Soluble CD14 in Children with Status Asthmaticus

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Key words: asthma, soluble CD14, monocytes, macrophages

Abstract

Background: Inflammation is a major component in the pathogenesis of asthma. CD14 is an endotoxin (lipopolysaccharide) receptor, and is expressed mainly on monocytes and macrophages. Binding of LPS to CD14 activates the monocyte or macrophage and causes the release of different cytokines. The soluble form of CD14 is present in serum, and its concentration increases in several clinical conditions, including infections, autoimmune disorders, allergic disorders, and lung diseases. The possible role of CD14/sCD14 in asthma has been investigated in a few adult patients only.

Objectives: To measure serum concentrations of sCD14 in children with status asthmaticus.

Methods: We compared serum concentration of sCD14 in 10 children with status asthmaticus measured within 24 hours of admission and after recovery from the acute episode.

Results: Levels of sCD14 were significantly higher during acute asthma attacks than at recovery.

Conclusions: The elevated serum levels of sCD14 during status asthmaticus may be the result of the activation of monocytes, macrophages or other cells. The influence of medications on serum sCD14 cannot be ruled out. The possible use of sCD14 as a marker of lung inflammation in asthma warrants further investigation.

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Inflammation is a major component in the pathogenesis of asthma [1–3]. The release of chemical mediators by mast cells, eosinophils and other cells in the lungs is considered to play an important role in this process [3–6]. There is some evidence that monocytes and macrophages may also be involved in the inflammatory process of acute asthma, but the studies investigating the role of these cells in asthma are still scarce [7,8].

The myelomonocytic differentiation antigen CD14 is expressed mainly on monocytes and various tissue macrophages [9], However it is also present in small amounts on granulocytes and B-lymphocytes. It consists of a glycoprotein that is anchored to the cell membranes and serves as a receptor for endotoxins, the LPS component of Gram-negative bacteria cell walls [10].

Binding of LPS to CD14 induces macrophage activation with release of cytokines such as interleukin-6, IL-8 and tumor necrosis factor-α. CD14 antigen may also be involved in monocyte-T cell or monocyte-endothelial cell interactions [11,12].

A soluble form of CD14 has been detected in the supernatants of cultured CD14+ cells as well as in serum and urine [9]. It appears from *in vitro* studies that sCD14 levels increase concomitantly with the cell-surface expression of CD14 [9]. Although the exact physiological functions of CD14 antigen and sCD14 within the immunological network are still unclear, sCD14 binding to LPS may serve as a mechanism of down-modulation of monocyte activation to protect cells from endotoxin-induced activation [11]. There is indeed evidence that sCD14 competes with CD14 for LPS binding in Gram-negative sepsis and its serum level is increased [12]. Levels of sCD14 are also altered in several autoimmune disorders, allergic disorders, lung diseases, and infections [13–17]. However, the clinical usefulness of sCD14 serum concentration measurements still needs to be explored.

The aim of the present study was to investigate the serum level of sCD14 in children with status asthmaticus during an acute attack and on recovery, as a possible indicator of monocyte, macrophage or other cell activation in this condition.

Materials and Methods

Patients

The study population included 10 children (6 males, 4 females) aged 3–18 years (mean 10 years) admitted with status asthmaticus to the Department of Pediatrics. Status asthmaticus was defined as severe respiratory distress, despite repeated treatment with inhaled beta 2-agonist and oxygen saturation below 95% at room air, requiring oxygen administration. All patients had been receiving inhaled β2-agonist therapy on an as-needed basis. Two had also received preventive treatment with low dose budesonide (400 μg/day). No patient had clinical evidence of lower respiratory tract infection or fever

LPS = lipopolysaccharide
SCD14 = soluble form of CD14

(>38°C). Three patients had mild rhinorrhea and three had mild atopic dermatitis. Mean immunoglobulin-E level for the whole group was 1,450 IU/ml (range 67–6,500 IU/ml); levels were elevated in seven patients and within normal range in three (<100 IU/ml).

On admission, the patients were treated with inhaled solbutamol 2.5 mg 6 to 8 times a day and intravenous methylprednisolone 2 mg/kg/day in two divided doses, the first of which was given immediately after admission. Recovery from the acute state occurred within 48–72 hours in eight patients and within 96 hours in two. Recovery was defined as lack of respiratory distress by clinical judgment, oxygen saturation ≥95% in room air, and no need for intravenous corticosteroid treatment.

Methods

Measurements of sCD14 levels were done within 24 hours of admission after the first methylprednisolone dose, and at recovery. Blood samples were collected, and the sera were separated and kept at -20°C until the assay. The sera taken during the acute and convalescent stages were tested simultaneously. Levels of soluble CD14 were determined by sCD14-EASIA ELISA (Medgenix Diagnostica, SA, Belgium); optical density was assessed by an ELISA reader.

Results

Levels of sCD14 were significantly higher during the acute asthma attack than at recovery ($P<0.02$) [Figure 1]. The reduction in sCD14 was very prominent in seven patients, and mild in one; in two patients, the sCD14 level mildly increased in the convalescent stage. There was no correlation between level of sCD14 or the change in sCD14 and serum IgE concentration.

Discussion

Activation of monocytes by different agents, including endotoxins, interferon-γ or anti-CD14 antibodies, results in the shedding of sCD14 into the surrounding medium [18]. Alterations in sCD14 are known to occur in various clinical situations. Serum sCD14 levels are reduced after polytrauma and increased on recovery [19]. In patients with human immunodeficiency virus infection, sCD14 serum levels rise with disease progression and correlate with the decrease in the CD_4/CD_8 T cell ratio [16,17]. Likewise, patients with systemic lupus erythematosus have higher levels of serum sCD14 than controls, and patients with active SLE have higher levels than do patients in remission [14]. Since there is a good correlation between sCD14 and C3 serum concentrations, but not with dsDNA titers, some authors have suggested that serum sCD14 may serve as a parameter of disease activity in SLE [14]. In addition, serum level of sCD14 is elevated in septic shock [20], and higher levels are associated with a higher mortality in patients with Gram-negative sepsis [21]. The explanation of these findings is controversial.

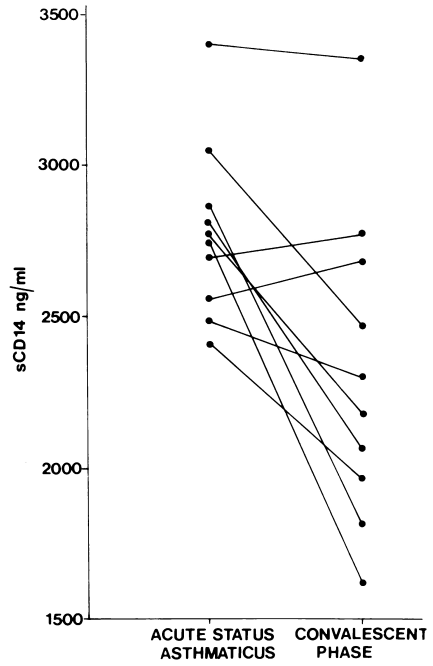

Figure 1. Serum sCD14 concentration in children during acute status asthmaticus and in the convalescent phase

Elevated serum sCD14 levels were reported in psoriasis [22], atopic dermatitis [15], and extrinsic allergic alveolitis [23]. In extrinsic allergic alveolitis, which is caused by an immune response to inhaled allergens, CD14 expression on alveolar macrophages was found to increase concomitantly with an increase in serum sCD14, suggesting that sCD14 may be released by alveolar macrophages into the circulation. Allergen avoidance decreased sCD14 levels, while allergen exposure increased them [23]. Elevated levels of sCD14 have been found in bronchoalveolar lavage fluid from patients with pulmonary diseases, such as tuberculosis, sarcoidosis and idiopathic pulmonary fibrosis [9,24,25], possibly reflecting an increased pulmonary monocyte/macrophage activation.

The involvement of monocytes and macrophages in asthma is supported by several observations. A significant increase in the number of macrophages in the airways was found 48 hours after antigen provocation [7]. It has been suggested that macrophages and monocytes participate directly in allergic responses by virtue of their IgE receptors ($FcER_2$) [26]. $FcER_2$ expression in peripheral blood monocytes is enhanced in asthmatic patients during wheezing attacks, exercise, or allergen provocation [26,27]. BAL macrophages from asthmatic patients have an enhanced capacity to release β-glucuronidase, leukotriene B_4, and prostaglandin $F_{2\alpha}$ in response to IgE stimulation *in vitro* [27]. Macrophages or monocytes may also interact with other cells involved in the inflammatory process. Activated neutrophils primed by supernatants of

SLE = systemic lupus erythematosus

BAL = bronchoalveolar lavage

peripheral blood monocytes from asthmatic subjects generated approximately three times more leukotriene B_4 than neutrophils primed by supernatants of monocytes from normal subjects [7,28]. Stimulated eosinophils incubated with supernatants from the alveolar macrophages of asthmatic subjects secreted more leukotriene C_4 and platelet-activating factor than eosinophils incubated with supernatants from alveolar macrophages of normal individuals [29].

In a study of eight adults with allergic asthma, stimulation by the inhalation of specific allergens yielded an increase in sCD14 in the BAL fluid at 18 hours after provocation (but not after 10 minutes) [30]; sCD14 levels remained unchanged when NaCl was inhaled. Interestingly, after 18 hours, a correlation was found between the sCD14 level and the number of granulocytes and eosinophils, but not with the number of macrophages. These findings may suggest that sCD14 is released locally, at the site of allergen stimulation, and that one source of sCD14 may be pulmonary granulocytes [30]. Increased sCD14 in BLA was obtained by segmental ragweed antigen challenge in 11 asthmatic adults with ragweed allergy [31].

The results of the present study show that the serum sCD14 level is elevated in children during status asthmaticus compared to the convalescent period. This finding may further support the assumption that monocytes or macrophages, the main source of sCD14 in the serum, are involved in the inflammatory process of status asthmaticus. However, it is also possible that some of the serum sCD14 in acute asthma may be due to activation of neutrophils or eosinophils [30]. In addition, all our patients were treated with inhaled β-agonists and methylprednisolone. Corticosteroids may reduce the number of monocyte $Fc_\gamma R_1$ and complement receptors, thereby suppressing cell activation [27]. Thus, the reduction of sCD14 in the serum of asthmatic patients during convalescence may have been partially related to the action of the large doses of corticosteroid administration during the acute state. Further studies are needed to differentiate the possible effects of treatment on serum concentration of sCD14.

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Be nice to people on the way up because you'll need them on your way down.

Wilson Mizner

Capsule

TT virus, HIV and HCV

The search for the cause of chronic hepatitis among individuals with non-A to G hepatitis has led to the discovery of a post-transfusion hepatitis-related DNA virus, designated TT virus, which, based on viral sequences, belongs to a new virus family. The principal modes of infection with TTV are poorly understood, and its role in human immunodeficiency virus type 1 (HIV-1) infection is unclear. The study objective was to determine if injection drug use and high risk heterosexual activity, principal modes of acquiring HIV1 infection, place individuals at greater risk of acquiring TTV. The authors analyzed DNA, extracted from sera or filter paper-blotted whole blood, obtained during August 1997 and June 1998 from 324 Vietnamese subjects (148 males, 176 females), for TTV sequences by hot-start, heminested polymerase chain reaction. The results showed a prevalence of TTV viremia that was similar among individuals engaging in injection drug use or high risk heterosexual activity (23.4% vs. 20.2%), with no age- or gender-specific differences. No association was found between TTV viremia and co-infection with HIV-1 or hepatitis C virus (HCV). Phylogenetic analysis of 30 TTV sequences revealed two distinct genotypes and four subtypes that did not segregate according to gender, HIV-1 and HCV risk behaviors, or geographic residence, suggesting that the usual route of TTV transmission in Vietnam is other than parenteral or sexual.

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