

CHEMOKINES AND THEIR ASSOCIATION WITH PAEDIATRIC DISEASES

Fahad Abdullah Al-Zamil¹

ABSTRACT

The knowledge about chemokines and their receptors activity is an essential need of health care professionals. Chemokines represent a superfamily of small, inducible, secreted, proinflammatory cytokines involved in a variety of function in leukocyte trafficking, recruiting, and recirculation. The purpose of this review is together and highlights the different studies concerning with chemokines and their potential effects with various diseases. Chemokines play a significant role in many pathophysiological processes such as inflammation, allergic responses, autoimmune diseases, angiogenesis and tumor growth. In addition, particular chemokines have been reported to act on hematopoietic progenitor cells, influence wound healing and have a close association between their receptors and certain human diseases. It is important to know the biological characteristics of chemokines and their receptors activity. It has been also observed that there is a close association between chemokines, chemokine receptors, cells and certain adult and paediatric diseases. In addition, chemokines play a significant role in many pathophysiological processes such as inflammation, infectious, allergic response and autoimmune diseases.

For this review we reviewed English-language MEDLINE publications from 1966 through January 2006 for experimental, observational and clinical studies having relation with chemokines with different diseases. Approximately 72 publications were reviewed based on the relevance, strength and quality of design and methods, 54 publications were selected for inclusion.

KEY WORDS: Chemokines, classification of chemokines, chemokine receptors, paediatric diseases.

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INTRODUCTION

Throughout the late 1960's and 1970's, stimulated leukocytes have been shown to contain chemoattractants for monocytes and granulocytes. Over the past twelve years, many of

these chemoattractants and their receptors have been purified and cloned; this has revealed an expanding family of homologous chemotactic cytokines now known as chemokines. The chemokines are 8-16kDa soluble proteins produced and released by a wide variety of cell types during the initial phase of host response to injury, allergens, antigens, or invading microorganisms. Chemokines are a superfamily of mostly small, secreted proteins that function in leukocyte trafficking (Chemo attractants), recruitment and recirculation. They also play a significant role in many pathophysiological processes such as inflammatory, allergic and autoimmune disorders, angiogenesis and tumor growth.¹⁻³ In addition, certain chemokines have been reported to act on hematopoietic

1. Dr. Fahad Abdullah Al-Zamil MBBS, ABP
Department of Paediatrics,
College of Medicine, King Khalid University Hospital,
King Saud University, Riyadh, Saudi Arabia.

Correspondences:

Dr. Fahad Abdullah Al Zamil,
Associate Professor,
Department of Paediatrics (39),
College of Medicine,
King Khalid University Hospital,
P.O. Box 2925, Riyadh 11461.
Kingdom of Saudi Arabia
E-Mail: fahad_alzamil@hotmail.com

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progenitor cells and on non-leukocytic cells such as fibroblasts, smooth muscle cells, keratinocytes and melanoma cell lines.⁴

Identification: Chemokines were initially identified on the basis of their biological activities others were discovered using subtractive hybridization or signal sequence trap and cloning strategies. Members of the chemokine family exhibit from 20% - 90% identity in their predicted amino acid sequences.⁵

Types: Chemokines are classified into four main classes based on the conserved amino acid sequence motifs. Most family members have at least four conserved cysteine residues that form two intramolecular disulfide bonds. The families are defined by the position of the first two cysteine residues.

1. CXC Chemokines: The CXC chemokines, also called alpha (α) subfamily chemokines, they have one amino acid separating the first two cysteine residues. Within the CXC subfamily, the chemokines can be further divided into two groups. One group of the CXC chemokines have the characteristic three amino acid sequence ELR (glutamic acid-leucine-arginine) motif immediately preceding the first cysteine residue near the amino terminus. The second group of CXC chemokines lacks such an ELR domain. The CXC chemokines with the ELR domain act primarily on polymorphonuclear neutrophils as chemoattractants and activators and inducing neutrophil degranulation with release of myeloperoxidase and other enzymes. Presently there are five CXC-specific receptors and they are designated CXCR1 to CXCR5.

The ELR chemokines bind to CXCR2 and generally act as neutrophil chemoattractants and activators. In addition, the ELR chemokines that bind to CXCR3 - CXCR5, act primarily on lymphocytes. The CXC chemokines without the ELR domain, the CC chemokines and the C chemokines, chemoattract and activate the monocytes, T-lymphocytes, B-lymphocytes, basophils, eosinophils, dendritic cells and natural killer cells.^{6,7}

2. CC Chemokines: The CC Chemokines also called the beta subfamily, in this family the first two cysteines are adjacent to one another with

no intervening amino acid. There are currently 24 distinct human β-subfamily members. The receptors for this group are designated CCR1 to CCR11.²

3. C Chemokines: The C Chemokines also called the gamma subfamily. This family of chemokines is deficient in the first and third of the four conserved cysteine residues. Lymphotactin is the single member of the gamma class C chemokine, which has missing the first and third cysteines.⁸

Chemokines and human chromosome: The genes for all CC chemokines have been found to cluster on human chromosome 17q and mouse chromosome 11. Most of the CXC chemokines genes have been found to cluster on human chromosome 4q. The gene for the C chemokine lymphotactin have been localized to human chromosome 10q and 1. The clustering of chemokine genes suggests that many cytokine family members arose through gene duplication and subsequent divergence.⁹

Mechanism of action of Chemokines: The specific effect of chemokines on their target cells are mediated by members of a family of 7-transmembrane-spanning, G-protein-coupled receptors. These chemokine receptors are part of a much bigger superfamily of G-protein-coupled receptors that include receptors for hormones, neurotransmitters, paracrine substances, inflammatory mediators, certain proteinases, taste and odorant molecules, and even photons and calcium ions. The communication of chemokines with these G protein-linked receptors causes a rapid reconfiguration of adhesion proteins on the surface of the responding cells, facilitating their adhesion to endothelial cells lining blood vessel walls. Once the inflammatory leukocytes migrate along a gradient of increasing concentration of the chemokine to the site of origin. In response to the higher chemokine concentration at the site of injury or microbial invasion, the leukocytes are activated to perform effector functions such as release of their granule contents and increased production of cytokines.¹⁰

Chemokine receptors: Receptors play a significant role not only in the transmission of

message signals but also helps in different chemical and biological cellular activities. Chemokines mediate their biological activities by binding to target cell surface chemokine receptors that belong to the large family of G protein-coupled, seven transmembrane (7 TM) domain receptors also called serpentine receptors.^{11,12}

Structural features of chemokine receptors: Chemokine receptors identified membrane-bound molecules composed of 7-transmembrane domains and coupled to G-proteins. They measure approximately 350 amino acids in length and require the introduction of few gaps in the primary sequence to be aligned to other chemokine receptors.¹³ The chemokine receptors that bind CXC chemokines are designated CXCRs and the receptors that bind CC chemokines are designated CCRs. Five CXC chemokine receptors (CXCR-1 CXCR-5) and five CC chemokine receptors (CCR-1 CCR-5) have been cloned and characterized.^{10,11}

CXC RECEPTORS

CXCR 1: The CXCR1 receptor is also called interleukin IL-8RA, or type I IL-8 receptor. It contributes approximately 77% amino acid sequence identity with CXCR2.^{14,15} It binds with N-terminal Glu-Leu-Arg (ELR) containing CXC chemokines. CXCR1 is expressed on granulocytes, monocytes, mast cells and some CD8⁺ T-cells and CD56⁺ natural killer (NK) cells.^{16,17}

CXCR 2: The CXCR 2 also called IL-8RB or type II IL-8 receptor. It was cloned by Murphy and Tiffany¹⁴ and contributes to approximately 77% homology to CXCR1. CXCR-2 has broad specificity and has been shown to bind with high-affinity to other ELR motif containing a chemokines including GROa, GROb, GROg, NAP-2 and ENA-78.

CXCR 3: CXCR-3 receptor is also known as the IP-10 / Mig receptor. It has recently been cloned as a chemokine receptor that shares approximately 40% protein sequence identity with CXCR-1 and CXCR-2, and 34.2 - 36.9% amino acid sequence identity with the five known CC chemokine receptors.¹⁸⁻²⁰

CXCR-4: The CXCR-4 receptor is also known as fusin or LESTR and was cloned by Loetscher *et al.* (1994).²¹ It was subsequently identified as a necessary cofactor for entry of T cell-tropic HIV viruses into CD4⁺ cells.²² The CXC chemokine PBSF/SDF-1 has now been shown to be a ligand for CXCR-4 and a powerful inhibitor of infection by T cell-tropic HIV-1 strains.^{23,24} CXCR4 have biologic functions significantly different from those of other chemokines and chemokine receptors.^{25,26}

CXCR5: CXC chemokine receptor to be identified and has been renamed CXCR5. It is strongly expressed in Burkitt's lymphoma cells, B-lymphocytes and is found to have significant homology with other CXC chemokine receptors.²⁷

CX₃CR1: Fractalkine (CX3CL1) is a novel class of unusual member of the chemokine family that is synthesized with its chemokine domain at the end of a mucin-rich, transmembrane stalk. This membrane-bound localization allows fractalkine to function as an adhesion molecule for cells bearing its receptor, CX₃CR1. In addition, fractalkine can be proteolytically released from the cell surface, generating a soluble molecule that functions as a chemoattractant similar to the other members of the chemokine family.²⁸ Fractalkine attached or detached from its stalk, binds to CX₃CR1,²⁹ and promotes adhesion of monocytes, NK cells, and T lymphocytes to endothelial, epithelial and dendritic cells.³⁰

CC Receptors: CCR-1: The CCR 1 receptor was first identified as C-C chemokine receptor and is expressed on monocytes, neutrophils or eosinophils. It was first cloned by Neote *et al*³¹ with 33% similarity to CXCR2 and 31% to CXCR1. CCR-1 binds to MIP-1a, RANTES, MCP-3 with high affinity and also MIP-1b, and MCP-1 with lower affinity.³²⁻³⁴

CCR-2A and CCR-2B: The CCR-2A and CCR-2B are differ in their alternatively spliced carboxy-terminus and are probably spliced variants of a single gene.³⁵ CCR-2A and B specifically binds MCP-1 and MCP-3. Yamagami *et al.*,³⁶ identified CCR2B, which enclose a 360 amino acids with 56% similarity to CCR1.

CCR-3: It has 63% similarity to CCR1 and 51% to CCR2B and binds several CC chemokines specifically, including eotaxin, eotaxin-2, RANTES, MCP-3, MCP-4, and MIP-5. It is a high affinity receptor for eotaxin, an eosinophil-specific chemo-attractant. In humans CCR-3 was found to be expressed exclusively on eosinophils. Daugherty *et al.*,³⁷ and Kitaura *et al.*,³⁸ reported and characterized the CCR-3 as a eosinophil-selective chemokine receptor.

CCR 4: The CCR 4 was recognized by Power *et al* as an innovative CC chemokine receptor. It shares 49% identity with CCR1 and 47% with CCR2B. This receptor was originally cloned from a human immature basophilic cell line and has since been shown to be expressed in T cells and IL-5-primed basophils. CCR-4 has been shown to mediate the biological activities of RANTES, MIP-1a and MCP-1. It binds with CCR4 particularly binds the CC chemokines TARC (thymus and activation-regulated chemokine) and MDC (macrophage-derived chemokine).³⁹

TARC and MDC (Ligands for chemokine receptor CCR4): The thymus and activation-regulated chemokine (TARC) and macrophage-derived chemokine (MDC) are ligands for the chemokine receptor CCR4. A number of cellular sources of TARC and MDC have been identified, including macrophages, dendritic cells, natural killer cells and also bronchial epithelial cells.⁴⁰ TARC is a selective chemoattractant of Th2 cells and is a reasonable applicant while a key regulator of Th2-mediated inflammation in allergic asthma. In experimental models of allergic airway inflammation, it has been shown that neutralization of TARC can not only inhibit T-cell and eosinophil infiltration into the lung but can also inhibit bronchial hyperresponsiveness. In addition, Sekiya *et al.*, (2002)⁴¹ suggested that it could be involved not only in allergic asthma, but also in the pathogenesis of allergic Th2-mediated diseases in general.

CCR-5: CCR-5 receptor and was originally cloned by Samson *et al.*⁴² It has 48-75% amino acid sequence identity to the other CC recep-

tors. CCR-5 is expressed in primary adherent monocytes, but not in neutrophils or eosinophils. CCR-5 mediates the activities of MIP-1a, MIP-1b and RANTES. Recently CCR-5 has also been shown to be a co-receptor on CD4⁺ target cells for infection with primary, monocyte-tropic HIV-1 viruses.

CCR6: Initially CCR6 was cloned by several groups as an orphan chemokine receptor and has been detected on memory T cells, B-lymphocytes, and dendritic cells but not on any other peripheral blood leukocyte. CCR6 mRNA has been shown to be up regulated by treatment with IL-2.

CCR7: CCR7 is the novel CC chemokine also called secondary lymphoid tissue chemokine (SLC). CCR7 is known to be expressed on activated T and B lymphocytes and dendritic cells and is strongly up-regulated in B cells infected with Epstein-Barr virus and in T cells infected with herpesvirus 6 or 7.

CCR8: CCR8 mRNA expression was detected in the thymus and monocytes but not in other peripheral blood leukocytes. It expressed on Th2-polarized cells and is transiently increased after T-cell receptor and CD28 engagement.

CCR9: CCR9 was identified by Zaballos *et al.*,⁴³ reported that thymus-expressed chemokine (TECK) with specific agonist for the human orphan receptor GPR-9-6. CCR9 expression is high in the thymus but low in lymph nodes and spleen, and it appears to be expressed on both immature and mature T cells. TECK is an activator of dendritic cells and thymocytes which implicates this CC chemokine in T-cell development.

CCR10: CCR10 is also called a D6; it displays approximately 30% homology with other CC chemokine receptors at the amino acid level. Bonini *et al.*,⁴⁴ reported that D6 is to bind MCP-1 and MCP-3 with high affinity, whereas Nibbs *et al.*,⁴⁵ suggest that D6 is more promiscuous because it is able to bind a number of CC chemokines with similar affinity. D6 is almost exclusively expressed in placenta with weak expression in the liver, lung, and thyroid.

Chemokine and disease activity in children:

Meningococcal disease: Serum levels of

chemokines in patients with meningococcal sepsis can predict mortality and can correlate strongly with disease severity. Chemokines may play a key role in the pathophysiology of meningococcal disease and are potentially new targets for therapeutic approaches. Furthermore, Serum levels of CXC and CC chemokines in children in the initial phase of meningococcal sepsis can predict disease severity and outcome.⁴⁶

Pneumococcal Infection: Palaniappan et al., (2006)⁴⁷ conducted a study for understanding the requirements for protection against pneumococcal carriage and pneumonia and have been shown that genetic polymorphisms can result in diminished expression of CCL5, which results in increased susceptibility to and progression of infectious diseases. They showed that CCL5, together with Th cytokine mRNA expression, is temporally up-regulated during pneumococcal carriage and CCL5 is an essential factor for the induction and maintenance of protective pneumococcal immunity. Frezzolini et al., (2002)⁴⁸ suggested that IL-16 could serve as a useful marker of disease activity in childhood.

Bacterial enterocolitis: Yeung et al., (2004)⁴⁹ demonstrated that serum IL-6, IL-8 and CRP are significantly elevated in children with bacterial enterocolitis. IL-6 has a higher sensitivity, specificity and positive predictive value than IL-8 and CRP.

Newborn Cardio respiratory variables: Yanowitz et al., (2002)⁵⁰ demonstrated that chorioamnionitis was associated with increased IL-6 and IL-1 β concentrations in cord blood. Additionally, chorioamnionitis was also associated with increased newborn heart rate and with decreased mean and diastolic blood pressures. Cord blood IL-6 concentration correlated inversely with newborn systolic, mean, and diastolic blood pressures. Furthermore, infants with fetal vessel inflammation had higher IL-6 concentrations, were more likely to have elevated IL-1 β , and displayed increased right ventricular cardiac output when compared with infants without fetal vessel inflammation.

Cardiocirculatory arrest: Antonelli et al., (1999)⁵¹ showed that children who underwent cardiopulmonary bypass with cardiocirculatory arrest cytokine production seem related to duration of operation and amplified by ischemia-reperfusion phenomena.

Pollen allergy: Blanco et al., (1999)⁵² made a conclusions that serum IL-12 and sCD30 showed different behaviors in children with food or pollen allergy. They observed that IL-12 and sCD30 levels in pollen-allergic patients that agree with the classical T-helper (Th) 1/Th2 paradigm of allergy. In contrast, serum IL-12 levels were increased in food-sensitized children, suggesting a different immunologic pathogenesis.

Atopic dermatitis: La Grutta et al., (2005)⁵³ reported that Th2 profile predominance in the peripheral blood of children with atopic dermatitis and evidences close relationship between the number of CD4(+)IL-13(+) T cells and the disease's severity.

Asthma: Leung et al., (2002)⁵⁴ suggested that plasma TARC concentrations are elevated in childhood asthma. This marker is also linked to plasma total IgE levels and cat allergen sensitization.

CONCLUSIONS

In the field of medical sciences it is essential to know the biological characteristics of chemokines and their receptors activity. It has also been observed that there is a close association between chemokines, chemokine receptors, cells and certain paediatric diseases. In addition, chemokines play a significant role in many pathophysiological processes such as inflammation, infectious, allergic response and autoimmune diseases.

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