Paediatrica Indonesiana

VOLUME 52 January • 2012 NUMBER 1

Original Article

Correlation between C-reactive protein and serum iron levels in children with pneumonia

I Gd. Oka Novi Purnawan[1], Ida Bagus Subanada[1], Sri Mulatsih[2]

Abstract

Background Pneumonia is an infectious disease often occuring in children under five years of age. At the time of infection, pro-inflammatory cytokines are released. It is thought that these pro-inflammatory cytokines cause changes to iron homeostasis in the body.

Objective To determine a correlation between CRP and serum iron levels in children with pneumonia.

Methods An analytical, cross-sectional study was performed in children aged 6 months-5 years with severe pneumonia at Sanglah Hospital, Denpasar, Bali from April-November 2010. Laboratory examinations included CRP and serum iron levels. The correlation between CRP and serum iron levels was analyzed by Pearson's correlation.

Results From 69 children with severe pneumonia, 23 children fulfilled the inclusion criteria. Subjects' median CRP level was 9.22 mg/L and median serum iron level was 25.55 ug/dL. The coefficient correlation between CRP and serum iron levels was -0.580 (P=0.004). The determination coefficient value was 0.316.

Conclusion In children with severe pneumonia, CRP level correlates negatively with serum iron levels. **[Paediatr Indones. 2012;52:38-42]**.

Keywords: *CRP, serum iron, pneumonia*

Pneumonia is an infectious disease that often occurs in children under five years of age. In developing countries, pneumonia causes a high morbidity and mortality rates.[1] During infection, cytokines such as pro-inflammatory interleukin-1 (IL-1), interleukin-6 (IL-6) and tumor necrotizing factor-α (TNF-α) are released. It is thought that these pro-inflammatory cytokines may lead to a change in iron homeostasis in the body.[2,3] Acute phase proteins, as products of pro-inflammatory cytokine release, are often evaluated as an indirect measure for these cytokines.

During acute infections, C-reactive protein (CRP), an acute phase protein, increases in concentration in the blood as a non-specific immune response. Induction of CRP synthesis is triggered by cytokines, especially IL-6, that are released in areas of inflammation. Laboratory examination of CRP concentration is often performed in patients with pneumonia.[4]

From the Department of Child Health, Medical School ,Udayana University, Sanglah Hospital, Denpasar, Indonesia.[1] Department of Child Health, Medical School, Gadjah Mada University, Dr. Sardjito Hospital, Yogyakarta, Indonesia.[2]

Reprint requests to: I Gd Oka Novi Purnawan, Department of Child Health, Medical School, Udayana University, Sanglah Hospital, Jalan Pulau Nias, Denpasar 80114, Indonesia. Tel. +62-361-244034, Fax. +62-361-244034. E-mail: *gedeokanovi@yahoo.com*

In Serbia, Nastasijevic *et al*.[5] found iron deficiency in adult patients suffering from community-acquired pneumonia. Their serum iron levels showed significant improvement after 6-8 days of therapy, despite the absence of iron supplementation, and CRP levels correlated negatively with serum iron levels. There have been few similar studies performed in children. The aim of our study was to determine if there is a correlation between CRP and serum iron levels in children with pneumonia.

Methods

This cross-sectional, analytical study was conducted at the Pulmonology Division, Department of Child Health, Udayana University Medical School, Sanglah Hospital, Bali from April to November 2010.

The number of required subjects was calculated using the single correlation test sample. We included 23 subjects with severe pneumonia aged 6 months - 5 years. We excluded patients with: 1) very severe pneumonia or severe pneumonia with concomitant systemic diseases, post-surgery and burns; 2) a history of preterm birth, low birth weight, and twin pregnancies; 3) food absorption problems; 4) anemia/ previous iron therapy; 5) incomplete data; and 6) lack of parent/guardian consent. This study was approved by the Research Ethics Committee of Sanglah Hospital, Denpasar.

Clinical examinations were performed by doctors in the emergency unit, outpatient ward, and the Pulmonology Division, Department of Child Health, Udayana University Medical School, Sanglah Hospital, Denpasar. Venous blood specimens for complete blood counts, CRP, and serum iron examinations were collected at the time patients were diagnosed with severe pneumonia. Serum iron concentration was measured by Beckman Coulter CX 7 using spectrophotometry. CRP concentration was measured by COBAS INTEGRA 400. Statistical analyses were performed using SPPS 15.0 version for windows. Pearson's correlation was used to analyze the association between CRP and serum iron levels.

Results

During the study period, of the 69 children with pneumonia, 23 children were included in our study. Among the excluded 46 children, 5 children had very severe pneumonia, 27 children had anemia (hemoglobin <11 g/dL), 3 children had a history of preterm birth and low birth weight, and 11 children had other systemic diseases (4 children with congenital heart disease, 1 child with liver disease, 1 child with a malignancy, 2 children with brain infections, 1 child with post-laparotomy surgery, and 2 children with sepsis).

The median concentration of CRP was 9.22 mg/L (range 0.22 to 53.5) and the median serum iron concentration was 25.55 μg/dL (range 9.13 to 76.46), as shown in **Table 1**.

Table 1. Basic characteristics of subjects

Subject characteristics	n = 23
Median age, months (minimum-maximum)	13.0 (6.0-58.0)
Sex	
Male	13
Nutritional status	
Well-nourished	10
Undernourished	8
Overweight	5
At risk of iron deficiency due to diet	23
Median laboratory parameters, (minimum-maximum)	
Hemoglobin,g/dL	11.8 (11-13.9)
WBC, $x10^3$ µ/L	11.2 (6.09-29.2)
Absolute neutrophil, $x10^3$ µ/L	5.74 (1.82-20.8)
CRP, mg/L	9.22 (0.22-53.5)
Iron serum, µg/dL	25.55 (9.13-76.46)
Median time of CRP and serum iron measurement after symptom onset, days (minimum-maximum)	4.0 (2.0-7.0)

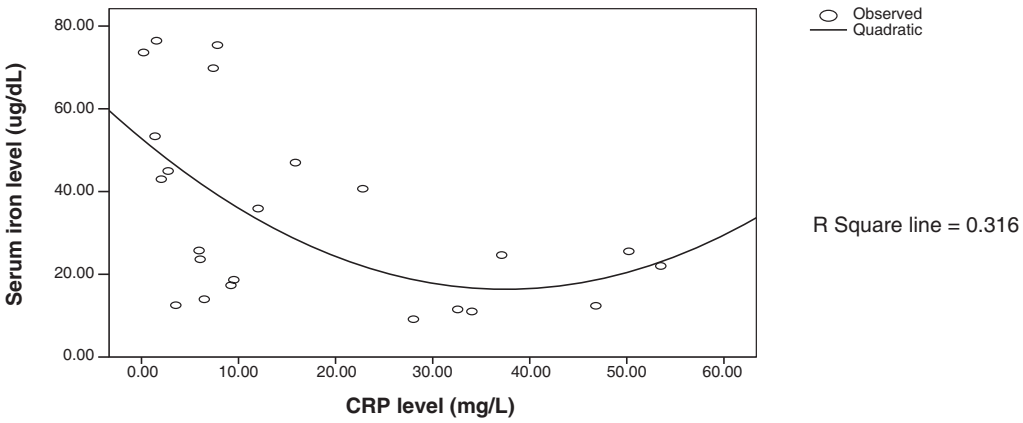

Figure 1. Scatter plot correlation of CRP and serum iron levels in children with severe pneumonia.

The Shapiro-Wilk test revealed abnormal data distribution in CRP and serum iron levels. To normalize this data distribution, we performed data transformation using the log10 method. Pearson's correlation test revealed a weak but statistically significant, negative correlation between CRP and serum iron levels ($r = -0.580$; $P = 0.004$). If CRP levels increased, serum iron levels decreased, and vice versa.

The relationship between CRP and serum iron levels is shown in **Figure 1**. Scatter plots showed that the relationship between CRP and serum iron levels was non-linear. The quadratic model was statistically significant ($F = 4.620$, $df = 20$, $P = 0.022$) and more precise than linear model. Scatter plot also showed the negative relationship between CRP and serum iron levels in children with severe pneumonia. The determination coefficient value was 0.316.

Discussion

In pneumonia patients, infection and tissue damage in the lung parenchyma may stimulate the immune system to release CRP into the bloodstream. Increased CRP levels are often seen in cases of pneumonia due to bacterial infection. Hence, CRP testing is useful to distinguish bacterial infections from other causes of infections. Increased CRP levels can also be found in viral infections such as influenza A and influenza B, with CRP levels ranging between 10-40 mg/L.[6,7] Other studies reported elevated levels of CRP > 12 mg/L in cases of pneumonia accompanied by empyema and necrotizing pneumonia, while average CRP concentrations in adenovirus infections can reach 6.71 ± 4.99 mg/L.[8,9]

In our study, the median CRP concentration was 9.22 mg/L, ranging from 0.22 - 53.5 mg/L, similar to that of previous studies.[6,7,9] These CRP levels suggest that the cause of pneumonia in most of our subjects was viral. Low CRP levels are also likely to be detected if CRP examination is performed after the subject has passed the peak increase stage of CRP. The average CRP examination time in our study was 4 days after onset of initial pneumonia symptoms, such as fever, cough and runny nose. In theory, CRP levels reach the lowest value at days 4 and 5 after stimulation.[12]

The inflammatory process may disrupt iron homeostasis. Impaired iron homeostasis begins with increased uptake and retention of iron in reticuloendothelial cells. Iron from the circulation is stored in the reticuloendothelial system, where its use is limited to stem cell erythropoiesis. Stimulation of pro-inflammatory cytokines (TNF-α, IL-1, IL-6) leads to the retention of iron in macrophages. Furthermore, down-regulation of ferroportin production withholds the release of ferrous iron from duodenum enterocytes into the circulation.[3] Kemna *et al*. found that after

humans were injected with lipopolysaccharide, serum iron levels gradually decreased, especially in the first 6 hours, and reached the lowest level after 22 hours.[13]

The median serum iron concentration of in our subjects was 25.55 μg/dL, ranging from 9.13 - 76.46 μg/dL, lower than that of a Serbian study which reported a mean serum iron level in adult pneumonia patients of 4.89 ± 3.23 μmol/L (27.38 ± 18.0 μg/dL).[5] This difference may be due to subjects' age differences. The median age of our subjects was 13 months, an age vulnerable to iron deficiency,[14] while the Serbian subjects were adults.[5]

A study of serum iron profiles reported mean serum iron concentrations in children with severe malnutrition to be 63.5 ± 0.97 μg/dL, in severely malnourished children with no acute respiratory infection to be 35.91 ± 0.70 μg/dL, and in severely malnourished children with acute respiratory infection to be 31.9 ± 1.35 μg/dL.[15] All of their results were higher than that in our study (25.55 μg/dL). This difference may be due to less severe acute respiratory infections in their study (upper acute respiratory infections), compared to acute lower respiratory infections in our study.[15]

Iron homeostasis disruption occurs during the inflammatory process.[3] In our study, we determined the degree of inflammation by measuring CRP levels, as an end result of pro-inflammatory cytokine stimulation, especially IL-6. We determined iron homeostasis disruption by measuring serum iron levels. We found a negative correlation between CRP and serum iron levels in children with severe pneumonia. The correlation coefficient was -0.580 ($P = 0.004$), indicating that increased CRP levels in children with pneumonia correlated to decreased serum iron levels. These results were consistent with a Serbian study which showed that serum iron levels negatively correlated with CRP levels ($r = -0.625$, $P < 0.01$).[5]

There was a positive relationship between CRP and serum iron levels after CRP concentration exceeded 40 mg/L, as shown by the ascending quadratic line. This condition may have been due to varying times of CRP and serum iron testing. The most rapid examination was at 2 days and the least was at 7 days after presentation of early clinical symptoms of pneumonia.

We found the correlation strength to be weaker than that of the Serbian study, which was of medium strength. This difference may be due to different etiologies of pneumonia. In Serbia, most infections were bacterial, while most infections in our study were likely to be viral. A limitation of our study is that we did not perform virus isolations or polymerase chain reaction (PCR) tests to determine pneumonia etiology. We only performed blood cultures, not lung tissue or pleural fluid. Blood cultures were sterile for all subjects. Blood culture results do not rule out the possibility that bacteria caused our subjects' pneumonia, since there is generally a low incidence of bacteremia during pneumonia.

Another limitation of our research was that CRP and serum iron levels were not measured after initiation of therapy. So we do not know if serum iron spontaneously increased after therapy. Since the testing times of CRP and serum iron levels varied, the data distribution was abnormal. Also, we did not account for the influence of diurnal variation on serum iron levels. Furthermore, all subjects were at risk for iron deficiency due to their dietary status. Since serum ferritin was not examined in our study, we do not know if the low serum iron levels in our subjects were caused by infection alone, or influenced by low iron reserves.

In conclusion, CRP level was negatively correlated with serum iron level in children with severe pneumonia. Further research is needed to evaluate iron status, including ferritin examinations, in order to provide more specific information on the impact of lower respiratory tract infection on iron metabolism.

Acknowledgments

We are deeply grateful to I Gde Raka Widiana, MD and IB Subanada, MD for their help in constructing methodology and statistical analysis in this study.

References

1. Bryce J, Boschi-Pinto C, Shibuya K, Black RE. WHO estimates of the causes of death in children. Lancet. 2005;365:1147-52.
2. Walter T, Olivares M, Pizarro F, Munos C. Iron, anemia, and infection. Nutr Rev. 1997;55:111-24.

3. Weiss G, Goodnough LT. Anemia of chronic disease. N Engl J Med. 2005;352:1011-23.
4. Kratz A, Lewandrowski EL. The plasma protein. In: Kenneth DM, editor. Clinical laboratory medicine. 2nd ed. Philadelphia: Lippincott Williams & Wilkins; 1997. p. 264-7.
5. Nastasijevic DB, Pejcic T, Radjenovic T, Radovic M. Inverse correlation of CRP and serum iron in patients with simple community-acquired pneumonia (CAP). Acta Fac Med NAISS. 2006;24:107-12.
6. Luna CM. C-reactive protein in pneumonia: let me try again. Chest. 2004;125:1192-5.
7. Melbye H. Point of care testing for C-reactive protein. J Fam Physic. 2006;35:313-5.
8. Lin CJ, Chen PY, Huang FL, Lee T, Chi CS, Lin CY. Radiographic, clinical, and prognostic features of complicated and uncomplicated community-acquired lobar pneumonia in children. J Microbiol. 2006;39:489-95.
9. Farng KT, Wu KG, Lee YS, Lin YH, Hwang BT. Comparison of clinical characteristics of adenovirus and non-adenovirus pneumonia in children. J Microbiol Immunol Infect. 2002;35:37-41.
10. Duga MK, Arora N, Prakash K, Jhamb R, Kumar N, Gupta N. C-reactive protein in lower respiratory tract infection. Lung India. 2005;22:41-4.
11. Almirall J, Bolibar I, Toran P, Pera G, Boquet X, Balanzo X, et al. Contribution of C-reactive protein to the diagnosis and assessment of severity of community-acquired pneumonia, for the community-acquired pneumonia. Chest. 2004;125:1335-42.
12. Gabay C, Kushner I. Acute phase protein and other systemic responses to inflammation. N Engl J Med. 1999;24:448-56.
13. Kemna E, Pickkers P, Nemeth E, van der Hoeven H, Swinkels D. Time-course analysis of hepcidin, serum iron, and plasma cytokine levels in humans injected with LPS. Blood. 2005;106:1864-6.
14. Brotanek JM, Gosz J, Weitzman M, Flores G. Iron deficiency in early childhood in the United States: risk factors and racial/ethnic disparities. Pediatrics. 2007;120:568–75.
15. Rahman MA, Mannan MA, Rahman MH, Aretin KE, Azad AK. Influence of acute respiratory infection on iron profile in severely malnourished children. Mymensingh Med J. 2008;17:S52-8.