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The Clinical Utility of Serum YKL-40 Levels in Community Acquired Pneumonia

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Abstract

Introduction: We aimed to investigate the changes in blood levels of YKL-40 in patients hospitalized with a diagnosis of community acquired pneumonia (CAP) before treatment and on the 7th day of treatment and to determine whether this can be used as a diagnostic and prognostic marker in the disease.
Methodology: Sixty-two subjects including 40 with CAP and 22 healthy as a control group were enrolled to the study. Serum YKL-40 levels were measured in patients with CAP before treatment and on the seventh day of the treatment. Degrees of severity of pneumonia were evaluated according to CURB-65 and the Pneumonia Severity Index (PSI).
Results: Mean serum YKL-40 levels of 89.24±98.67 ng/ml and 74.37±56.28 ng/ml were measured on the 1st and 7th days, respectively. The difference between two measurements was significant (p=0.003). A significant difference was also determined in serum YKL-40 level between control group and patient with CAP group on 1st and 7th days (p=0.001 and p<0.001, respectively). PSI and CURB-65 scores were not correlated with serum YKL-40 levels in patients with CAP.
Conclusion: The results show higher blood YKL-40 levels in patients in the CAP group compared to the controls. Elevated YKL-40 levels in blood specimens at the start of treatment in our pneumonia group, followed by a decrease one week later, may be regarded as evidence that blood YKL-40 levels can be used as an inflammation marker in clinical practice.

Keywords: Pneumonia; YKL-40; C-reactive protein

Introduction

Community-acquired pneumonia (CAP) is an acute lower respiratory tract infection characterized by clinical features of lower respiratory tract infection with new radiological shadowing in the lungs in a person who has not

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been hospitalized recently. Aging, existence of co-morbidities, living in a crowded environment, malnutrition, immunization deficiency, HIV infection and exposure to smoking constitute risk factors for development of CAP (1). While a decrease has been observed in the death rates of infectious diseases due to the effectiveness of modern vaccination programs, the development of new antibiotics and the widespread use of antibiotics, CAP still exhibits high levels of morbidity and mortality (2). Pneumonia stands in 6th place in causes of death in the USA and Great Britain, and in first place among deaths of infectious origin. Approximately 5.6 million patients are diagnosed with CAP in the USA each year, of which 1.1 million require hospitalization. Proportional to severity of the disease, mean mortality in patients treated on an outpatient basis is 1-5%, 12% in those hospitalized for treatment and rises to 40% in patients treated in intensive care (3-6).

Diagnostic and prognostic approaches depending on inflammatory biomarkers such as C-reactive protein (CRP), procalcitonin, and cytokines have been further investigated in CAP (7). Promising results have also been obtained by novel biomarkers such as pro-adrenomedullin, pro-vasopressin, and YKL-40 in daily clinical practice in the management of CAP (7,8). Serum YKL-40 is a glycopeptide largely released from chondrocytes, osteoblasts, macrophages, neutrophils, epithelium and smooth muscle cells, this secretion increasing in systemic inflammation (9-11). Human YKL-40 possesses a similar amino acid sequence to bacterial chitinases and exhibits enzymatic activity, as in bacteria (12,13). Serum YKL-40 has previously been investigated in asthma, COPD, sarcoidosis, pulmonary tuberculosis, cystic fibrosis, lung cancer and pneumonia, and various studies have shown that it increases in association with the activity or severity of these and other similar diseases with an inflammatory course (14). The purpose of this study was to investigate changes in blood levels of YKL-40, previously shown to rise in various diseases of an inflammatory nature, in patients hospitalized with a diagnosis of CAP before treatment and on the 7th day of treatment and to determine whether this can be used as a diagnostic and prognostic marker in the disease.

Materials and methods

Study Design and Patients

Recep Tayyip Erdoğan University Faculty of Medicine ethical committee approval was obtained before the study began. The study was performed at Recep Tayyip Erdoğan University Faculty of Medicine Chest Diseases Clinic between 1 April and 1 December, 2012. The design was prospective, observational and descriptive. Forty patients hospitalized with a diagnosis of CAP and 22 healthy controls were included. Written informed consent was obtained from all participants. Patients' total blood count, blood biochemistry analyses and arterial blood gases were investigated. Chest x-rays were taken. Diagnosis of CAP was based on Infectious Diseases Society of America (IDSA, Arlington, VA, USA) and American Thoracic Society (ATS, New York, NY, USA) guidelines. Infiltrative changes at chest x-ray or at least one clinical finding, such as thick, yellow phlegm, coughing or fever (>37.8 oC), or at least two minor criteria, such as tachypnea, dyspnea, pleural pain, chest pain, confusion and impaired orientation, consolidation in the lungs or WBC count >12,000 cells/microL were determined as criteria for CAP. Degrees of severity of pneumonia were calculated according to CURB-65 (Table 1) and the Pneumonia Severity Index (PSI) (Table 2). Treatments were arranged empirically. Venous

Parameter	Point
1. Confusion	+1
2. Urea >42.8 mg/dL, (BUN >20 mg/dL)	+1
3. Respiratory rate ≥30/min	+1
4. Blood pressure (Systolic <90 mmHg or Diastolic ≤ 60 mmHg)	+1
5. Age ≥ 65 year	+1

Table 1: CURB-65 scores

blood specimens were collected from the pneumonia group on the 1st and 7th days of treatment and from the healthy control group. White blood cell (WBC), C- reactive protein (CRP) and erythrocyte sedimentation rate (ESR) tests were performed on patients on the 1st and 7th days. Serum YKL-40 levels were investigated separately for each case in the patient and control groups. Patient and control group demographic characteristics, comorbidities, pneumonia symptoms and findings and laboratory results were recorded.

Characteristics	Point
Demographics	
Male	Age (years)
Female	Age (years) − 10
Nursing home resident	+ 10
Comorbid illness	
Neoplastic disease	+ 30
Liver disease	+ 20
Congestive heart failure	+ 10
Cerebrovascular disease	+ 10
Renal disease	+ 10
Physical examination findings	
Altered mental status	+ 20
Respiratory rate > 30 breaths per minute	+ 20
Systolic blood pressure < 90 mm Hg	+ 20
Temperature < 35˚C (95˚F) or > 40˚C (104˚F)	+ 15
Pulse rate > 125 beats per minute	+ 10
Laboratory and radiographic findings	
Arterial pH < 7.35	+ 30
Blood urea nitrogen > 64 mg per dL (22.85 mmol per L)	+ 20
Sodium < 130 mEq per L (130 mmol per L)	+ 20
Glucose > 250 mg per dL (13.87 mmol per L)	+ 10

Hematocrit < 30 percent		+ 10
Partial pressure of arterial oxygen < 60 mm Hg	or	+ 10
oxygen percent saturation < 90 percent		
Pleural effusion		+ 10

Table 2: Pneumonia Severity Index (PSI)

Exclusion Criteria:

Outpatients, patients who had attended a different hospital within the previous 3 weeks or had been transferred from a different hospital, who had been started on antibiotic therapy before presentation, and patients with coronary artery disease, pulmonary edema, pulmonary embolism, kidney failure, liver failure, malignity, severe immune deficiency such as severe neutropenia (WBC count 1.0 × 109 cells /L), with organ or tissue transplant or with HIV infection were excluded.

YKL-40 measurement:

Venous blood specimens were centrifuged for 10 min at 2500 rpm. Serum was separated and stored in a freezer at -80 C. Serum levels of human Chitinase-3-like Protein 1 were quantified with enzyme-linked immunosorbent assay (ELISA) using commercially available matched antibodies (MyBiosource, San Diego, USA). The intra- and inter-assay coefficients of variation (CV) were <10.0% and <12.0%, respectively. Sensitivity was calculated at 0.52 ng/ml.

Arterial blood gas analysis:

Arterial blood gas was collected from the radial artery after the patient had rested in a sitting position at room temperature for 15 min. Blood gas analysis was performed immediately using a blood gas device (RAPIDLYLab 248/348 system, Siemens AG Healthcare, Germany).

Statistical Analysis:

Data analysis was performed on SPSS (SPSS version 15; SPSS Inc., Chicago, IL, USA) software. Relations between variables were investigated using Pearson correlation analysis and those between groups using Wilcoxon test correlation analysis. Significance was set at $P<0.05$.

Results

Sixty-two subjects, 40 with CAP and 22 from the control group, were enrolled. Demographic features, clinical and laboratory findings of the patients with pneumonia and control group were shown in Table 3. Demographic analysis revealed that the groups were similar in terms of gender and level of cigarette use. Mean YKL-40 levels in blood specimens taken from the pneumonia group was 89.24±98.67 ng/ml on the 1st day and 74.37±56.28 ng/ml on the 7th day. The difference between the two measurements was significant (p=0.003). The control group YKL-40 level was 47.44±7.16 ng/ml. YKL-40 and other inflammatory markers are studied in first and seventh day of treatment and shown in Table 4. A significant difference was determined between control group YKL-40 level and the levels in blood specimens from the 1st and 7th days (p=0.001 and p<0.001). CRP levels in blood specimens from the pneumonia group were 22.2±12.95 mg/dL on the 1st day and 5.78±5.6 mg/dL on the 7th day. The difference in CRP levels between the two groups was also significant (p<0.001).

Blood specimen WBC counts in the same groups were 23759±3950/mm3 on the 1st day and 9687±4286/mm3 on the 7th day. A

significant difference was determined between the two groups in terms of leukocytes (p<0.001). Blood YKL-40 levels on the 7th day after antibiotic therapy were significantly lower than those at the start of treatment (p=0.003). Correlation analysis between YKL-40 and the other clinical and laboratory results were studied and detailed in Table 5. PSI and CURB-65 scores were not correlated with serum YKL-40.

Characteristics	Controls (n=22)	CAP 1st day (n=40)	CAP 7th day (n=40)	P* value
YKL-40, ng/mL	47,44 ± 7,16	89,24±98,67	74,37±56,28	<0.001[a] 0.001[b] 0.003[c]
Age, year	59,8 ± 13,0	70,53 ± 16,2	-	NS
Smoking, pack-year	19,45 ± 18,4	33,78 ± 34,20	-	0.03
Sex, male/female	14/8	29/11	-	NS
CRP, mg/L	-	22,2±12,95	5,78±5,6	<0.001
WBC, x10^3/mL	-	23759±3950	9687±4286	<0.001
ESR, /hour	-	66,4±33	67,18±28,07	NS
Fibrinogen, gr/L	-	717,1± 190,97	683,21±145,35	NS

Table 3: Comparison of sex, age, smoking history, CRP, WBC, ESR, fibrinogen and initial YKL-40 levels between study and control groups, the result are given as mean standard deviation with the ranges of ages

Wilcoxon test was used for comparison of parameters between groups. The results are shown as mean ± standard deviation. *$p < 0.05$ is significant, NS: not significant, a: compared to control and pneumonia at 1st day YKL-40 serum levels, b: compared to control and pneumonia at 7th day YKL-40 serum levels, c: compared to pneumonia at 1st day and 7th day YKL-40 serum levels. CAP 1st day: patients with CAP first day in treatment, CAP 7th day: patients with CAP seventh day in treatment, CRP: C-reactive protein, WBC: white blood cell, ESR: erythrocyte sedimentation rate.

Discussion

The most prevalent symptoms in CAP are fever, cough, phlegm, trembling and chest pain. Chest x-rays, blood count, biochemical tests and phlegm and blood culture tests must be performed, if pneumonia is suspected. Chest x-rays are required to support diagnosis and for differential diagnosis. New pulmonary infiltrates are valuable for diagnosis, but x-rays may be normal in the early stages of the disease in some patients, or may be confused with radiological findings of lung cancer, heart failure or various chronic diseases involving the lung. The expected increases in some blood tests may not be seen in the early stages of pneumonia, and there may be no growth in blood and phlegm cultures. For this and other similar reasons, there may be delays in diagnosing pneumonia and starting treatment (15). However, pneumonia treatment needs to be started as early as possible. This makes it essential to start an empirical treatment. While markers such as CRP and procalcitonin are useful in the diagnosis of pneumonia and in monitoring the severity of disease and treatment, novel

markers that can be used in the early diagnosis of CAP, which still has high morbidity and mortality levels, and that can show disease severity and success of treatment are also needed (6,7).

YKL-40 is released by synovial fibroblasts, chondrocytes, neutrophils, smooth muscle cells, macrophages and malign tumor cells and contributes to inflammation, angiogenesis, remodeling and cell proliferation, adhesion and migration (16). YKL-40 is a relative marker in inflammation, the role of which has been investigated in several inflammatory diseases (17). Recent research into various systemic diseases has shown a significant difference between serum YKL-40 levels in disease prognosis and severity. Blood YKL-40 levels have been shown to rise in *Streptococcus pneumoniae* bacteremia (18).

Serum YKL-40 levels have been shown to peak at the beginning of treatment in CAP and to decrease gradually on the 3rd and 10th days thereafter in recent studies (3). These results are in agreements with our study. Elevated YLK-40 levels have been shown in bronchial biopsy and blood specimens in inflammatory diseases such as asthma and COPD (19). That same study also showed than blood YLK-40 levels rose in parallel with severity of COPD attacks. Parapneumonic effusions may occur as a result of inflammation in patients with pneumonia. Parapneumonic effusions have been compared with congestive heart failure-related pleural effusions, and serum and pleural fluid YKL-40 levels were determined to be higher in cases with parapneumonic pleural effusion (20).

Conclusion

In this study, we focused on changes in YKL-40 levels in control and pneumonia group blood specimens. The results show higher blood LK-40 levels in patients in the CAP group compared to the controls. Our own and other studies show that blood YKL-40 levels may be an inflammatory marker. Elevated YKL-40 levels in blood specimens at the start of treatment in our pneumonia group, followed by a decrease one week later, may be regarded as evidence that blood YKL-40 levels can be used as an inflammation marker in clinical practice. Further studies with a wider patient group are now needed to determine what contribution the clinical use of YKL-40 can make as an inflammation marker.

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