-Shield, Oslo, Norway). Erythrocytes were lysed with ice-cold ammonium chloride buffer, and neutrophils were washed in Hanks's balanced salt solution without Ca^{2+}/Mg^{2+} (HBSS^{-/-}; Gibco/Life Technologies, Breda, the Netherlands). RNA was isolated using the RNeasy Plus Mini kit (Qiagen), and cDNA was prepared and individual gene real-time polymerase chain reaction (PCR) analyses were carried out as described in the 'Chemokine expression analysis' section. Primer-probe sets specific for human CXCR1 (Hs00174146_m1), CXCR2 (Hs00174304_m1) and glyceraldehyde-3-phosphate dehydrogenase (GAPDH) (Hs99999905_m1) (Applied Biosystems) were used.

Intracellular calcium measurements

For single-cell calcium imaging, CiGEnC were grown on glass coverslips (30 mm Ø) in six-well plates (60 000 cells/ well) at 33°C for 1 day before thermoswitching to 37°C. Cells were washed briefly in loading buffer (150 mM NaCl, 2 mM KCl, 2 mM CaCl₂, 1 mM MgCl₂, 10 mM HEPES/NaOH, 10 mM glucose, pH ~ 7.35) and loaded with 5 μ M Fura-2(AM) (TEF Laboratories, Austin, TX, USA) in loading buffer in the presence of 0.08% Pluronic® F-127 (Molecular Probes, Invitrogen) at 37°C for 30 min. After an additional washing step in loading buffer for 30 min in the presence or absence of 15 µg/ml anti-CXCR1 and/or anti-CXCR2 blocking antibodies (R&D systems), the coverslips were fixed in a perfusion chamber (37°C) and attached to an inverted microscope (Axiovert 35M; Zeiss, Sliedrecht, the Netherlands) equipped with a 12-bit Sensicam CCD camera (PCO, Kelheim, Germany) supported by Imaging Workbench 5.0 software (INDEC BioSystems, Santa Clara, CA, USA). Using a 16× plan-neofluar objective, digital images were taken at an emission wavelength of 510 nm using paired exposure at 340 nm and 380 nm excitation wavelengths at a frequency of 1Hz. Changes in intracellular calcium levels upon IL-8 treatment (100 ng/ml; R&D systems) were detected as changes in the ratio of the 340 and 380 nm excitation wavelengths in time (seconds).

In vivo anti-CXCR2 treatment

The inhibitory goat anti-murine CXCR2 serum was raised against a peptide (MGEFKVDKFNIEDFFSG) of the ligandbinding region of CXCR2. In previous studies, this antiserum (0·5–1·0 ml) has been shown to abrogate neutrophil influx in lung inflammatory mouse models without affecting circulating neutrophil numbers [19,20]. Mice (n = 6/group) received 0·8 ml anti-CXCR2 or normal goat serum (AbD Serotec, Düsseldorf, Germany) intraperitoneally every other day, starting with the first treatment 24 h before anti-MPO IgG administration. In a separate experiment, mice (n = 3/group) received a daily dose (30 mg/kg) of the CXCR2-inhibitor repertaxin (Sigma) subcutaneously, starting with the first treatment 1 h before anti-MPO IgG administration, and were killed after 7 days.

Statistical analysis

Statistical significance was determined using one-way analysis of variance (ANOVA) with Bonferroni's multiple comparison test (whole kidney mRNA analysis, intracellular calcium measurements and CXCL1/CXCL2/CXCL5 protein measurements) or two-tailed Student's *t*-test (mRNA analysis of dissected renal compartments and quantification of glomerular neutrophils).

B. S. van der Veen et al.

Table 1. List of the genes that were analysed using a custom low-density array.

CC CCL1 TCA-3 1-309 Mm0041226, ml Cd1 CCL2 IE MCP-1 Mm0041236, ml Cd3 CG13 MIP-16 MIP-16 Mm0041238, ml Cd3 CG17 CG17 MIP-16 Mm0041318, ml Cd5 CG27 MARC MCP-3 Mm0043112, ml Cd61 CG17 MARC MCP-3 Mm004318, ml Cd11 CGL11 Eotaxin Eotaxin Mm004128, ml Cd22 CCL2 MIP-3a Mm004128, ml Mm004128, ml Cd11 CGL11 Eotaxin Eotaxin Mm004128, ml Cd20 CCL2 ABCD-1 MDC Mm00428, ml Cd21 CCL2 ABCD-1 MDC Mm004382, ml Cd22 CCR2 ABCD-1 MDC Mm004382, ml Cd23 CCR4 Mm003820, ml Mm003820, ml Mm003820, ml Cd24 CCR4 Mm003820, ml Mm003820, ml Mm003820, ml Cd25 CCR4	Gene	Protein	Mouse synonym †	Human synonym †	Assay ID
Cd1 CC1 TCA3 1-399 Mn0041124_pn1 Cd2 IF MCP-1 Mn0041124_pn1 Cd3 CC13 MIP-1 α MIP-1 β Mn0041124_pn1 Cd4 CC13 MIP-1 α MIP-1 β Mn0041125, pn1 Cd5 CC15 RANTES RANTES Mn0041123, pn1 Cd61 CC15 RANTES RANTES Mn0041228, pn1 Cd7 CC17 MARC Mn0041228, pn1 CC17 Cd11 CC11 CC17 TARC Mn0041228, pn1 Cd17 CC17 TARC Mn0041228, pn1 CC122 Cd17 CC17 TARC Mn0041228, pn1 CC120 Cd17 CC17 TARC Mn0041228, pn1 CC122 MDC Mn0041228, pn1 Cd2 CC12 CC12 MP- α Mn0041228, pn1 CC12 MN0041228, pn1 Cd2 CC12 CC12 MP- α Mn0041228, pn1 CC12 Mn0041228, pn1 Cd2 CC12 MP- α Mn0043619, pn1 Mn0043619, pn1 Mn0043619, pn1 CC42 Mn00126171, pn1 <td>CC chemokines and receptors</td> <td></td> <td></td> <td></td> <td></td>	CC chemokines and receptors				
Cd2CC12FMCP-1Mn004128, m1Cd3CC13MIP-1αMIP-1αMn004128, m1Cd4CC4MIP-1βMIP-1βMn004131. m1Cd5CC15RANTESMn004311. m1Cd7CC17MARCMCP-2Mm0043113, m1Cd8CC18MCP-2Mm0043128, m1Cd17CC11EotainEotainMm0044128, m1Cd17CC12ABCD-1MDCMm0044128, m1Cd20CC12ABCD-1MDCMm004428, m1Cd21CC12ABCD-1MDCMm004428, m1Cd2CC12CC12ABCD-1MDCMm004428, m1Cd2CC12CC13MIP-3αMIP-3αMm004428, m1Cd2CC12CC14MID-3aMID-3aMID-3aCd7CC23CC12MDCMm0035827, m1MID-3aCd7CC24CC23MID-3aMID-3aMID-3aCd7CC24CC24MID-2GR0βMm0034869, m1Cd2CCC4CXC1KCGR0βMm0034869, m1Cd2CXC1MID-2GR0βMm0034869, m1Cd1CXC1KC1KC1MID-3aMn0034869, m1Cd2CXC13MID-2GR0βMm00343859, m1Cd2CXC14FF4Mm00343859, m1Cd2CXC13MIRMm00432859, m1Cd12CXC13MIRMm0043529, m1Cd212CXC13MIRMm0043529, m1Cd213C	Ccl1	CCL1	TCA-3	I-309	Mm00441236 m1
Cd3CC13MIP-1aMIP-1aMIP-1bMIM0401258_m1Cd4CC14MIP-1bMIM041258_m1MID041258_m1Cd7CG17MARCMCP-3Mm0041258_m1Cd7CC17MARCMCP-2MCP-3Mm0041258_m1Cd17CG17CG17MARCMIP-3aMm0041258_m1Cd11CC111ExarinExarinMm0041258_m1Cd17CG2MCP-2MCP-3Mm0041258_m1Cd12CC117TARCTARCMm0041258_m1Mm0041258_m1Mm0041258_m1Mm0044228_m1Cd22CC122ABCD-1MIP-3aMIP-3aMm0041258_m1Mm0044228_m1Cd2CC12CC2ABCD-1MID-3Mm0044228_m1Mm0044228_m1Cd7CCR1CCR1MID003871_m1Mm003871_m1Mm0038721_m1Mm0038721_m1Cd7CCR3CCR4Mm003871_m1Mm0038721_m1Mm0038721_m1Mm0038721_m1Cd7CCR4CCR4MM003871_m1Mm0038721_m1Mm0038721_m1Mm0038721_m1Cd7CCR5CCR4Mm003871_m1Mm003885_m1Mm0034946_m1Cd7CCR4CCR4PF4PF4Mm003895_m1Cd2CXC15LIXEN-78Mm0034946_m1Cd2CXC10IP10IP10Mm0044238_m1Cd2CXC10IP10IP10Mm0034946_m1Cd11CXC11IP10IP10Mm0034946_m1Cd12CXC12SDF-1SDF-1Mm0034946_m1Cd12CXC11IP10	Ccl2	CCL2	JE	MCP-1	Mm00441242_m1
Ccl4CL4MIP-1βMIP-1βMIP-1βMIP-00Cd5CL5RANTESRANTESMO0130248.mlCd7CCL7MARCMCP-3MI004311.s.mlCd8CCL8MCP-2MCP-3MID013718.s.mlCd11EotaxinEotaxinMID013718.s.mlCd12CCL11EotaxinMID0441128.mlCd12CCL12MIP-3αMIP-3αCd22CCL2ABCD-1MDCCd13CCR1MID043012.s.mlCd14CCR1MID043012.s.mlCd2CCR2MIP-3αMID033620.s1Cd7CCR3MID043012.s.mlMID0033573.s1Cd7CCR4MID0033573.s.mlMID0033573.s1Cd7CCR5MID0033573.s.mlMID0033573.s1Cd7CCR5MID0033573.s1MID0033573.s1Cd7CCR4MIP-2GGRO2MID043085.p.mlCd7CCR5MID0033573.s1MID0033573.s1Cd7CCR5MID0033573.s1MID0033573.s1Cd7CCR5MID033665.p.mlMID033655.g1Cx41CXC1MIP-2GGRO2MID033655.g1Cx42CXC1MIP-2GGRO3MID033655.g1Cx41CXC1MIP-2GGRO3MID033655.g1Cx41CXC1MIP-2GGRO3MID033655.g1Cx42CXC1MIP-2GGRO3MID033655.g1Cx43CXC1MIP-2GMID03395.g1Cx41CXC1MID04392.g1MID03395.g1Cx41CXC1	Ccl3	CCL3	MIP-1a	MIP-1a	Mm00441258 m1
Cd5CL5RANTESRANTESMm010212E_m1Cd7CC17MARCMCP-3Mm0041138_m1Cd8CC18MCP-2MCP-2Mm01927183_m1Cd11CC111ExtaxinExtaxinMm0041128_m1Cd12CC12MIP-3αMIP-3αMm004128_m1Cd22CC12ABCD-1Mm004128_m1Mm004128_m1Cd2CC12ABCD-1Mm004128_m1Mm004428_m1Cd2CC12ABCD-1Mm004350_m1Mm0043520_m1Cd2CCR3Mm0043521_m1Mm0043521_m1Cc73CCR4Mm0043521_m1Mm0043521_m1Cc74CCR5CCR4Mm0043521_m1Cc75CCR5Mm0043521_m1Cc76CCR6Mm0043521_m1Cc78CCR4Mm0043521_m1Cc78CCR4Mm0043521_m1Cc78CCR5Mm0043521_m1Cc79CCR5Mm0043521_m1Cc79CCR6Mm0043525_m1Cx21CXC1KCCx21CXC1KCCx21CXC1Mm004352_m1Cx21CXC1KCCx21CXC1Cx21CXC1Cx21CXC1Cx21CXC1Cx21CXC1Cx21CXC1Cx21CXC1Cx21CXC1Cx21CXC1Cx21CXC1Cx21CXC1Cx21CXC1Cx21CXC1Cx21CXC1Cx21CXC1Cx21	Ccl4	CCL4	MIP-1B	MIP-1B	Mm00443111 m1
Cd7CL7MARCMCP.3Mm0043115_m1Cd8CCB8MCP.2MCP.2Mm0127183_m1Cd11CCL11ForaxinEotxinMm0041128_m1Cd12CCL17TARCTARCMm0031016_m1Cd20CCL20MFP.3cMm0044223_m1Cd21CCL2ABCD-1MDCMm0043649_m1Cd22CCL2ABCD-1MDCMm0043642_m1Cd2CCR2CCR2Mm0034642_m1Mm0034642_m1Cd7CCR3CCR3Mm0034642_m1Cd7CCR4Mm003457_m1Mm003457_m1Ccr5CCR5Mm003457_m1Mm003457_m1Ccr6CCR6Mm003457_m1Mm003457_m1Ccr6CCR2Mm003464_m1Mm003455_m1Ccr6CCR2Mm0034645_m1Mm003454_m1Ccr6CCR2Mm0034645_m1Mm0034645_m1Ccd1CXCL2MFP-2GROftMm003455_m1Ccd2CXCL3LTXENA-78Mm0034654_m1Pi4CXCL10IP-10IP-10Mm003457_m1Ccd10CXCL10IP-10IP-10Mm003452_m1Ccd11CXCL11ITACITACMm003452_m1Ccd12CXCL3BLCBCA-1Mm003452_m1Ccd13CXCR1Mm003452_m1Mm003452_m1Ccc73CXCR3CXCR3Mm003452_m1Ccc74CXCR4FacalkineMm003452_m1Ccc75CXCR4Mm003452_m1Mm003452_m1Ccc76CXCR4Facalkine<	Ccl5	CCL5	RANTES	RANTES	Mm01302428 m1
Cd8CC18MCP-2MCP-2MP013718_m1Cd11CCL11FatarinEntarinMm014128, m1Cd17CACTARCTARCMm0316136_m1Cd20CCL20MIP-3αMIP-3αMm0044128, m1Cd12CCL2ABCD-1MIP-3αMm0043820_m1Cr1CCR1KMm0038271_m1Cr2CCR3Mm00438271_m1Cr3CCR4Mm00138271_m1Cr6CCR3Mm00138271_m1Cr6CCR3Mm00138271_m1Cr6CCR4Mm00138271_m1Cr6CCR5Mm00138271_m1Cr6CCR4Mm00138571_m1Cr6CCR5Mm00136352_m1Cr6CCR4Mm00136452_m1Cr6CCR5Mm00136452_m1Cr6CCR4Mm00136452_m1Cr6CCR5Mm00136452_m1Cr61CXCL1MIP-2Cr63CXC14PF-4Pf4CXC14PF-4Pf4Mm00136452_m1Cr40CXC10IP-10Pf4CXC11IFACCr41CXC11IP-10Cr41CXC11IP-10Cr41CXC11IP-10Mm0043525Mm0043552_m1Cr41CXC12SDF-1Cr41CXC13Cr41CXC14Cr41CXC14Cr41CXC14Cr41CXC14Cr41CXC13Cr41CXC14Cr41CXC14Cr41CXC14Cr41 </td <td>Ccl7</td> <td>CCL7</td> <td>MARC</td> <td>MCP-3</td> <td>Mm00443113 m1</td>	Ccl7	CCL7	MARC	MCP-3	Mm00443113 m1
Cd11Cd11FaranFaranMan0041235_m1Cd17CCL17TARCTARCMan051616_n1Cd20CL20MIP-3aMIP-3aMIP-3aGd22CCL22ABCD-1MDCMm0043639_m1Car1CCR1MTCMm0043639_m1Mm0043639_m1Car2CCR2MID-3aMm0043639_m1Car3CCR3Mm0043821_m1Mm0043826_s1Car4CCR4Mm0043827_m1Mm0043827_m1Car5CCR6Mm0043827_m1Mm0043827_m1Car6CCR6Mm0043827_m1Mm0043827_m1Car6CCR6Mm0043859_m1Mm0043649_m1Car6CCR6Mm0043649_m1Mm0043649_m1Cacl2CCR2MFP-2GRO3Mm0043649_m1Cacl3CXCL4MFP-2GRO3Mm0043649_m1Cacl4CXCL4PF-4Mm00436451_g1Cacl5CXCL5LIXENA-78Mm00436451_g1Cacl9CXCL9MfgMm0043651_g1Cacl10CXCL10IP-10IP-10Mm0043552_m1Cacl11CXCL12SDF-1SDF-1Mm0043552_m1Cacl12CXCL13BLCBC-1Mm0043555_m1Cacl13CXCR1Mm0043528_m1Mm0043829_m1Cacl2CXCR4Mm0043829_m1Mm0043829_m1Cacl3CXCR4Mm0043829_m1Cacl4CXCR4Mm0043829_m1Cacl5CXCR4Mm0043829_m1Cacl5CXCR4Mm0043829_m1Cacl6CXCR4 <t< td=""><td>Ccl8</td><td>CCL8</td><td>MCP-2</td><td>MCP-2</td><td>Mm01297183 m1</td></t<>	Ccl8	CCL8	MCP-2	MCP-2	Mm01297183 m1
Cd17TARCTARCMRCMm0041613_m1Cd20CCL20MIP-3αMIP-3αMm00436439_m1Cd22CCL2ABCD-1Mm00436439_m1Cd2CCR1Mm00436439_m1Cd2CCR2MR00436430_m1Cd2CCR3Mm004364321_m1Ccr4CCR4Mm004364321_m1Ccr5CCR5Mm00131671_m1Ccr6CCR6Mm00131671_m1Ccr6CCR6Mm00436459_m1Ccr6CCR6Mm00436459_m1Ccc8CCR6Mm00436459_m1Ccc4CCR1KCCcd1CCR6Mm00436459_m1Ccc4CCR1MIP-2Ccd1CCR2MIP-3Ccd2CXCL4MP-4Pf-4Pf-4Mf-6Ccd5LIXENA-78M0043645Mm0043645Ccd10CXCL9MigCcd11CXCL10IP-10Ccd12CXCL1MfP-10Ccd13CXCL13BLCCcd13CXCL1Mm0044552_m1Ccd13CXCL3Mm0043208_m1Ccc73CXCL4Mm0043208_m1Ccc74CXCR4Mm0043208_m1Ccc75CXCL5Mm0043208_m1Ccc76CXCL1Mm0043208_m1Ccc77CXCL1Mm0043208_m1Ccc78CXCL1Mm0043208_m1Ccc79CXCL1Mm0043208_m1Ccc79CXCL1Mm0043208_m1Ccc79CXCL1Mm0043208_m1Ccc79CXCL1Mm0043208_m1 <td>Ccl11</td> <td>CCL11</td> <td>Eotaxin</td> <td>Eotaxin</td> <td>Mm00441238 m1</td>	Ccl11	CCL11	Eotaxin	Eotaxin	Mm00441238 m1
Cd20Cd20MP-3αMP-3αMm0044422a_m1Cd22CC122ABCD-1MDCMm00436439_m1Cr1CCR1MDCMm00436439_m1Cr2CCR2Mm003580_s1Cr3CCR3Mm0043827_m1Cr4CCR4Mm0043827_m1Cr5CCR6Mm0043827_m1Cr6CCR6Mm0043827_m1Cr6CCR6Mm0043827_m1Cr6CCR6Mm0043859_m1Cr6CCR6Mm0043859_m1Cr6CCR6Mm0043859_m1Cr2CR6Mm0043859_m1Cr2CR1MF-2GR0βMm0043645PF-4PF-4Cr20CCL1MF-2Mm0043645Cr21CCL1ITP-2Mm0043645Cr21CCL1ITP-2Mm0043645Cr21CCL1ITP-10IP-10Cr21CCL1SDF-1SDF-1Cr21CCCL3SDF-1SDF-1Cr21CCCR3Wm0043455Cr23CCCR3Wm0043825Cr24CCCR4Mm0043825Cr31CCCR5Wm0043825Cr31CCCR4Mm0043825Cr31CCCR5Mm0043825Cr31CCCR3Wm004372Cr31CCCR3Wm004372Cr31CCCR4Mm004372Cr31CCCR5Mm004372Cr31CCCR4Mm004372Cr31CCCR5Mm004372Cr31CCCR5Mm004372Cr31CCCR5Mm004372	Ccl17	CCL17	TARC	TARC	Mm00516136 m1
Cd22Cd22ABCD-1MDCMm004383_m1Ccr1CCR1Mm0043820_s1Ccr2CCR2Mm0051554_s1Ccr3CCR3Mm0015851_s1Ccr4CCR4Mm0015827_m1Ccr5CCR5Mm0015151_s1Cr6CCR6Mm0013852_m1Cr6CCR6Mm0013852_m1Cr6CCR6Mm0013852_m1Cr6CCR1KCGR06Cr8CCR2Mm0043852_m1Cx6CXCL1MFP-4PF-4Pf4CXCL2MIP-2GR06Mm0043651_g1CXCL5LIXENA-78Cxd10CXCL10IP-10IP-10Mm0043645_1g1Cxd10CXCL10IP-10IP-10Mm00436451_g1Cxd11CXCL11I-TACMm00436451_g1Cxd12CXCL13BLCBCA-1Mm00443525_m1Cxd13CXCL13BLCBCA-1Mm0043825_m1Cxc73CXCR2Mm0043825_m1Mm0043825_m1Cxc74CXCR4Mm0043825_m1Cxc75CXCR5Mm0043825_m1Cxc74CXCR1NumphotactinMm0043854_m1Cxc74CXCR1NumphotactinMm0043854_m1Cxc75CXCR2Mm00436451_m1Cxc76CXCR3Mm0043654_m1Cxc77CXCR4Mm0043654_m1Cxc78CXCR2Mm0043654_m1Cxc79CXCR1NumphotactinCxc70CXCR2Mm0043655_m1Cxc71CXCR4Mm0043655_m1Cxc71	Ccl20	CCL20	MIP-3α	MIP-3a	Mm00444228 m1
Cr1CR1Mm0943820_11Cr2CR2Mm0999051_gHCr3CR3Mm001554.31Cr4CCR4Mm001554.31Cr5CCR5Mm001357_m1Cr6CCR6Mm09999115_s1Cr7CCR6CCR8Cr8CR8Mm09999115_s1CXC chemokines and receptorsKCGR0\alpha Mm0043859_m1Cxc1CXCL1KCGR0\alpha Mm0043859_m1Cxc2CXCL2MIP-2GR0\alpha Mm0043859_m1Cxc2CXCL4PF-4PF-4Mm0045135_g1Cxc3CXCL4PF-4Mm0043946[m1]Cxc4CXCL10IP-10IP-10Mm0043945[m1]Cxc10CXCL10IP-10Mm0043945[m1]Cxc11CXCL10IP-10Mm0043945[m1]Cxc11CXCL12SDF-1SDF-1Mm0043945[m1]Cxc11CXCL13BLCBC-1Mm0043325[m1]Cxc13CXCR1Mm0043325[m1]Cxc13Mm0043325[m1]Cxc3CXCR2Mm0043325[m1]Cxc3Mm0043325[m1]Cxc3CXCR4Mm004320[m2]Mm004320[m1]Cxc1CXCR4Mm004320[m1]Mm004320[m2]Cxc1CXC11LymphatactinMm004347[m1]Cxc1CXCR4Mm004320[m2]Mm004320[m2]Cxc3CXCR4Mm004320[m2]Mm004320[m2]Cxc3CXC11LymphatactinMm004320[m2]Cxc3CXCR4Mm004320[m2]Mm004320[m2]Cxc3CXCR4Mm004320[m2]Mm004320[m2	Ccl22	CCL22	ABCD-1	MDC	Mm00436439 m1
Cr2CCR2Mm999905_dflCr3CCR3Mm0051354_slCr4CCR4Mm00128271_m1Cr5CCR5Mm9999114_slCr6CCR6Mm99999115_slCr6CCR8Mm00438851_m1CxC1CXCMm00438851_m1Cxc1CXC12MIP-2GR00Mm0043855Mm0043655_m1Cxc2CXC12MIP-3GR06Mm004385CXC15LIXENA-78Pf4CXC10IP-10IP-10Mm0043651_g1Cxc10CXC10IP-10P1-10Mm0043651_m1Cxc11CXC11ITACI-TACMm0043651_m1Cxc13CXC110IP-10IP-10Mm0043985_m1Cxc110CXC1111-TACI-TACMm0043532_m1Cxc111CXC113BLCBCA-1Mm0043532_m1Cxc13CXC13BLCMm0043532_m1Cxcr3CXCR12Mm0043625_m1Mm0043625_m1Cxcr4CXCR2Mm0043625_m1Mm0043625_m1Cxcr5CXCR2Mm0043625_m1Mm0043625_m1Cxcr4CXCR1NeurotactinImm0043625_m1Cxcr4CXCR1NeurotactinMm0043654_m1Cxcr5CXC13NeurotactinMm0043625_m1Cxcr4CXCR2Mm0043625_m1Mm0043625_m1Cxcr5CXCR1NeurotactinMm0043625_m1Cxcr6CXCR2Mm0043625_m1Mm0043625_m1Cxcr6CXCR2Mm0043625_m1Cxcr6CXCR2Mm0043625_m1<	Ccr1	CCR1			Mm00438260 s1
Cr3 CCR3 Mm00515543_m Cr4 CCR4 Mm0043821_m1 Cr6 CCR6 Mm9999114_s1 Cr76 CCR6 Mm9999115_s1 CXC chemokines and receptors Mm00433859_m1 CxC1 KC GR0α Mm00433859_m1 CxC1 MC GR0α Mm00433859_m1 CxC2 MP-2 GR0β Mm0043315_g1 CxC1 KC GR0β Mm00434946_m1 Cxc10 CXC14 PF.4 Mm00436131_g1 Cxc13 CXC19 Mig Mm00436131_g1 Cxc14 CXC19 Mig Mm004364462_m1 Cxc10 CXC11 I-TAC Mm0044523_sm1 Cxc11 CXC11 I-TAC Mm0044533_m1 Cxc11 CXC11 Mm0044533_m1 Mm0044533_m1 Cxc11 CXC1 Mm0044533_m1 Mm00433359_m1 Cxc11 CXC1 Mm00433359_m1 Mm00433359_m1 Cxc3 CXC2 Mm00433325_m1 Cxc4	Ccr2	CCR2			Mm99999051 gH
Cr4CCR4Mm0043827]m1Cr5CCR5Mm0121671_m1Cr6CCR6Mm09999114_s1Cr7CCR5Mm09999115_s1CXCCCR8Mm09999115_s1CXCCCR1KCGR0 α Mm00433859_m1CxC12MIP-2GR0 β Cxd1CXC14PF-4PF-4P4CXC14PF-4PF-4Cxd5CXC15L1XENA-78Cxd10CXC10IP-10IP-10Cxd11CXC110IP-10Mm00434552_m1Cxd12CXC10IP-10IP-10Cxd13CXC112SDF-1SDF-10Cxd13CXC112SDF-1Mm00433259_m1Cxd13CXC13BLCBCA-1Mm0043325_m1Mm0043325_m1Cxcr3CXCR4Mm00433259_m1Cxcr4CXCR4Mm00433259_m1Cxcr5CXCR4Mm00433259_m1Cxcr4CXCR4Mm00436454_m1Cxcr5CXCR4Mm00436454_m1Cxcr4CXCR1Mm00436454_m1Cxc74CXCR4Mm00436454_m1Cxc74CXCR1Mm00436454_m1Cxc74CXCR1Mm0043625_m1Xcl1Xcl1IymphotactinMm0043727_m1Xcl1Mm0043625_m1Xcl1CX1IymphotactinMm0043725Mm0043725_m1Xcl1CX1IymphotactinMm004372Mm0043725_m1Xcl1Mm004372Xcl1Mm004372Mm004372Mm0043725_m1<	Ccr3	CCR3			Mm00515543 s1
Cr5CCR5Mm01216171_m1Cr6CCR6Mm9999115_s1Cr7CCR6Mm99999115_s1CXC chemokines and receptorsCXC1KCCxd1CXC12MIP-2GR0 α Mm00436450_m1CXC12MIP-2GR0 β Mm00436450_m1CXC12MIP-2GR0 β Pf4CXC12MIP-2GR0 α Cxd5CXC15LIXENA-78Cxd10CXC19MigMm0043645_g1Cxd10CXC19MigMm0043645_g1Cxd11CXC111I-TACI-TACCxd11CXC112SDF-1SDF-1Cxd13CXC13BLCBCA-1Cxd13CXCR1Mm0043255_m1Cxc73CXCR2Mm0043255_m1Cxc74CXCR4Mm0043255_m1Cxc74CXCR4Mm0043255_m1Cxc74CXCR4Mm0043255_m1Cxc74CXCR4Mm0043255_m1Cxc74CXCR4Mm0043255_m1Cxc74CXCR4Mm0043265_m1Cxc74CXCR4Mm0043265_m1Cxc74CXCR1Mm0043265_m1Cxc74CXCR1Mm0043265_m1Cxc74CXCR1Mm0043265_m1Cxc74CXCR1Mm0043265_m1Cxc74CXCR1Mm0043265_m1Cxc74CXCR1Mm0043265_m1Cxc74CXCR1Mm0043265_m1Cxc74CXCR1Mm0043265_m1Cxc74CXCR1Mm0043265_m1Cxc74CXCR1Mm0044265_m1Cxc74C	Ccr4	CCR4			Mm00438271 m1
CréCCR6Mm99999114_s1Cr6CCR8Mm99999115_s1CXC chemokines and receptorsMm00433859_m1Cxcl2CXC.1KCGRO α Mm00433859_m1Cxcl2CXC.12MIP-2GRO β Mm00433859_m1Cxcl3CXC.14PF-4PF-4Mm0043315_g1Cxcl4PF-4PF-4Mm00436451_g1GROCxcl5LIXENA-78Mm00436451_g1Cxcl0CXC.10IP-10IP-10Mm00446252_m1Cxcl10CXC.11I-TACI-M0044525_m1Cxcl11CXC.12SDF-1SDF-1Mm0044552_m1Cxcl12CXC.13BLCBCA-1Mm0044532_m1Cxcl3CXC.R3Mm00438259_m1Cxcr3CXC.R4Mm00438259_m1Cxcr4CXC.R4Mm00436254_m1Cxcr5CXC.R5Mm00438254_m1Xcl1XCl1IymphotactinMm00436354_m1Xcr1CXC.R4Mm00436266_m1Cxcr4CXC.R1Mm00436354_m1Xcr4CXC.R1Mm00436354_m1Xcr5CXC.R1Mm00436354_m1Xcr4MM0043206_m1Mm00436354_m1Xcr4CXG.R1Mm00436354_m1Xcr4CXG.R1Mm00436354_m1Xcr4CXG.R1Mm0043206_m1Xcr5CXG.R1Mm0043206_m1Xcr4Mm0043206_m1Xcr5Mm0043206_m1Xcr5Mm0043206_m1Xcr4Mm0043206_m1Xcr4Mm0043206_m1Xcr5 <t< td=""><td>Ccr5</td><td>CCR5</td><td></td><td></td><td>Mm01216171 m1</td></t<>	Ccr5	CCR5			Mm01216171 m1
Ccr8CCR8Mm99999115_s1CXC hemokines and receptorsCxcl1CXCL2MCCL2CXCL2Cxcl2CXCL2CXCL3MIP-2GROGMm00436450_m1Pf4CXCL4PF-4Cxcl5LIXEX09CXCL9MigMigMm00436451_g1Cxcl0CXCL9Cxcl10CXCL10Cxcl11F1ACCxcl12CXCL12Cxcl13BLCCxcl13CXCL12Cxcl13BLCCxcl13CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4Cxcr4CXCR4CXcr11ImphotactinCxcr5CXCR4CXcr11Mm0043825_m1Cxc73CXCR4CXCR4Mm0043825_m1CXc74CXCR4CXCR4Mm0043825_m1CXc74CXCR4CXCR4Mm0043825_m1CXC74CXCR4CXC74Mm0043825_m1CXC74CXCR4CXC74Mm0043825_m1CXC75Mm0043825_m1C	Ccr6	CCR6			Mm999999114_s1
CXC chemokines and receptors KC GROα Mm00433859_m1 Cxcl1 CXCL1 KC GROα Mm00433859_m1 Cxcl2 CXCL4 PF-4 PF-4 Mm00436450_m1 Pf4 CXCL4 PF-4 PF-4 Mm00436451_g1 Cxcl5 L1X ENA-78 Mm00436451_g1 Cxcl9 CXCL0 IP10 IP-10 Mm00436452_m1 Cxcl10 CXCL10 IP-10 IP-10 Mm0043652_m1 Cxcl11 CXCL12 SDF-1 SDF-1 Mm0044552_m1 Cxcl13 CXCL12 SDF-1 SDF-1 Mm00438259_m1 Cxcl13 CXCR1 Mm00438259_m1 Mm00438259_m1 Cxcr3 CXCR1 Mm00438259_m1 Mm00438259_m1 Cxcr3 CXCR2 Mm00438259_m1 Mm00438259_m1 Cxcr4 CXCR4 Mm00436454_m1 Mm00436454_m1 Cxr4 CXCR1 Mm00436454_m1 Mm00436454_m1 Cxf2 CACR1 Mm00436454_m1 Mm00436454_m1 Cxf2	Ccr8	CCR8			Mm99999115_s1
Circl influenties and receptors CXCL1 KC GROα Mm00433859_ml Cxcl2 CXCL2 MIP-2 GROβ Mm00433859_ml Cxcl2 CXCL4 PF-4 PF-4 Mm00433151_gl Cxcl5 CXCL5 LIX ENA-78 Mm00436451_gl Cxcl9 Mig Mig Mig Mm0043946_ml Cxcl10 IP-10 IP-10 Mm0044525_ml Cxcl11 CXCL10 IP-10 Mm0044525_ml Cxcl12 CXCL13 BLC BCA-1 Mm0044532_ml Cxcl13 CXCL13 BLC BCA-1 Mm0043825_ml Cxr3 CXCR1 Mm0043825_ml Mm0043825_ml Mm0043825_ml Cxr4 CXCR4 Xm0043825_ml Mm0043825_ml Mm0043825_ml Cxr4 CXCR4 Xm0043825_ml Mm0043825_ml Mm0043825_ml Cxr4 CXCR4 Mm0043825_ml Mm0043825_ml Mm0043825_ml Cxr4 CXCR4 Mm00434772_ml Mm00434772_ml Mm00434772_ml Cxr4 <td>CYC chemokines and recentors</td> <td></td> <td></td> <td></td> <td></td>	CYC chemokines and recentors				
CxtlCxtlMD2GR0AMm00436450_m1Pf4CXCL2MIP-2GR0BMm00436450_m1Cxd2CXCL5LIXENA-78Mm00436451_g1Cxd9CXCL9MigMigMm00436452_m1Cxd10CXCL9MigMigMm00434946_m1Cxd11CXCL10IP-10IP-10Mm00445252_m1Cxd12CXCL11I-TACI-TACMm00445325_m1Cxd13CXCL12SDF-1SDF-1Mm00445325_m1Cxd13CXCR1Mm0043525_m1Mm0043525_m1Cxr3CXCR2Mm00438258_m1Cxr4CXCR4Mm00438259_m1Cxr5CXCR5Mm0043825_m1Cxr4CXCR1Mm00438259_m1Cxr5CXCR3Mm00438259_m1Cxr4CXCR4Mm0043825_m1Cxr4CXCR1Mm00438259_m1Cxr4CXCR1Mm00438259_m1Cxr4CXCR1Mm00434772_m1Xcr1CXCR1Mm00434772_m1Xcr1XCR1Mm00434772_m1Xcr1XCR1Mm00434772_m1Xcr1CD31Mm00434772_m1SeleE-selectinMm00434772_m1AngtP1Angiopeitin 1Mm00434232_m1AngtP2Angiopeitin 2Mm00434232_m1AngtP3SelectinMm00434232_m1AngtP4I88Hs9999901_s1Nphs1NephrinMm0043755_m1Ang2Aquaporin 2Mm0043755_m1App2MegainMm0132817_m1	Cycl1	CXCI 1	KC	GROG	Mm00433859 m1
CkriftCkriftMit 2OrdpMino 450102, minor 20,	Cycl2	CXCL2	MIP_2	GROß	Mm00436450 m1
I.P.CACLYI.P.I.P.Min0031512_g1CxdbCXCL5LIXENA-78Mm00436451_g1Cxdl0CXCL9MigMigMm0043946_m1Cxdl1CXCL10IP-10IP-10Mm00445235_m1Cxdl12CXCL11I-TACI-TACMm0044552_m1Cxdl3CXCL12SDF-1SDF-1Mm00445552_m1Cxd13BLCBCA-1Mm0044535_m1IBrbCXCR1Mm004352_m1Cxr3CXCR3Mm0043525_m1Cxr4CXCR4Mm0043525_m1Cxr5CXCR5Mm00436454_m1Cxr61CXCR1NeurotactinCxr4CXCR4Mm00436454_m1Cxr5CXCR5Mm00436454_m1Cxr1CXCR1NeurotactinCxr4CXCR1Mm00436454_m1Cxr5CXCR1Mm00436454_m1Cxr1CX,CR1NeurotactinCxr1CXCR1Mm00436454_m1Cxr2Mm00436454_m1Cxr4CXCR1Mm00436454_m1Cxr4CXCR1Mm00436454_m1Cxr1CXR1Mm00436454_m1Cxr1Mm00436454_m1Cxr2Mm00436454_m1Cxr4CXCR1Mm00436454_m1Cxr4CXCR1Mm00436454_m1Cxr4CXCR1Mm00436454_m1Cxr4Mm00436454_m1Mm00436454_m1Cxr4CXCR1Mm00436454_m1Cxr4Mm00436454_m1Cxr5Mm0043656_m1Mm0044206_m1 <td>Df4</td> <td>CYCL4</td> <td>PE_{-4}</td> <td>PE-4</td> <td>Mm00451315_g1</td>	Df4	CYCL4	PE_{-4}	PE-4	Mm00451315_g1
CxdbCxdbLinLinLinInit 10Init 10000112, generationCxdbCxdl0IP-10IP-10Mm00434235, m1Cxd11CXCL11I-TACI-TACMm00443552, m1Cxd12CXCL12SDF-1SDF-1Mm00445552, m1Cxd13CXCL13BLCBCA-1Mm00443552, m1IBraCXCR1ImmonostrateMm00438258, m1Cxr3CXCR3ImmonostrateMm00438259, m1Cxr4CXCR4ImmonostrateMm00438259, m1Cxr5CXCR5ImmonostrateMm00438259, m1Cxr4CXCR4ImmonostrateMm00438259, m1Cxr5CXCR5ImmonostrateMm00436454, m1Cx3c1CXCR5ImmonostrateMm00436454, m1Cx3c1CXCR1ImmonostrateImmonostrateCx3c1CXCR1ImmonostrateImmonostrateCx4CXCR1ImmonostrateImmonostrateCx4CXCR5ImmonostrateImmonostrateCx4CXCR1ImmonostrateImmonostrateCx4CXCR1ImmonostrateImmonostrateCx4CXCR1ImmonostrateImmonostrateCx4CXCR5ImmonostrateImmonostrateCx4CXCR1ImmonostrateImmonostrateCx4CXCR1ImmonostrateImmonostrateCx5CXCR5ImmonostrateImmonostrateCx6CXCR1ImmonostrateImmonostrateCx6CXCR1ImmonostrateImmonostrate <td>Cycl5</td> <td>CXCL5</td> <td>I I X</td> <td>FNA-78</td> <td>Mm00436451_g1</td>	Cycl5	CXCL5	I I X	FNA-78	Mm00436451_g1
CXCDAngAngAngAngAngAngAngAngAnd Madde Ma	Cyclo	CYCL9	Mig	Mig	Mm00434946 m1
CxrliCXCL10F1 TACF1 TACH1 M0044352_m1Cxcl11CXCL12SDF-1SDF-1Mm0044662_m1Cxcl3CXCL13BLCBCA-1Mm0044353_m1Il8raCXCR1Mm0073132_g.1Mm00438258_m1Cxcr3CXCR3Mm00438259_m1Mm00438258_m1Cxcr4CXCR4Mm00438259_m1Cxcr5CXCR5Mm00438258_m1Cxcr4CXCR4Mm00438258_m1Cxcr5CXCR5Mm00438258_m1Cxcr4CXCR4Mm00438354_m1Cxf2 and C chemokines and receptorsKCR1Mm00438354_m1Cxf2 and C chemokines and receptorsKCR1Mm00438354_m1Xcl1CX_CR1Mm00438354_m1Xcl1Xcr1XCL1LymphotactinLymphotactinXcr1CXCR1Mm0043672_m1Xcr1XCR1Mm0043672_m1SelpP-selectinMm0041278_m1Angpt1Angiopoietin 1Mm0043632_m1Angpt1Angiopoietin 2Mm0043242_m1Housekeeping and otherIte-2Mm0043242_m1Mps1NephrinMm004372575_m1Nphs2PodocinMm00437575_m1Aqp2Aquaporin 2Mm00437575_m1Lrp2MegalinMm0032817_m1	Cxcl10	CXCL10	IVIIg IP-10	IP-10	Mm00445235_m1
Cxrl1CxCl12CXCl12SDF-1Mn00+45552_m1Cxcl13CXCL13BLCBCA-1Mn00445532_m1IlaraCXCR1Mn0073132_s1Mn0043528_m1IlarbCXCR2Mn0043528_m1Cxr3CXCR3Mn0043828_m1Cxr4CXCR4Mn0043828_m1Cxr5CXCR5Mn00438259_m1Cxr4CXCR1Mn00438259_m1Cxr5CXCR4Mn00438259_m1Cxr1CX,CR1Mn00438259_m1Cxr2CXCR5Mn00438254_m1Cx3c11CX,CC11NeurotactinCx3c11CX,CR1Mn00438454_m1Xcr1XCL1LymphotactinXcr1XCR1Mn00438772_m1Xcr1XCR1Mn00443772_m1SeleE-selectinMn0041278_m1SelpP-selectin1Mn00441285_m1Angto1Angiopoietin 1Mn0045652_m1Angt0GAPDHMn0043242_m1Housekeeping and otherIe-2Mn0043242_m1Mph2Agiapoietin 2Mn0047828_m1Nph51NephrinMn00497828_m1Nph52PodocinMn00497828_m1Aqp2Aquaporin 2Mn0043775_m1Lrp2MealinMn0043775_m1	Cycl11	CXCL11			Mm00444662 m1
CX112SDP-1SDP-1MIN0P4352_m1CX113BLCBCA-1Min0044532_m1Il8raCXCR1Min00438258_m1Cxcr3CXCR2Min00438258_m1Cxcr4CXCR4Min00438258_m1Cxcr5CXCR3Min00438258_m1Cxcr4CXCR4Min00438258_m1Cxc75CXCR4Min00438258_m1Cxc74CXCR4Min00438258_m1Cxc75CXCR5Min00438258_m1Cx3c1CX_5C1Nin00438268_m1Cx3c1CX_5C1Min00438258_m1Xcr1CX_5C1Min00438254_m1Xcr1CXG1Min00438254_m1Xcr1XCR1Min0044720_m1SeleE-selectinMin0044720_m1SelpP-selectinMin00441278_m1Angpt1Angiopoietin 1Min0044522_m1Angpt2Angiopoietin 2Min00443222_m1Housekeeping and otherIte-2Min0044322_m1InselGAPDHMin00497828_m1Nphs1NephrinMin00497828_m1Nphs1NephrinMin00497828_m1Nphs2PodocinMin00497828_m1Aqp2Aquaporin 2Min0043755_m1Lrp2MealinMin0043275_m1	Crell2	CYCL12	SDE 1	SDE 1	Mm00444002_1111
CARLYCARLYMIN0044713/29_x1Il8raCXCR1MIN0044713/29_x1Il8rbCXCR2MIN004313/29_x1Cxr3CXCR3MIN0043285_m1Cxr4CXCR4MIN0043285_m1Cxr5CXCR5MIN0043285_m1Cxr5CXCR5MIN0043286_m1Cx3Cal C chemokines and receptorsKCR1MIN0043286_m1Cx3Cal C chemokines and receptorsCXCR1MIN0043286_m1Cx3Cal C Chemokines and receptorsKCR1MIN00438354_m1Cx3Cal C Chemokines and receptorsKCR1MIN0043270_m1Xcr1XCL1LymphotactinLymphotactinXcr1XCR1MIN0043270_m1Xcr1XCR1MIN00441295_m1SeleE-selectinMIN00441295_m1Angiopoietin 1MIN004456503_m1Angiopoietin 2MIN00456503_m1Angiopoietin 2MIN00456503_m1Angiopoietin 2MIN00456503_m1Housekeeping and otherIte-2GapdhGAPDHMinphi1NephrinNphs1NephrinNphs2PodocinAquaporin 2MIN00497575_m1Lrp2MegalinMino1328171_m1	Cxcl12	CYCL13	BLC	BCA 1	Mm00445552_m1
Inda CACR MIND073129_31 IBrb CXCR2 MIN0073129_31 Cxcr3 CXCR3 MIN0073129_31 Cxcr4 CXCR3 MIN0073129_31 Cxcr4 CXCR3 MIN0073129_31 Cxcr4 CXCR3 MIN0073129_31 Cxcr4 CXCR4 MIN0073129_31 Cxcr5 CXCR4 MIN0073129_31 Cxcr4 CXCR4 MIN0073129_31 Cxcr5 CXCR5 MIN00438259_m1 Cxcr5 CXCR5 MIN00432086_m1 CX3c1 KCR1 MIN00438354_m1 Cx3c1 CX3CR1 MIN00438354_m1 Xc11 XCR1 MIN00438354_m1 Xcr1 XCR1 MIN00476702_m1 Selp CAS1 MIN00476702_m1 Selp Pselectin MIN00476702_m1 Angtp2 Angiopoietin 1 MIN00476703_m1 Angtp3 Angiopoietin 1 MIN00476703_m1 Angtp4 Angiopoietin 1 MIN00476703_m1 Angtp4 Angiopoietin 2 MIN00476703_		CYCD1	BEC	DCA-1	Mm00721220_01
Into CXCR2 Minloot38259_m1 Cxcr3 CXCR3 Mm00438259_m1 Cxcr5 CXCR4 Mm9999055_m1 Cxc75 CXCR5 Mm00438259_m1 Cxc74 CXCR5 Mm00438259_m1 Cxc75 CXCR1 Mm00438254_m1 Cx3c1n CX ₅ CR1 Mm00438354_m1 Xc1 XCR1 Mm00438256_m1 Xc1 XCR1 Mm00438256_m1 Endothelial and angiogenic KCR1 Mm00438256_m1 Pecam1 CD31 Mm00476702_m1 Selp P-selectin Mm00441278_m1 Angpt1 Angiopoietin 1 Mm00456503_m1 Angpt2 Angiopoietin 2 Mm00456503_m1 Housekeeping and other Ire2 Mm00456503_m1 Selp Selp Mm00456503_m1 Angiopoietin 2 Mm00456503_m1 Angiopoietin 3 Mm00456503_m1 Angiopoietin 4 Mm00456503_m1 Angpt2 Angiopoietin 2 Mm004545822_m1 Mphs1 Selp Mm00497828_m1	Ilorb	CYCR2			Mm00429259_m1
CXr13CXr24Minlour3255_minCxr24CXCR4Min0043255_minCxr25CXCR5Min0043206_minCX3C1CX3CL1NeurotactinFractalkineCX3cr1CX_3CR1Min00438354_minXcr1XCL1LymphotactinLymphotactinXcr1XCR1Min00438354_minXcr1XCR1Min00438354_minSeleE-selectinMin00442206_s1SelpP-selectinMin00441278_minSelpP-selectinMin00441295_minAngpt1Angiopoietin 1Min00441295_minAngpt2Angiopoietin 2Min00443224_minHousekeeping and otherIie-2Min00438354_minGapdhGAPDHMin00443224_minNphs1NephrinMin0043828_minNphs2PodocinMin0043828_minNphs2Aquaporin 2Min00437575_minAqp2Aquaporin 2Min00432875_minAngp2MegalinMin00432871_min	Cycr3	CYCP3			Mm00438259_m1
CXr4*CXr4*Mm997903_minCXr4*CXr4*Mm0043086_mCXr5CXr4*Mm00436454_m1CXr1CXr1NeurotactinFractalkineCx3cr1CXr1CXr1Mm00436454_m1Xc1XCL1LymphotactinLymphotactinXcr1XCR1Mm00436772_m1Xcr1XCR1Mm00442206_s1Endothelial and angiogenicMm00476702_m1SeleE-selectinMm0041278_m1SelpP-selectinMm0044205_m1Angpt1Angiopoietin 1Mm00442653_m1Angpt2Angiopoietin 2Mm00443032_m1Housekeeping and otherTie-2Mm0043242_m1Hs185Hs9999915_g1185185Hs9999901_s1Nphs1NephrinMm00497828_m1Nphs2PodocinMm00497828_m1Nphs2Aquaporin 2Mm00499257_m1Lrp2MegalinMm01328171_m1	Cxcr4	CYCP4			Mm0000055 m1
CxcribKerrolMintod-92006_minCX3C and C chemokines and receptorsCx3cl1CX3CL1NeurotactinFractalkineMm00436454_m1Cx3cr1CX3CR1Mm0043854_m1Xcl1XCL1LymphotactinMm0043854_m1Xcr1XCR1Mm0043006_s1Endothelial and angiogenicMm0043206_s1Pecam1CD31Mm00476702_m1SeleE-selectinMm00476702_m1SelpP-selectinMm00441278_m1Angpt1Angiopoietin 1Mm00445503_m1Angpt2Angiopoietin 2Mm00445503_m1Housekeeping and otherMm00443242_m1Hs185185Hs9999901_s1Nphs1NephrinMm00497828_m1Nphs2PodocinMm00437575_m1Aqp2Aquaporin 2Mm00437575_m1Lrp2MegalinMm01328171_m1	Cxcr5	CYCP5			Mm00432086 m1
CX3C and C chemokines and receptorsCX3Cl1CX3CL1NeurotactinFractalkineMm00436454_m1CX3cr1CX3CR1Mm0043854_m1Xcl1XCL1LymphotactinMm00434772_m1Xcr1XCR1Mm00442206_s1Endothelial and angiogenicE-selectinMm00441278_m1SeleE-selectinMm00441295_m1Angpt1Angiopoietin 1Mm00456503_m1Angpt2Angiopoietin 2Mm00456503_m1Angpt2Tie-2Mm0043242_m1Housekeeping and otherIfie-2Mm9999915_g118S18S18SHs9999901_s1Nphs1NephrinMm00497828_m1Nphs2Aquaporin 2Mm00437575_m1Aqp2Aquaporin 2Mm00437575_m1Lrp2MegalinMm01328171_m1		CACRO			WIII00452080_III1
Cx3c1Cx3c1NeurotactinFractakineMm00436434_m1Cx3cr1CX3CR1Mm00438354_m1Xc1XCL1LymphotactinLymphotactinXcr1XCR1LymphotactinMm00434772_m1Kr1CD31Mm00476702_m1SeleE-selectinMm0041278_m1SelpP-selectinMm0041295_m1Angpt1Angiopoietin 1Mm00456503_m1Angpt2Angiopoietin 2Mm00456503_m1TekTie-2Mm0043242_m1Housekeeping and otherIseHs9999901_s1Nphs1NephrinMm0047828_m1Nphs2OdocinMm0049828_m1Aquaporin 2Mm0049829_m1Aquaporin 2Mm0047875_m1Lrp2MegalinMm0043775_m1	CX_3C and C chemokines and receptors	OV OL 1			N 00426454 1
Cx3cr1CX3cR1Mm00438354_m1Xcl1XCL1LymphotactinLymphotactinMm00434772_m1Xcr1XCR1Mm0042206_s1Endothelial and angiogenicMm00476702_m1Pecam1CD31Mm00476702_m1SeleE-selectinMm0041278_m1SelpP-selectinMm0041295_m1Angpt1Angiopoietin 1Mm0045653_m1Angpt2Angiopoietin 2Mm0045653_m1TekTie-2Mm0043222_m1Housekeeping and otherISSMm9999915_g118S18SHs9999901_s1Nphs1NephrinMm00497828_m1Nphs2PodocinMm00497828_m1Aqp2Aquaporin 2Mm00437575_m1Lrp2MegalinMm01328171_m1		CX ₃ CL1	Neurotactin	Fractalkine	Mm00436454_m1
Xcl1LympnotactinLympnotactinLympnotactinMm004347/2_miXcr1XCR1Mm00442206_s1Endothelial and angiogenicEndothelial and angiogenicMm00476702_m1Pecam1CD31Mm00476702_m1SeleE-selectinMm00441278_m1SelpP-selectinMm00441295_m1Angpt1Angiopoietin 1Mm00456503_m1Angpt2Ite-2Mm00443242_m1TekTie-2Mm00443242_m1Housekeeping and otherIte-2Mm0043242_m1I88185Hs9999901_s1Nphs1NephrinMm00497828_m1Nphs2PodocinMm00497828_m1Aqu2Aquaporin 2Mm00437575_m1Lrp2MegalinMm01328171_m1	Cx5cr1	CA ₃ CKI		T 1 4 4	Mm00438354_m1
XcriXcRiMm00442206_siEndothelial and angiogenicPecamlCD31SeleE-selectinSelpP-selectinAngpt1Angiopoietin 1Angpt2Angiopoietin 2TekTie-2Housekeeping and otherGapdhGAPDHNphs1NephrinNphs2PodocinAqu20Aquaporin 2Angp2Aquaporin 2Mm00437575_m1Lrp2MegalinMegalinMm00437575_m1Mrg1328171_m1	XcII	XCLI	Lymphotactin	Lymphotactin	Mm00434//2_m1
Endothelial and angiogenic Mm00476702_m1 Pecam1 CD31 Mm00476702_m1 Sele E-selectin Mm00441278_m1 Selp P-selectin Mm00441295_m1 Angpt1 Angiopoietin 1 Mm00456503_m1 Angpt2 Angiopoietin 2 Mm00456503_m1 Tek Tie-2 Mm00454822_m1 Housekeeping and other Tie-2 Mm0043242_m1 ISS GAPDH Mm9999915_g1 18S ISS Hs9999901_s1 Nphs1 Nephrin Mm00497828_m1 Nphs2 Podocin Mm00437575_m1 Aquaporin 2 Mq0aprin 2 Mm00437575_m1 Lrp2 Megalin Mm01328171_m1	Xcr1	XCRI			Mm00442206_\$1
Pecam1 CD31 Mm00476702_m1 Sele E-selectin Mm00441278_m1 Selp P-selectin Mm00441295_m1 Angpt1 Angiopoietin 1 Mm00456503_m1 Angpt2 Angiopoietin 2 Mm00456503_m1 Tek Tie-2 Mm00443242_m1 Housekeeping and other Mm00443242_m1 ISS ISS Mm00497828_m1 Nphs1 Nephrin Mm00497828_m1 Nphs2 Podocin Mm00497828_m1 Aquaporin 2 Mm00437575_m1 Lrp2 Megalin Mm004328275_m1	Endothelial and angiogenic				
Sele E-selectin Mm00441278_m1 Selp P-selectin Mm00441295_m1 Angpt1 Angiopoietin 1 Mm00456503_m1 Angpt2 Angiopoietin 2 Mm00456503_m1 Tek Tie-2 Mm0043242_m1 Housekeeping and other Mm00443242_m1 ISS GAPDH Mm9999915_g1 18S 18S Hs9999901_s1 Nphs1 Nephrin Mm00497828_m1 Nphs2 Podocin Mm00437575_m1 Aquaporin 2 Mgalin Mm00437575_m1	Pecam1	CD31			Mm00476702_m1
Selp P-selectin Mm0041295_m1 Angpt1 Angiopoietin 1 Mm00456503_m1 Angpt2 Angiopoietin 2 Mm00545822_m1 Tek Tie-2 Mm0043242_m1 Housekeeping and other Mm9999915_g1 I8S I8S Hs9999901_s1 Nphs1 Nephrin Mm00497828_m1 Nphs2 Podocin Mm0049755_m1 Aqu2 Aquaporin 2 Mm00437575_m1 Lrp2 Megalin Mm01328171_m1	Sele	E-selectin			Mm00441278_m1
Angpt1 Angiopoietin 1 Mm00456503_m1 Angpt2 Angiopoietin 2 Mm00545822_m1 Tek Tie-2 Mm0043242_m1 Housekeeping and other Mm0043242_m1 Gapdh GAPDH Mm9999915_g1 18S 18S Hs9999901_s1 Nphs1 Nephrin Mm00497828_m1 Nphs2 Podocin Mm0049755_m1 Aquaporin 2 Mm00437575_m1 Lrp2 Megalin Mm01328171_m1	Selp	P-selectin			Mm00441295_m1
Angpt2 Angiopoietin 2 Mm00545822_m1 Tek Tie-2 Mm00443242_m1 Housekeeping and other Mm00443242_m1 Gapdh GAPDH Mm9999915_g1 18S 18S Hs9999901_s1 Nphs1 Nephrin Mm00497828_m1 Nphs2 Podocin Mm0049755_m1 Aquaporin 2 Mm00437575_m1 Lrp2 Megalin Mm01328171_m1	Angpt1	Angiopoietin 1			Mm00456503_m1
Tek Tic-2 Mm00443242_m1 Housekeeping and other GAPDH Mm9999915_g1 I8S I8S Hs9999901_s1 Nphs1 Nephrin Mm00497828_m1 Nphs2 Podocin Mm00497828_m1 Aqp2 Aquaporin 2 Mm00437575_m1 Lrp2 Megalin Mm01328171_m1	Angpt2	Angiopoietin 2			Mm00545822_m1
Housekeeping and other GAPDH Mm9999915_g1 Gapdh GAPDH Mm9999901_s1 18S 18S Hs9999901_s1 Nphs1 Nephrin Mm00497828_m1 Nphs2 Podocin Mm00497828_m1 Aqp2 Aquaporin 2 Mm00437575_m1 Lrp2 Megalin Mm01328171_m1	Tek	Tie-2			Mm00443242_m1
Gapdh GAPDH Mm9999915_g1 18S 18S Hs9999901_s1 Nphs1 Nephrin Mm00497828_m1 Nphs2 Podocin Mm00497828_m1 Aqp2 Aquaporin 2 Mm00437575_m1 Lrp2 Megalin Mm01328171_m1	Housekeeping and other				
18S 18S Hs9999901_s1 Nphs1 Nephrin Mm00497828_m1 Nphs2 Podocin Mm0049929_m1 Aqp2 Aquaporin 2 Mm00437575_m1 Lrp2 Megalin Mm01328171_m1	Gapdh	GAPDH			Mm99999915_g1
Nphs1 Nephrin Mm00497828_m1 Nphs2 Podocin Mm0049929_m1 Aqp2 Aquaporin 2 Mm00437575_m1 Lrp2 Megalin Mm01328171_m1	18S	18S			Hs99999901_s1
Nphs2 Podocin Mm00499929_m1 Aqp2 Aquaporin 2 Mm00437575_m1 Lrp2 Megalin Mm01328171_m1	Nphs1	Nephrin			Mm00497828_m1
Aqp2 Aquaporin 2 Mm00437575_m1 Lrp2 Megalin Mm01328171_m1	Nphs2	Podocin			Mm00499929_m1
Lrp2 Megalin Mm01328171_m1	Aqp2	Aquaporin 2			Mm00437575_m1
	Lrp2	Megalin			Mm01328171_m1

[†]BCA-1, B cell-activating chemokine-1; BLC, B lymphocyte chemoattractant; ENA-78, epithelial cell-derived neutrophil-activating factor, 78 amino acids; GAPDH, glyceraldehyde-3-phosphate dehydrogenase; GRO α/β , growth-related oncogene α/β ; IP-10, interferon-inducible protein-10; I-TAC, interferon-inducible T cell α -chemoattractant; KC, keratinocyte-derived chemokine; LIX, lipopolysaccharide-induced CXC chemokine; MARC, mast cell activation-related chemokine; MCP-#, monocyte chemoattractant protein-#; MDC, macrophage-derived chemokine; Mig, monokine induced by interferon- γ ; MIP-#, macrophage inflammatory protein-#; PF-4, platelet factor-4; RANTES, regulated on activation normal T cell expressed and secreted; SDF-1, stromal cell-derived factor-1; TARC, thymus- and activation-related chemokine; TCA-3, T-cell activation protein-3.



Fig. 1. Relative gene expression of chemokines and chemokine receptors in mouse kidney during the acute inflammation phase of anti-myeloperoxidase (MPO) immunoglobulin (Ig)G-induced necrotizing crescentic glomerulonephritis (NCGN). (a–e) Relative mRNA levels of (a) CC-chemokines, (b) CC-chemokine receptors, (c) CXC-chemokines, (d) CXC-chemokine receptors and (e) CX₃C- and XC-chemokines and their receptors in kidneys from healthy control mice (white bars), mice that received lipopolysaccharide (LPS) only (grey bars) and mice that received anti-MPO IgG and LPS (black bars) 1 day after administration. Bars represent mean \pm standard deviation (s.d.). **P* < 0.05 *versus* control mice; #*P* < 0.05 *versus* LPS-treated mice. (f–j) Relative mRNA levels in laser-microdissected glomeruli (striped bars) and tubulo-interstitial areas (dotted bars) from mice that received anti-MPO IgG and LPS 1 day after administration. (f) CC-chemokines, (g) CC-chemokine receptors, (h) CXC-chemokines, (i) CXC-chemokine receptors and (j) CX₃C- and XC-chemokines and their receptors. Bars represent mean \pm s.d. **P* < 0.05; n.d.: not detected; n.t.: not tested – CXCR4 could not be analysed due to insufficient amplification reactions.

Results

Chemokine and chemokine receptor expression in the acute inflammatory phase of anti-MPO IgG-induced NCGN

Gene expression analysis of chemokines and chemokine receptors in renal tissue 1 day after induction of experimen-

tal anti-MPO IgG-mediated NCGN revealed induction or up-regulation of several chemokines when compared to expression levels in non-treated and LPS-treated mice (Fig. 1a–e). These chemokines belonged to the CC-family of chemokines, CCL3 and CCL5, and the CXC-family, CXCL2, CXCL5 and CXCL13. Interestingly, CXCR2 was the only chemokine receptor with a significantly higher expression level in the acute phase of disease development. Subse-

	Pla	Plasma (pg/ml)			Renal homogenate (ng/mg total protein)		
Group	CXCL1	CXCL2	CXCL5	CXCL1	CXCL2	CXCL5	
Control	166·1 ± 80·9	n.d.	n.t.	0.80 ± 0.15	1.94 ± 0.30	5.30 ± 0.93	
LPS day 1	601.6 ± 210.4	$78\cdot3 \pm 32\cdot7$	n.t.	0.86 ± 0.23	1.83 ± 0.46	4.81 ± 0.97	
Anti-MPO IgG + LPS day 1	$432{\cdot}5\pm47{\cdot}5$	60.1 ± 22.8	n.t.	0.78 ± 0.10	1.80 ± 0.14	5.65 ± 0.26	
LPS day 7	172.1 ± 22.9	n.d.	n.t.	0.62 ± 0.15	1.41 ± 0.29	4.64 ± 1.09	
Anti-MPO IgG + LPS day 7	$1075.6 \pm 1030.5^*$	n.d.	n.t.	0.59 ± 0.05	1.34 ± 0.17	3.73 ± 0.76	

Table 2. Plasma and renal tissue protein levels of CXCL1, CXCL2 and CXCL5 in mice suffering from anti-MPO IgG-induced NCGN.

*P < 0.05 compared to both control and lipopolysaccharide (LPS) day 7; mean \pm standard deviation; n.d., not detected; n.t., not tested. Ig, immunoglobulin; MPO, myeloperoxidase; NCGN, necrotizing crescentic glomerulonephritis.

quently, we determined the localization of chemokine expression by comparing expression levels between laser microdissected glomeruli and the tubulo-interstitial area. We confirmed accurate separation of the compartments with laser microdissection by analysing expression of glomerulusspecific (podocin and nephrin) and tubulus-restricted (megalin and aquaporin-2) genes (data not shown). We found that gene expression of most of the chemokines and chemokine receptors was localized predominantly in the glomerular compartment (Fig. 1f–j).

Chemokine and chemokine receptor expression in the crescentic phase of anti-MPO IgG-induced NCGN

Gene expression analysis in renal tissue obtained from mice 7 days after induction of anti-MPO IgG-mediated glomerulonephritis demonstrated the induction or up-regulation of various chemokines and chemokine receptors compared to non-treated and LPS-treated mice (Fig. 2a-e). Most of the chemokines with increased expression at day 7 belonged to the CC-chemokines: CCL2, CCL5, CCL7, CCL8, CCL17 and CCL20. In addition, increased expression was observed for the CC-chemokine receptors CCR2 and CCR8 and CXCchemokine CXCL10. In contrast to day 1, no increased expression of CCL3 and CXCL13 was found at day 7. CXCL1, CXCL2 and CXCL5, the ligands for CXCR2, were also up-regulated in the crescentic phase, although the receptor itself was expressed at a level similar to control mice. Furthermore, CX₃CL1 and its receptor CX₃CR1 were expressed to an increased extent. Analysis of dissected renal tissue showed that expression of the majority of chemokines and receptors was also localized in glomeruli at this later time-point (Fig. 2f-j). However, for some chemokines the pattern changed. CXCL2 was expressed highly in glomeruli but less so in the tubulo-interstitium on day 1, while on day 7 no significant difference was found.

Protein expression of CXCR2 and CXCR2 ligands

Because a temporal induction of CXCR2 was detected in the acute phase and induction of its ligands in both the acute and the crescentic phases, we aimed to investigate further the

role of CXCR2 and its ligands in anti-MPO IgG-induced NCGN. Analysis of circulating CXCL1 and CXCL2 protein levels demonstrated increased CXCL1 levels in the crescentic phase of anti-MPO IgG-induced NCGN (Table 2). In contrast, CXCL1, CXCL2 and CXCL5 protein levels in renal homogenates did not change during the course of anti-MPO IgG-induced NCGN (Table 2). In addition, we wanted to determine to what extent intrinsic glomerular cells contribute to glomerular CXCR2 expression. Recent data indicate that expression of CXCR2 is not restricted to inflammatory cells, but can also be detected on microvascular endothelial cells. Furthermore, endothelial CXCR2 has been demonstrated to be involved in LPS-induced neutrophil infiltration in the lungs [21]. We speculated that CXCR2 was expressed on glomerular endothelial cells and that endothelial CXCR2 could contribute to the early neutrophil accumulation, as observed in anti-MPO/LPS-induced glomerulonephritis. We studied whether human CiGEnC expressed CXCR2. CXCR2 mRNA was detected in CiGEnC, but its expression was much lower compared to expression in human neutrophils (Fig. 3a). CXCR1 mRNA was not detected in CiGEnC. Treatment of CiGEnC with IL-8 induced an intracellular calcium flux in approximately 70% of the cells, which was mediated by CXCR2 but not by CXCR1 (Fig. 3b). These results indicate that glomerular endothelial cells express a functional CXCR2 receptor.

In vivo inhibition of CXCR2 does not attenuate renal injury in anti-MPO IgG-induced NCGN

To study the functional role of the CXCR2–CXCR2 ligand axis in anti-MPO IgG/LPS-mediated glomerulonephritis, we blocked the receptor employing a CXCR2 blocking serum. Compared to control-treated mice, mice that had received anti-CXCR2 serum showed an increase in glomerular neutrophil accumulation 1 day after anti-MPO IgG/LPS administration (Fig. 4a and b). Moreover, inhibition of CXCR2 did not change the percentage of glomeruli positive for neutrophils (control 65.7 ± 5.0 and anti-CXCR2 $66.3 \pm 6.3\%$) but rather increased the mean number of neutrophils per positive glomerulus (control 1.75 ± 0.30 and anti-CXCR2 2.59 ± 0.50 , P < 0.05). Treatment of mice suffering from anti-MPO IgG-induced NCGN with the anti-CXCR2



Fig. 2. Relative gene expression of chemokines and chemokine receptors in mouse kidney during the crescentic phase of anti- myeloperoxidase (MPO) immunoglobulin (Ig)G-induced necrotizing crescentic glomerulonephritis (NCGN). (a–e) Relative mRNA levels of (a) CC-chemokines, (b) CC-chemokine receptors, (c) CXC-chemokines, (d) CXC-chemokine receptors and (e) CX₃C- and XC-chemokines and their receptors in kidneys from healthy control mice (white bars), mice that received lipopolysaccharide (LPS) only (grey bars) and mice that received anti-MPO IgG and LPS (black bars) 7 days after administration. Bars represent mean \pm standard deviation (s.d.). **P* < 0.05 *versus* control mice; #*P* < 0.05 *versus* LPS-treated mice. (f–j) Relative mRNA levels in laser-microdissected glomeruli (striped bars) and tubulo-interstitial areas (dotted bars) from mice that received anti-MPO IgG and LPS 7 days after administration. (f) CC-chemokines, (g) CC-chemokine receptors, (h) CXC-chemokines, (i) CXC-chemokine receptors and (j) CX₃C- and XC-chemokines and their receptors. Bars represent mean \pm s.d. **P* < 0.05; n.d.: not detected; n.t.: not tested – CXCR4 could not be analysed due to insufficient amplification reactions.

blocking serum did not reduce haematuria and albuminuria (Fig. 4c and d). In addition, CXCR2 inhibition did not influence glomerular crescent formation, as a similar percentage of crescentic glomeruli was detected in control and anti-CXCR2-treated mice (Fig. 4e). To confirm the effects of the CXCR2 blocking serum, a group of anti-MPO IgG-induced NCGN mice were treated with the CXCR2 inhibitor repertaxin. No differences were found between vehicle- and repertaxin-treated animals with respect to albuminuria on day 1 (vehicle 28.9 ± 16.0 and repertaxin $21.7 \pm 0.8 \,\mu\text{g/}$ 17 h, mean \pm standard deviation, n = 3) and day 7 (110.8 ± 119.7 and $119.1 \pm 52.2 \,\mu\text{g/}17$ h) and the extent of glomerular crescent formation on day 7 (11.0 ± 3.6 and $9.3 \pm 1.5\%$).



Fig. 3. Human glomerular endothelial cells functionally express CXCR2 *in vitro*. (a) Relative CXCR2 mRNA levels in conditionally immortalized glomerular endothelial cells (CiGEnC) and human neutrophils. Bars represent mean \pm standard deviation (s.d.) of measurements in two independent cell preparations (CiGEnC) or three donors (neutrophils). (b) Percentage of CiGEnC responding to interleukin (IL)-8 treatment by intracellular calcium flux in the presence or absence of CXCR1- and CXCR2-blocking antibodies. Bars represent the mean percentage of cells responding to IL-8 in three independent experiments (n = 34-142 cells/condition). *P < 0.01.

Discussion

In the present study, we demonstrate spatiotemporal differences in chemokine and chemokine receptor gene expression levels in a mouse model of anti-MPO IgG-induced NCGN. Our objective was to identify those chemokines and chemokine receptors whose expression patterns suggest involvement in the recruitment of specific leucocyte subsets.

Induction of CXCR2 was restricted to the acute inflammation phase. Its ligands, CXCL1, CXCL2 and CXCL5, were up-regulated in both the acute and crescentic phases of the disease. Our findings suggest that CXCL2 expression is restricted to glomeruli in the acute phase of anti-MPO IgGinduced NCGN, whereas in the crescentic phase CXCL2 is expressed in both the glomerular and the tubulo-interstitial compartment, which correlates with the presence of both glomerular and interstitial inflammatory infiltrates at this time-point. We chose CXCR2 as an initial target for intervention because the temporal induction of CXCR2 suggested involvement of CXCR2 in recruitment of neutrophils, which are pivotal effector cells in anti-MPO IgG-induced NCGN [4]. Furthermore, several studies have demonstrated reduced glomerular neutrophil influx and albuminuria in models of glomerulonephritis upon inhibition of CXCR2 or its ligands [22-25]. The measurements of CXCL1, CXCL2 and CXCL5 protein in kidney tissue did not confirm the increase in tissue-specific expression of these chemokines suggested by analysis of mRNA levels. In plasma, we observed a higher CXCL2 level at day 7. Interestingly, increased circulating levels of IL-8 have been observed in ANCA-associated vasculitis patients [26].

Interestingly, blocking CXCR2 in our model of anti-MPO IgG-induced NCGN did not prevent early glomerular neutrophil influx but increased glomerular accumulation of neutrophils. In vitro flow assays have revealed that inhibition of neutrophil CXCR2 reduces transendothelial migration of ANCA-activated neutrophils [27]. We speculate that in our experiment neutrophils were recruited to glomeruli via chemoattractants other than CXCR2 ligands but, due to inhibition of CXCR2-mediated transendothelial migration, were retained within the vascular compartment. In line with this, Cockwell et al. hypothesized that frustrated neutrophil transmigration due to high levels of intravascular IL-8 contributes to glomerular injury in ANCA-associated glomerulonephritis [7]. The observation that inhibition of CXCR2accelerated glomerular neutrophil accumulation would also imply a worsening of other disease parameters, but we did not observe increased urinary abnormalities or crescent formation, which was supported by the repertaxin experiment. Based on the experiments conducted, we cannot explain this discrepancy, although our data suggest that the extent of kidney injury is determined primarily by the number of affected glomeruli. A probable neutrophil chemoattractant involved in the neutrophil recruitment in anti-MPO IgGinduced NCGN is C5a. Recently, genetic ablation or inhibition of complement factor C5, and consequently C5-derived C5a, was found to abrogate disease development completely in this model [16,28]. Moreover, neutrophils from CXCR2^{-/-} mice retain chemotactic activity towards C5a [29]. An additional possibility is that the activity of other chemokines and chemokine receptors becomes more important, as there is redundancy in the chemokine system [30].

The observed increase in CXCR2 mRNA at day 1 in our disease model is caused most probably by the glomerular influx of neutrophils and not by up-regulation of endothelial CXCR2, as neutrophils express relatively high levels of CXCR2. Reutershan and colleagues demonstrated that endothelial CXCR2 was involved in LPS-induced neutrophil transmigration in the lungs [21]. Although lung tissue has a higher expression of CXCR2 compared to kidney, we cannot exclude the possibility that inhibition of endothelial CXCR2 in glomeruli contributed to neutrophil transmigration impairment in our CXCR2-intervention experiment.

Other chemokines that had increased expression in the acute phase were CCL3 and CCL5, suggesting their involvement in leucocyte influx. Increased protein levels of CCL3 and CCL5 have been found in vasculitic lesions in lungs of Wegener's granulomatosis [31]. None of the receptors for CCL3 and CCL5 (e.g. CCR1, CCR5), however, were up-regulated in our model.

In the crescentic phase of anti-MPO IgG-induced NCGN, CCR2 was up-regulated and expressed predominantly in glomeruli. The CCR2 ligands CCL2 and CCL7 were also expressed to an increased extent and mainly in glomeruli. Up-regulation of CCR2 and its ligands suggests involvement in the monocyte/macrophage influx observed in the crescen-



Fig. 4. Inhibition of CXCR2 in anti-myeloperoxidase (MPO) immunoglobulin (Ig)G-induced necrotizing crescentic glomerulonephritis (NCGN) enhanced early glomerular neutrophil influx but did not affect urinary abnormalities and crescent formation. (a) Immunohistochemical staining of neutrophils (Ly6G) on kidney cryosections of mice treated with control serum (left) or CXCR2 blocking serum (right) 1 day after anti-MPO IgG/lipopolysaccharide (LPS) administration. Original magnification $400\times$. (b) Quantification of neutrophil influx at day 1. gcs: glomerular cross-section. **P* < 0.01. (c) Administration of anti-MPO IgG/LPS caused marked haematuria after 1 and 7 day(s). No differences were observed between mice treated with control serum (\Box) and mice treated with CXCR2-blocking serum (\blacktriangledown). (d) Albuminuria at 1 and 7 days after anti-MPO IgG/LPS administration. Treatment with CXCR2-blocking serum did not prevent albuminuria. Open bars: control treatment; filled bars: anti-CXCR2 treatment. Bars represent mean ± standard deviation (s.d.). The average level of albumin in urine of untreated mice was $40.7 \pm 24.0 \,\mu$ g/17 h. (e) Quantification of glomerular crescents revealed no difference between control-treated and anti-CXCR2-treated mice. Bars represent mean ± s.d.

tic phase of anti-MPO IgG-induced NCGN. Neutralization studies have demonstrated important roles for CCR2 and CCL2 in monocyte/macrophage influx and, in some cases, crescent formation in other models of crescentic glomerulonephritis [32–34]. Furthermore, CCL2 protein has been detected in glomerular and interstitial cells in ANCAassociated vasculitis patients with renal involvement [35].

In addition, we observed up-regulation of CX₃CL1 and its receptor CX₃CR1 in the crescentic phase. CX₃CL1 is a transmembrane domain-containing chemokine with monocyte attracting properties. CX₃CL1 mRNA expression has been observed in glomerular lesions of vasculitic patients [36], whereas inhibition of CX₃CR1 in a rat model of crescentic glomerulonephritis attenuated glomerular leucocyte influx and reduced crescent formation [37]. These data suggest that CX₃CL1 may be involved in the pathogenesis of anti-MPO IgG-induced NCGN. Our finding that several chemokines were up-regulated during anti-MPO IgG-induced glomerulonephritis raises questions on the cellular source of chemokine production. We speculate that intrinsic renal (predominantly glomerular) cells are, in part, responsible for the production of chemokines, as are infiltrating inflammatory cells. The relative contributions of the different intrinsic glomerular cells – glomerular endothelial cells, podocytes and mesangial cells – in chemokine production could be investigated *in vitro* in future studies.

In this study, we have used murine anti-MPO IgGinduced glomerulonephritis as a model for human ANCAassociated vasculitis. For the translation of our data to the human situation, it is important to realize that the mouse model mimics ANCA-induced effects on neutrophils (and other cells) but does not involve a genuine autoimmune response. In addition, the model encompasses a proinflammatory stimulus, which may have modulated expression of chemokines and receptors in a way that does not occur in human ANCA-associated vasculitis.

In conclusion, various chemokine receptors and chemokines, including CXCR2, CCR2 and CX₃CR1 and their ligands, are up-regulated during the course of anti-MPO IgG-induced NCGN, and they probably contribute to shaping the inflammatory response. These chemokines and chemokine receptors can be tested further as potential targets in anti-MPO IgG-induced glomerulonephritis. Inhibition of one potential target, CXCR2, did not diminish anti-MPO IgG-induced NCGN. On the contrary, CXCR2 inhibition may, hypothetically, accelerate the vasculitic process by retaining neutrophils in the vascular compartment. Intervening with multiple chemokine targets is probably necessary to avoid redundancy of the chemokine system.

Acknowledgements

Peter Heeringa is supported by the Dutch Organization of Scientific Research (ZonMW VIDI 917·066·341). We thank Hester I. Bakker for excellent technical assistance, Tjerk Feenstra and Peter J. Zwiers for help with laser microdissection and Attje S. Hoekstra and Hilmar R. J. van Weering for help with intracellular calcium measurements.

Disclosure

The authors declare no conflict of interest.

References

- 1 Falk RJ, Jennette JC. Anti-neutrophil cytoplasmic autoantibodies with specificity for myeloperoxidase in patients with systemic vasculitis and idiopathic necrotizing and crescentic glomerulonephritis. N Engl J Med 1988; **318**:1651–7.
- 2 Xiao H, Heeringa P, Hu P *et al.* Antineutrophil cytoplasmic autoantibodies specific for myeloperoxidase cause glomerulone-phritis and vasculitis in mice. J Clin Invest 2002; **110**:955–63.
- 3 Huugen D, Xiao H, van Esch A *et al.* Aggravation of antimyeloperoxidase antibody-induced glomerulonephritis by bacterial lipopolysaccharide: role of tumor necrosis factor-alpha. Am J Pathol 2005; **167**:47–58.
- 4 Xiao H, Heeringa P, Liu Z *et al.* The role of neutrophils in the induction of glomerulonephritis by anti-myeloperoxidase antibodies. Am J Pathol 2005; **167**:39–45.
- 5 Murdoch C, Finn A. Chemokine receptors and their role in inflammation and infectious diseases. Blood 2000; 95:3032–43.
- 6 Olson TS, Ley K. Chemokines and chemokine receptors in leukocyte trafficking. Am J Physiol Regul Integr Comp Physiol 2002; 283:R7–28.
- 7 Cockwell P, Brooks CJ, Adu D, Savage CO. Interleukin-8: a pathogenetic role in antineutrophil cytoplasmic autoantibody-associated glomerulonephritis. Kidney Int 1999; 55:852–63.
- 8 Hsieh SC, Yu HS, Cheng SH et al. Anti-myeloperoxidase antibodies enhance phagocytosis, IL-8 production, and glucose uptake of

polymorphonuclear neutrophils rather than anti-proteinase 3 antibodies leading to activation-induced cell death of the neutrophils. Clin Rheumatol 2007; **26**:216–24.

- 9 Salcedo R, Resau JH, Halverson D *et al*. Differential expression and responsiveness of chemokine receptors (CXCR1-3) by human microvascular endothelial cells and umbilical vein endothelial cells. FASEB J 2000; **14**:2055–64.
- 10 Addison CL, Daniel TO, Burdick MD *et al.* The CXC chemokine receptor 2, CXCR2, is the putative receptor for ELR+ CXC chemokine-induced angiogenic activity. J Immunol 2000; 165:5269–77.
- 11 Li A, Dubey S, Varney ML, Dave BJ, Singh RK. IL-8 directly enhanced endothelial cell survival, proliferation, and matrix metalloproteinases production and regulated angiogenesis. J Immunol 2003; **170**:3369–76.
- 12 Heidemann J, Ogawa H, Dwinell MB *et al.* Angiogenic effects of interleukin 8 (CXCL8) in human intestinal microvascular endothelial cells are mediated by CXCR2. J Biol Chem 2003; 278:8508– 15.
- 13 Panzer U, Steinmetz OM, Stahl RA, Wolf G. Kidney diseases and chemokines. Curr Drug Targets 2006; 7:65–80.
- 14 Segerer S, Nelson PJ, Schlondorff D. Chemokines, chemokine receptors, and renal disease: from basic science to pathophysiologic and therapeutic studies. J Am Soc Nephrol 2000; 11:152–76.
- 15 Aratani Y, Koyama H, Nyui S, Suzuki K, Kura F, Maeda N. Severe impairment in early host defense against *Candida albicans* in mice deficient in myeloperoxidase. Infect Immun 1999; 67:1828–36.
- 16 Huugen D, van Esch A, Xiao H et al. Inhibition of complement factor C5 protects against anti-myeloperoxidase antibodymediated glomerulonephritis in mice. Kidney Int 2007; 71:646–54.
- 17 Asgeirsdottir SA, Kamps JA, Bakker HI *et al.* Site-specific inhibition of glomerulonephritis progression by targeted delivery of dexamethasone to glomerular endothelium. Mol Pharmacol 2007; 72:121–31.
- 18 Satchell SC, Tasman CH, Singh A *et al.* Conditionally immortalized human glomerular endothelial cells expressing fenestrations in response to VEGF. Kidney Int 2006; 69:1633–40.
- 19 Mehrad B, Strieter RM, Moore TA, Tsai WC, Lira SA, Standiford TJ. CXC chemokine receptor-2 ligands are necessary components of neutrophil-mediated host defense in invasive pulmonary aspergillosis. J Immunol 1999; 163:6086–94.
- 20 Moore TA, Newstead MW, Strieter RM, Mehrad B, Beaman BL, Standiford TJ. Bacterial clearance and survival are dependent on CXC chemokine receptor-2 ligands in a murine model of pulmonary *Nocardia asteroides* infection. J Immunol 2000; **164**:908–15.
- 21 Reutershan J, Morris MA, Burcin TL *et al.* Critical role of endothelial CXCR2 in LPS-induced neutrophil migration into the lung. J Clin Invest 2006; 116:695–702.
- 22 Wada T, Tomosugi N, Naito T *et al.* Prevention of proteinuria by the administration of anti-interleukin 8 antibody in experimental acute immune complex-induced glomerulonephritis. J Exp Med 1994; **180**:1135–40.
- 23 Wu X, Wittwer AJ, Carr LS, Crippes BA, DeLarco JE, Lefkowith JB. Cytokine-induced neutrophil chemoattractant mediates neutrophil influx in immune complex glomerulonephritis in rat. J Clin Invest 1994; 94:337–44.
- 24 Feng L, Xia Y, Yoshimura T, Wilson CB. Modulation of neutrophil influx in glomerulonephritis in the rat with anti-macrophage inflammatory protein-2 (MIP-2) antibody. J Clin Invest 1995; 95:1009–17.

- 25 Wu X, Dolecki GJ, Sherry B, Zagorski J, Lefkowith JB. Chemokines are expressed in a myeloid cell-dependent fashion and mediate distinct functions in immune complex glomerulonephritis in rat. J Immunol 1997; 158:3917–24.
- 26 Ohlsson S, Wieslander J, Segelmark M. Circulating cytokine profile in anti-neutrophilic cytoplasmatic autoantibody-associated vasculitis: prediction of outcome? Mediat Inflamm 2004; 13:275– 83.
- 27 Calderwood JW, Williams JM, Morgan MD, Nash GB, Savage CO. ANCA induces beta2 integrin and CXC chemokine-dependent neutrophil–endothelial cell interactions that mimic those of highly cytokine-activated endothelium. J Leukoc Biol 2005; 77:33– 43.
- 28 Xiao H, Schreiber A, Heeringa P, Falk RJ, Jennette JC. Alternative complement pathway in the pathogenesis of disease mediated by anti-neutrophil cytoplasmic autoantibodies. Am J Pathol 2007; 170:52–64.
- 29 Lee J, Cacalano G, Camerato T, Toy K, Moore MW, Wood WI. Chemokine binding and activities mediated by the mouse IL-8 receptor. J Immunol 1995; 155:2158–64.
- 30 Mantovani A. The chemokine system: redundancy for robust outputs. Immunol Today 1999; 20:254–7.
- 31 Zhou Y, Huang D, Farver C, Hoffman GS. Relative importance of

CCR5 and antineutrophil cytoplasmic antibodies in patients with Wegener's granulomatosis. J Rheumatol 2003; **30**:1541–7.

- 32 Bird JE, Giancarli MR, Kurihara T *et al.* Increased severity of glomerulonephritis in C-C chemokine receptor 2 knockout mice. Kidney Int 2000; **57**:129–36.
- 33 Lloyd CM, Minto AW, Dorf ME *et al.* RANTES and monocyte chemoattractant protein-1 (MCP-1) play an important role in the inflammatory phase of crescentic nephritis, but only MCP-1 is involved in crescent formation and interstitial fibrosis. J Exp Med 1997; 185:1371–80.
- 34 Panzer U, Thaiss F, Zahner G *et al.* Monocyte chemoattractant protein-1 and osteopontin differentially regulate monocytes recruitment in experimental glomerulonephritis. Kidney Int 2001; 59:1762–9.
- 35 Tam FW, Sanders JS, George A *et al.* Urinary monocyte chemoattractant protein-1 (MCP-1) is a marker of active renal vasculitis. Nephrol Dial Transpl 2004; 19:2761–8.
- 36 Cockwell P, Chakravorty SJ, Girdlestone J, Savage CO. Fractalkine expression in human renal inflammation. J Pathol 2002; 196:85– 90.
- 37 Feng L, Chen S, Garcia GE *et al.* Prevention of crescentic glomerulonephritis by immunoneutralization of the fractalkine receptor CX3CR1 rapid communication. Kidney Int 1999; 56:612–20.