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ORIGINAL ARTICLE

Interleukin-6 is prone to be a candidate biomarker for predicting incomplete and IVIG nonresponsive Kawasaki disease rather than coronary artery aneurysm

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Abstract

Kawasaki disease (KD) is an acute, systemic vasculitis and occurs mainly in childhood. Interleukin-6 (IL-6) is a pleiotropic cytokine synthesized predominantly by neutrophils and monocytes/macrophages and plays an important role in systemic inflammatory disease. However, a little information is currently available on the relationship of serum IL-6 with conventional inflammatory mediators, clinical classification, IVIG response and coronary artery aneurysm (CAA). 165 Chinese children with KD were enrolled and divided into six subgroups, including complete KD, incomplete KD, IVIG-responsive KD, IVIG-nonresponsive KD, coronary artery noninvolvement KD and coronary artery involvement KD. Blood samples were collected from all subjects within 24-h pre- and 48-h post-IVIG therapy, respectively. Serum IL-6 and conventional inflammatory mediators were detected. (1) Serum IL-6 markedly increased in the acute phase of KD, whereas declined to normal after IVIG therapy; it was positively correlated with C-reactive protein and erythrocyte sedimentation rate. (2) Serum IL-6 was significantly elevated in patients with incomplete KD when compared with their complete counterparts. The area under receiver operating characteristic curve (AUC) value for serum IL-6 in prediction of incomplete KD was 0.596, and the estimated sensitivity and specificity were 77.80% and 54.40% with a cutoff of IL-6 > 13.25 pg/ml, respectively. (3) Serum IL-6 was significantly elevated in patients with IVIG-nonresponsive KD when compared with their IVIG-responsive counterparts; the AUC value for serum IL-6 in prediction of IVIG-nonresponsive KD was 0.580, and the estimated sensitivity and specificity were 60.00% and 66.30% with a cutoff of IL-6 > 26.40 pg/ml, respectively. (4) No significant differences in IL-6 were found between KD patients with and without CAA. IL-6 is prone to be a candidate biomarker for predicting incomplete and IVIG nonresponsive KD rather than CAA.

Keywords Interleukin-6 · Intravenous immunoglobulin · Kawasaki disease · Vascular endothelial cell · Coronary artery aneurysm

Introduction

Kawasaki disease (KD), also known as mucocutaneous lymph node syndrome, is an acute, systemic vasculitis and occurs mainly in children less than 5 years old. According to a latest epidemiological survey in Shanghai, China, the average annual incidence rate of KD was 50.5 per 100,000 children during the period of 2008–2012 [1]. The 2017 American Heart Association (AHA) guidelines for KD diagnosis include ≥ 5 days of fever and ≥ 4 of the following five major features: conjunctival injection, cervical lymphadenopathy, oral mucosal changes, polymorphous eruption and swelling or redness of the extremities. However, patients with fever for ≥ 5 days and at least two of the major features can be diagnosed as having incomplete KD, if no other disease processes could explain the illness [2]. Coronary artery aneurysm (CAA) is an important prognostic factor in KD and acts as the leading cause of acquired heart disease. Daniels et al. [3] reviewed the medical records of 261 young adults with suspected myocardial ischemia and found that 5% of them had a KD history in their childhoods.

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Intravenous immunoglobulin (IVIG) combined with high-dose aspirin is used as the mainstay treatment for KD, which not only shortens fever duration and suppresses systemic inflammation, but prevents CAA onset to a large extent. A multicenter, randomized trials from USA documented that timely usage of IVIG could reduce the incidence of CAA from 20 to 6.8% at the 2-week visit [4].

Currently, there is no definitive test for the diagnosis of KD except for the increases in several inflammatory mediators, such as C-reactive protein (CRP), erythrocyte sedimentation rate (ESR), platelet (PLT) count and so on. Song et al. [5] evaluated the diagnostic efficiency of CRP, ESR and PLT in 67 KD patients and found that the estimated sensitivity of CRP for predicting KD was 69.0%, and specificity was 72.7%; the estimated sensitivity of ESR for diagnosing KD was 93.9%, and specificity was 83.3%; and estimated sensitivity of PLT for diagnosing KD was 70.6%, and specificity was 75%. Tumor necrosis factor (TNF)-α is one of the cytokines produced chiefly by activated monocyte/macrophages, and significantly elevates in KD [6, 7]. A recent retrospective study from Hangzhou, China, showed that serum TNF-α had a sensitivity of 66.7% and a specificity of 74.2% for predicting IVIG-nonresponsive KD [8]. Infliximab is a monoclonal antibody that binds to TNF-α and has been used as a novel therapy for IVIG-nonresponsive KD. Song et al. [9] administered a single infusion of infliximab (5–6.6 mg/kg) to 16 patients with IVIG-nonresponsive KD and found that all patients had a rapid fall in TNF-α and 80% of them obtained complete response without any coronary artery lesions. However, these above inflammatory mediators have limited diagnostic efficacies, and infliximab cannot be prescribed to the pediatric patients since it lacks the certification of the China Food and Drug Administration (CFDA). In this circumstance, a better biomarker is warranted to cover these limitations.

Interleukin-6 (IL-6) is a pleiotropic cytokine synthesized predominantly by neutrophils and monocytes/macrophages and plays an important role in immune regulation, inflammatory response, regenerative processes and metabolism [10]. The elevation in serum IL-6 has been documented to be associated with disease activity in patients with systemic juvenile idiopathic arthritis (sJIA) [11]. Spîrchez et al. [12] measured IL-6 concentrations in 40 sJIA patients and found that the estimated sensitivity and specificity for sJIA diagnosis were 40% and 90%, respectively, and its cutoff value obtained from receiver operating characteristic (ROC) curve was 8.33 pg/ml. Tocilizumab is the first humanized monoclonal antibody against IL-6 receptor and solely approved for JIA treatment by CFDA. Lai et al. [13] observed the therapeutic efficiency of tocilizumab on 40 Chinese patients with sJIA and discovered that arthritis symptom, fever and cutaneous lesion were alleviated significantly in more than 90% of patients after a 6-month intervention with tocilizumab. Similar to sJIA, KD is also identified as a prevalent systemic inflammatory disease in childhood and exhibited with increased circulating IL-6 [14]. However, a little information is currently available on the relationship of serum IL-6 with conventional inflammatory mediators, clinical classification, IVIG response and coronary arteritis.

Methods

Subjects

The present study was conducted retrospectively in the Department of Pediatrics, the First Affiliated Hospital of Anhui Medical University, Anhui province, PR China. A total of 165 KD children were recruited from July 2015 to August 2018. According to the 2017 AHA guidelines, the diagnostic criteria for complete KD comprise the presence of ≥ 5 days of fever and ≥ 4 of the following five major features: (1) bilateral conjunctival injection without exudates; (2) changes in the oral mucosa, such as erythema and cracking lips, erythema of the pharynx, strawberry tongue; (3) changes in extremities, such as redness and swelling in the acute phase, periungual desquamation in the subacute phase; (4) polymorphous exanthema; and (5) cervical lymphadenopathy (≥ 1.5 cm in diameter), usually unilateral. Patients with fever for ≥ 5 days and at least two of the principal features can be diagnosed as having incomplete KD, if no other disease processes could explain the illness. All patients received the standard therapy for KD, including a single infusion of high-dose IVIG (2 g/kg) and aspirin (30–50 mg/kg/d), within 10 days of the fever onset. IVIG-nonresponsive KD is defined as persistent or recrudescent fever ≥ 36 h after completion of the initial IVIG infusion. Echocardiographic evaluation of the coronary arteries is mainly based on the quantitative assessment of the internal diameters. Coronary arteritis is defined as a coronary artery having an internal diameter of at least 3 mm in children < 5 years or at least 4 mm in children ≥ 5 years, or a segment with an internal diameter at least 1.5 times larger than that of an adjacent segment by echocardiogram [2].

Laboratory analysis

Approval for this research was acquired from the Medical Ethic Committee of the First Affiliated Hospital of Anhui Medical University and obtained consent from parents. Blood samples were collected from all subjects within 24-h pre- and 48-h post-IVIG therapy, respectively. Venous blood (2 ml) was collected in a gel coagulation-promoting vacuum tube and centrifuged immediately at 2800 *g* for 15 min at room temperature, and plasma samples were stored at −80 °C. White blood cells counts (WBC), absolute

neutrophil counts (ANC) and PLT were performed using a flow cytometer (Sysmex XE-2100). ESR and CRP were determined by the Westergren method and immunoturbidimetry, respectively. Serum IL-6 was measured by enzyme-linked immunosorbent assay (Immulite) according to the manufacturer's protocols.

Statistical analysis

Normally distributed continuous data were expressed as mean ± SD. Comparisons of the frequencies between groups were analyzed using Chi-square tests. A standard two-tailed *t* test was used for analyzing serum data, and paired t test was used in comparison of pre-IVIG to post-IVIG data. Pearson correlation coefficients were reported for IL-6 and other variables of interest. Cutoff value, sensitivity and specificity of IL-6 were identified by receiver operating characteristic (ROC) curve. A value of $p < 0.05$ was considered significant. Statistical analysis was performed using the statistical package for social studies SPSS version 16.0.

Results

Patient characteristics and laboratory findings

During the observational period of 3 years, 92 boys and 73 girls were diagnosed as having KD, with a mean age at diagnosis of 35.17 ± 30.12 months and a range from 3 months to 13 years. The serum levels of IL-6 and five conventional inflammatory mediators were compared before and after IVIG therapy in all the patients (Table 1). Serum IL-6, WBC, ANC, CRP, ESR and PLT markedly increased in the acute phase of KD, in which IL-6, WBC, ANC and CRP declined to normal after IVIG therapy ($p < 0.05$). As compared with the time before IVIG therapy, ESR remained significantly higher after IVIG therapy (66.77 ± 22.46 mm/h vs. 60.25 ± 21.24 mm/h, p = NS), and PLT even exhibited a significant increase after IVIG therapy (355.46 ± 127.31 × 10^9/l vs. 500.83 ± 162.43 × 10^9/l, $p < 0.05$). The relationship between serum IL-6 and conventional inflammatory mediators before IVIG therapy is shown in Fig. 1. Serum IL-6 was positively correlated with CRP (Fig. 1c; $r = 0.35$, $p < 0.05$) and ESR (Fig. 1d; $r = 0.21$, $p < 0.05$), but no correlation existed between serum IL-6 and WBC (Fig. 1a; $r = 0.14$, $p > 0.05$), ANC (Fig. 1b; $r = 0.01$, $p > 0.05$), PLT (Fig. 1e; $r = -0.08$, $p > 0.05$), respectively.

Clinical classification

All five classic diagnostic criteria for KD were met in 50 cases (30.30%), four criteria in 67 (40.60%), three criteria in 28 (16.97%) and two criteria in 20 (12.12%). Therefore, 117 children (70.91%) had complete KD, including 68 males and 49 females with the mean age of 35.99 ± 30.27 months; 48 children (29.09%) had incomplete KD, including 24 males and 24 females with the mean age of 33.17 ± 29.98 months. The male/female ratio, mean age and internal diameter of coronary artery were almost identical between the complete KD patients and the incomplete KD patients ($p > 0.05$). The differences of IL-6 and conventional inflammatory mediators between complete KD and incomplete KD are presented in Table 2. Serum IL-6 was significantly elevated in patients with incomplete KD when compared with their complete counterparts ($t = 4.34$, $p < 0.05$). However, no significant differences in WBC, ANC, CRP, ESR and PLT were observed between the two groups ($p > 0.05$). When serum IL-6 levels were substituted in a ROC curve analysis, the area under receiver operating characteristic curve (AUC) was 0.596. The estimated sensitivity of serum IL-6 for the diagnosis of incomplete KD was 77.80%, and specificity was 54.40% with a cutoff of IL-6 > l3.25 pg/ml (Fig. 2a).

IVIG therapy

According to the fever duration after the initial IVIG therapy, 11 KD patients (6.67%) were identified as IVIG-nonresponsive KD with the persistent fever about 71.00 ± 12.27 h, including four males and seven females with the mean age of 33.72 ± 22.45 months. In contrast, 154 patients were diagnosed with IVIG-responsive KD and exhibited a dramatic decrease in fever duration (8.25 ± 7.16 h; $t = 8.47$, $p < 0.05$), including 88 males and 66 females with the mean age of 35.27 ± 33.07 months. There was no noted discrepancy

Table 1 IL-6 and conventional inflammatory mediators in KD children before and after IVIG therapy

	Pre-IVIG	Post-IVIG	*T* value	*p* value
IL-6 (pg/ml)*	49.73 ± 8.89	7.05 ± 5.96	4.95	0
WBC (× 10^9/l)*	13.15 ± 4.86	9.54 ± 3.55	8.38	0
ANC (× 10^9/l)*	10.14 ± 8.86	4.25 ± 5.12	13.16	0
PLT (× 10^9/l)*	355.46 ± 127.31	500.83 ± 162.43	11.94	0
CRP (mg/l)*	62.80 ± 44.26	10.68 ± 7.82	15.98	0
ESR (mm/h)	66.77 ± 22.46	60.25 ± 21.24	1.65	0.12

*$p < 0.05$

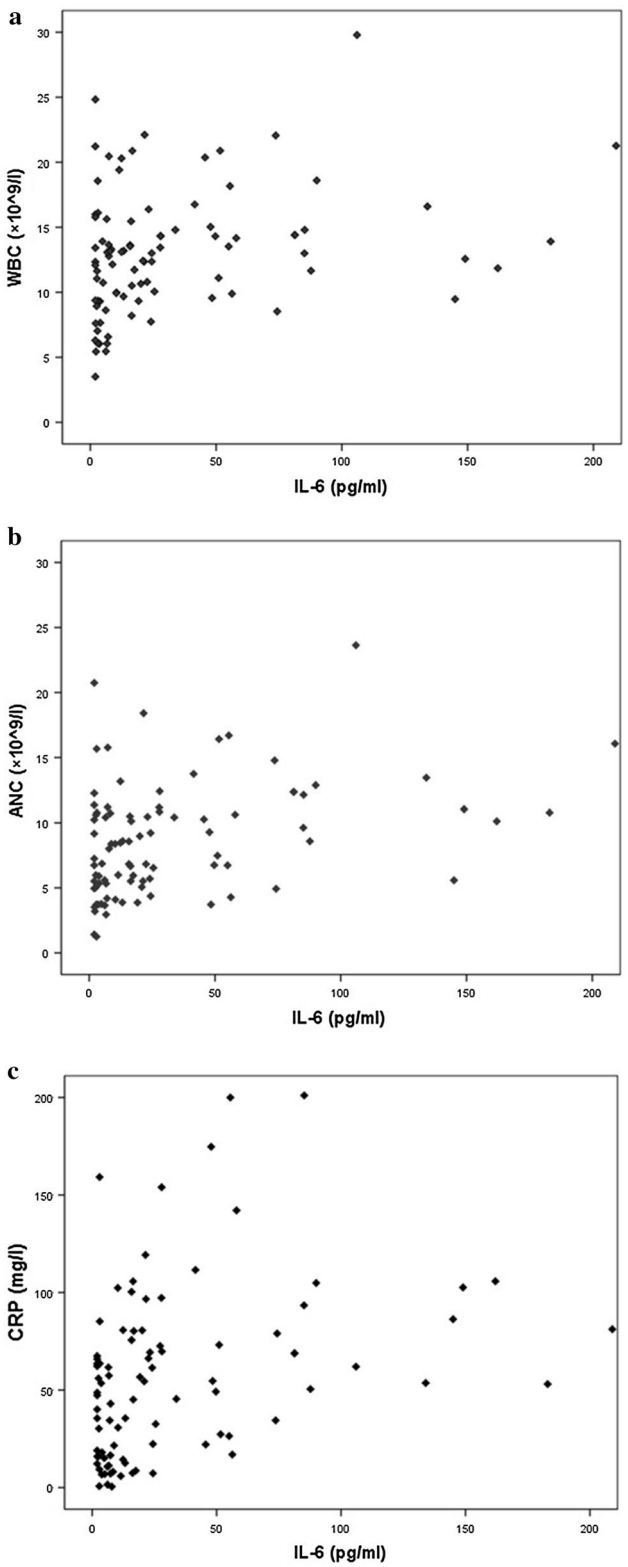

Fig. 1 Relationship between plasma IL-6 and conventional inflammatory mediators before IVIG therapy. Serum IL-6 was positively correlated with CRP (**c** $r=0.35$, $p<0.05$) and ESR (**d** $r=0.21$, $p<0.05$), but no correlation existed between serum IL-6 and WBC (**a** $r=0.14$, $p>0.05$), ANC (**b** $r=0.01$, $p>0.05$), PLT (**e** $r=-0.08$, $p>0.05$), respectively

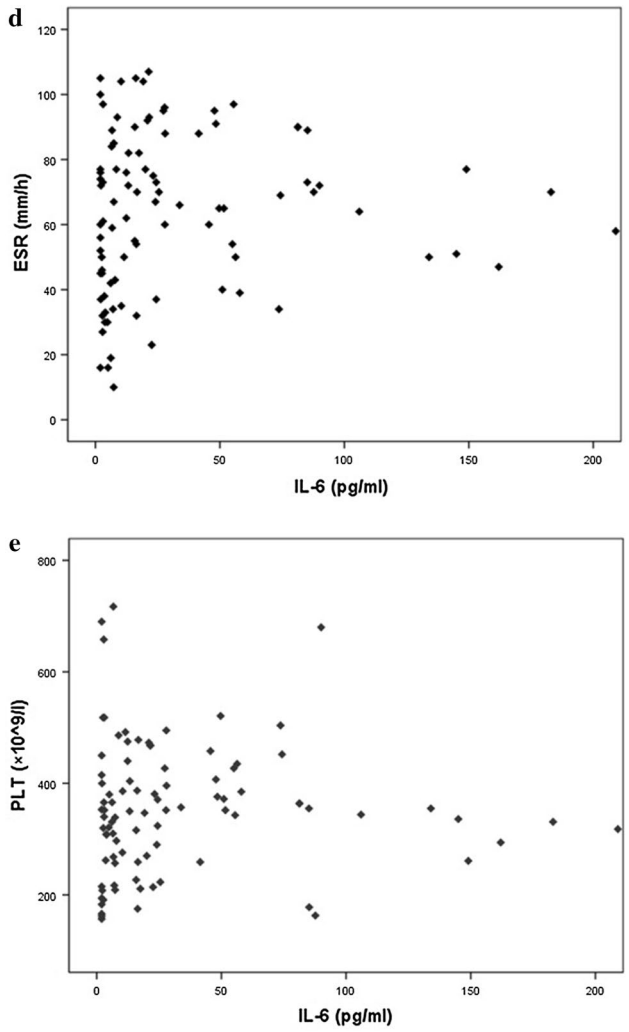

Fig. 1 (continued)

regarding the male/female ratio and mean age regardless of IVIG response or not ($p>0.05$). IVIG-nonresponsive KD was significantly associated with coronary artery abnormalities both in the left (2.44 ± 0.76 mm vs. 2.13 ± 0.45 mm; $t=2.47$, $p<0.05$) and in the right (2.42 ± 0.61 mm vs. 2.06 ± 0.60 mm; $t=2.95$, $p<0.05$), when compared with IVIG-responsive KD. The differences of IL-6 and conventional inflammatory mediators between IVIG-responsive KD and IVIG-nonresponsive KD are presented in Table 2. Serum IL-6 and ANC were significantly elevated in patients with IVIG-nonresponsive KD when compared with their IVIG-responsive counterparts (IL-6: $t=3.29$, $p<0.05$; ANC: $t=2.49$, $p<0.05$). However, no significant differences in WBC, CRP, ESR and PLT were observed between the two groups ($p>0.05$). The AUC value for serum IL-6 in prediction of IVIG-nonresponsive KD was 0.580, and the estimated sensitivity and specificity were 60.00% and 66.30% with a cutoff of IL-6 > 26.40 pg/ml, respectively (Fig. 2b).

Table 2 IL-6 and conventional inflammatory mediators in patients with different types of KD

KD patients ($n=165$)	IL-6 (pg/ml)	WBC ($\times 10^9$/l)	ANC ($\times 10^9$/l)	CRP (mg/l)	ESR (mm/h)	PLT ($\times 10^9$/l)
Clinical classification						
Complete ($n=117$)	54.60 ± 10.82	13.21 ± 4.72	10.78 ± 11.16	58.90 ± 45.80	61.49 ± 24.61	357.10 ± 133.61
Incomplete ($n=48$)	76.16 ± 19.56*	14.38 ± 6.20	11.76 ± 14.17	64.92 ± 43.62	64.62 ± 22.52	344.56 ± 106.41
IVIG therapy						
Response ($n=154$)	33.01 ± 44.08	13.35 ± 4.95	8.19 ± 4.50	59.15 ± 46.26	61.65 ± 24.13	351.21 ± 123.64
Nonresponse ($n=11$)	52.08 ± 33.04*	16.24 ± 7.72	12.46 ± 7.37*	60.31 ± 27.53	72.64 ± 22.95	384.91 ± 160.52
Coronary artery						
Normal ($n=154$)	54.31 ± 31.92	13.59 ± 5.25	9.12 ± 4.87	60.86 ± 45.97	62.87 ± 23.81	351.92 ± 123.28
CAA ($n=11$)	47.48 ± 31.82	12.95 ± 4.77	7.81 ± 3.74	43.85 ± 29.90	55.72 ± 28.85	375.00 ± 166.60

*$p<0.05$

Coronary artery involvement

Based on the internal diameter of coronary artery, 11 KD patients (6.67%) were defined as having coronary arteritis after IVIG therapy (left coronary artery: 3.20 ± 0.64 mm; right coronary artery: 3.29 ± 0.37 mm), including seven males and four females with the mean age of 27.64 ± 32.35 months. In contrast, 154 patients had normal coronary arteries after IVIG therapy (left coronary artery: 2.07 ± 0.37 mm, right coronary artery: 1.99 ± 0.33 mm), including 85 males and 69 females with the mean age of 35.71 ± 29.99 months. The internal diameters of both coronary arteries were significantly larger in KD patients undergoing coronary arteritis (left coronary artery: $t=5.77$, $p<0.05$; right coronary artery: $t=2.80$, $p<0.05$), when compared with KD patients without coronary arteritis. The mean age and male/female ratio were almost identical in the two groups ($p>0.05$). The differences of IL-6 and conventional inflammatory mediators between KD patients with and without coronary arteritis are presented in Table 2. No significant differences in IL-6, WBC, ANC, CRP, ESR and PLT were found between the two groups ($p>0.05$). Relationship between inflammatory mediators and coronary artery involvement is shown in Fig. 3. No correlation of the internal diameter of coronary artery with IL-6 (Fig. 3a; $r=-0.02$, $p>0.05$), WBC (Fig. 3b; $r=-0.04$, $p>0.05$), ANC (Fig. 3c; $r=-0.04$, $p>0.05$), CRP (Fig. 3d; $r=-0.03$, $p>0.05$), ESR (Fig. 3e; $r=-0.08$, $p>0.05$) and PLT (Fig. 3f; $r=-0.05$, $p>0.05$) was determined, respectively.

Discussion

IL-6 is a 26 kDa, four-helical glycopeptide and participates in the pathogenesis of multiple systemic inflammatory diseases, such as sJIA, systemic lupus erythematosus, dermatomyositis, Behcet's disease and KD [11, 12, 15–17]. A prospective study conducted by Ye et al. [18] revealed that serum IL-6 markedly increased in Chinese patients with KD (89.30 pg/ml) when compared with normal controls (4.00 pg/ml). More specifically, a clinical trial from Yonsei University College of Medicine, Korea, demonstrated that mean serum IL-6 significantly increased in the acute phase of KD (123.0 pg/ml) compared to those in the subacute phase (25.7 pg/ml) [19]. The regulatory mechanisms of IL-6 production in systemic inflammatory diseases remain unclear. Adenine–thymine-rich interactive domain 5a (Arid5a), a novel IL-6-regulating molecule, specifically stabilizes interleukin-mRNA and maintains its overproduction; thus, Arid5a plays an important role in accelerating inflammation and autoimmune diseases [20]. On the contrary, in Arid5a-knockout mice, experimental autoimmune encephalomyelitis does not develop, and lipopolysaccharide stimulation does not induce elevated expression of IL-6 [21]. Since the main objective of the present study was not to confirm these previous findings, normal controls were not recruited by our research group. Uniquely, the present study shed light on the relationship of serum IL-6 with clinical classification, IVIG response and coronary arteritis. Our findings indicated that IL-6 markedly increased in the acute phase, whereas it declined to normal after IVIG therapy and significantly elevated in those who had incomplete KD or IVIG-nonresponsive KD. However, no significant difference in IL-6 was found regardless of coronary arteritis or not.

Although several conventional inflammatory mediators could not serve as the definitive diagnostic evidence for KD, they are really beneficial to heighten or reduce its suspicion. As outlined in the 2017 American Heart Association guidelines, conventional inflammatory mediators including WBC, ANC, CRP, ESR and PLT usually reached the summit during the acute phase, whereas PLT peaked later [2]. Besides the above consensuses, the present study observed the relationship between serum IL-6 and conventional inflammatory mediators and found that IL-6 was positively correlated with CRP and ESR. Similarly, a retrospective study encompassing 60 KD patients by Lin et al. [22] indicated

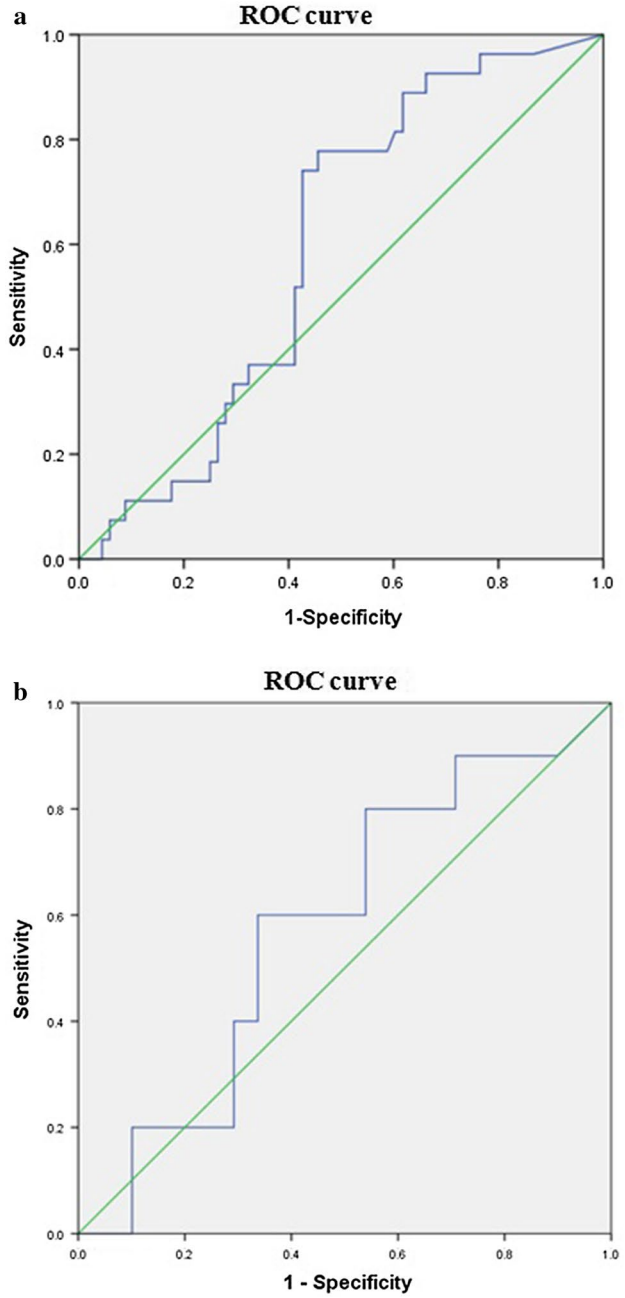

Fig. 2 **a** Receiver operating characteristic (ROC) curve of serum IL-6 for the diagnosis of incomplete KD. **b** Receiver operating characteristic (ROC) curve of serum IL-6 for the diagnosis of IVIG-nonresponsive KD

that serum IL-6 was highly correlated with CRP and ESR during the first week; however, there was no significant correlation between them thereafter. Peng et al. [23] reviewed the medical records of 32 KD patients and discovered that IL-6 not only positively correlated with CRP but returned to normal more rapidly than CRP after IVIG therapy, which may be ascribed to the stimulation of IL-6 to CRP expression in hepatocytes [24]. In contrast, IL-6-deficient mice show a compromised acute phase response and fail to control infection with the pathogen [25]. On this background, we hypothesize that IL-6 may rise ahead of CRP and fluctuate more sensitively in KD.

It is a challenge for pediatricians to distinguish incomplete KD due to the lack of typical clinical manifestations and the shortage of specific laboratory biomarkers for the early stage of disease. In addition, the delayed recognition of incomplete KD acts as an important risk factor for CAA onset. A systematic review and meta-analysis by Ha et al. [26] identified the risk factors of CAA in 4504 KD patients and demonstrated that the incomplete KD increased significantly the risk of CAA (OR 1.447, 95% CI 1.158–1.808). In the past two decades, several epidemical surveys from East Asia have devoted to screen the clinical features and laboratory biomarkers for the early recognition of incomplete KD. In 2012, Liu et al. [27] analyzed the clinical data of 145 Taiwan children who were diagnosed with KD since 1993 to 2008 and revealed that the infants younger than 6 months old took the longest time from the day of symptoms onset to diagnosis, and they had the largest proportion of patients fulfilling the criteria of incomplete KD. In 2017, another prospective study from Shaanxi, China, indicated that incomplete KD had a significant increase in CRP and ESR, whereas it experienced a marked reduction in PLT as compared with complete KD [28]. Complementally, the present study noted that incomplete KD exhibited a higher IL-6 than its counterparts, similar to the report of Peng et al. [29]. However, whether IL-6 can serve as a novel biomarker for predicting incomplete KD is still to be elucidated. Based on this hypothesis, serum IL-6 levels were substituted in a ROC curve and the AUC values for the prediction of incomplete KD were 0.596 with a sensitivity of 77.80% and a specificity of 54.40%.

IVIG-nonresponsive KD is at the great risk of developing CAA [2]. Kobayashi et al. [30] reviewed the medical records of 750 KD patients and found that CAA occurred in 32% of IVIG nonresponders and 0.8% of IVIG responders in the acute phase, and CAA occurred in 15% of IVIG nonresponders and 0.2% of IVIG responders at 4 weeks. Predicting IVIG nonresponders before initiating therapy would assist pediatricians in selecting more judicious and aggressive treatment. A multicenter and nationwide survey conducted in Korea showed that laboratory predictive factors for IVIG nonresponders included increased ANC, CRP, pro-brain natriuretic peptide, alanine aminotransferase, aspartate aminotransferase, hyponatremia and hypoproteinemia [31, 32]. Tocilizumab is the first humanized monoclonal antibody against IL-6 receptor and solely approved to be prescribed to the pediatric patients by CFDA. Therefore, study on the predictive effect of IL-6 may provide more forceful evidence on tocilizumab usage in IVIG nonresponders. In 2017, Xie et al. [33]

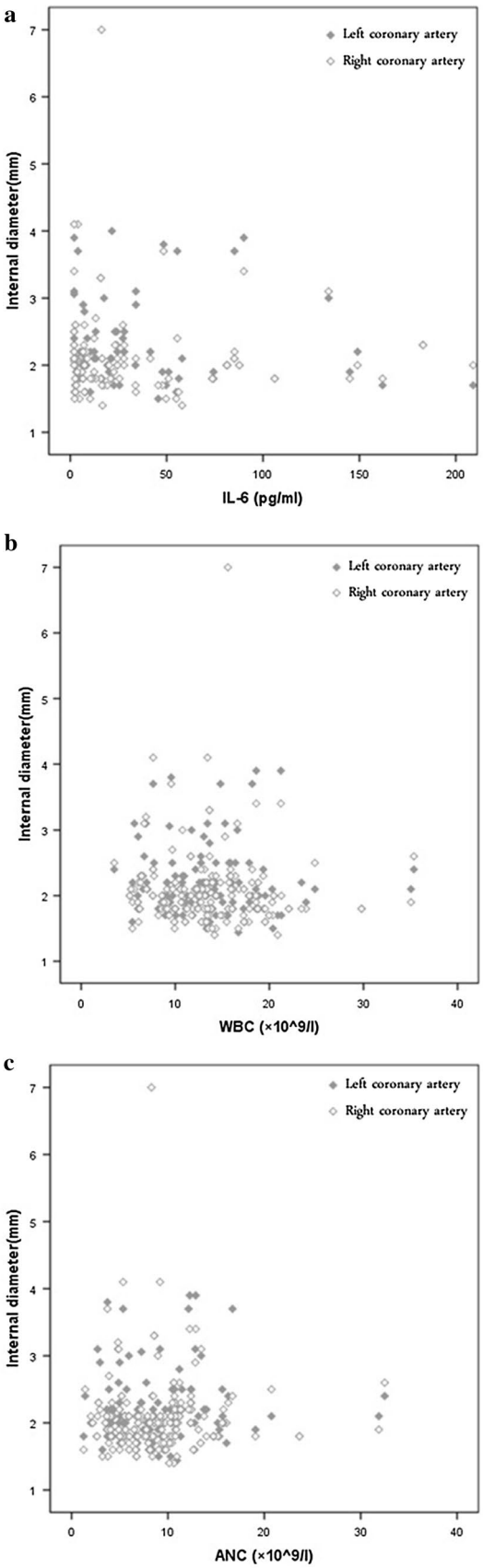

Fig. 3 Relationship between inflammatory mediators and coronary artery involvement. No correlation of the internal diameter of coronary artery with IL-6 (**a** $r=-0.02$, $p>0.05$), WBC (**b** $r=-0.04$, $p>0.05$), ANC (**c** $r=-0.04$, $p>0.05$), CRP (**d** $r=-0.03$, $p>0.05$), ESR (**e** $r=-0.08$, $p>0.05$) and PLT (**f** $r=-0.05$, $p>0.05$) was determined, respectively

established a multivariable logistic regression analysis to predict the risk of IVIG nonresponders and discovered that elevated IL-6 was identified as an independent risk factor with a sensitivity of 76.19% and a specificity of 61.59%. In the present study, we also found that serum IL-6 was significantly elevated in IVIG nonresponders, and furthermore, had a sensitivity of 60.00% and a specificity of 66.30% at a cutoff value of > 26.40 pg/ml to predict IVIG nonresponders. Up to now, the potential mechanisms for the inflammatory modulations of IVIG are not clearly understood, but may be largely dependent on Th2 cytokines, such as IL-6. Gupta et al. [34] examined the effects of IVIG (16.7 mg/ml) on peripheral blood cells treated with lipopolysaccharides (LPS, 50 ng/ml) and found that LPS-induced upregulation of IL-6 was suppressed by IVIG to some extent. However, overexpressed IL-6-related genes exceeding the inhibitory ability of IVIG have been proved to be involved in IVIG nonresponse [35].

CAA influences the long-term prognosis of KD and is associated with many risk factors. Maric et al. [36] retrospected the medical records of 111 KD patients between 2003 and 2012 and found that disease duration before hospitalization > 6 days, age < 6 months old and platelet count > 771×10^9/l after the seventh day of disease were the candidate risks for CAA.

In the latest report of Si et al. [37], IL-6 was subjected to a 2.13-fold increase in KD patients with CAA as compared to those with normal coronary arteries. As we all know, migration of vascular smooth muscle cells (VSMC) into the intima is considered to be a vital step in the pathophysiology of atherosclerosis. An in vitro study conducted by Lee et al. [38] revealed that IL-6 could obviously inhibit Toll-like receptor 4 activation-induced VSMC migration. The present study also investigated the relationship between serum IL-6 and CAA; however, no significant difference in IL-6 was detected between the above two groups, and moreover no significant correlation was observed between IL-6 and the internal diameter of coronary artery. Consistently, a cross-sectional study in Japan indicated that IL-6 was not helpful in screening CAA [39]. Therefore, whether IL-6 participates in CAA onset is still controversial, based on the current evidence. Further studies are warranted to elucidate this issue in detail.

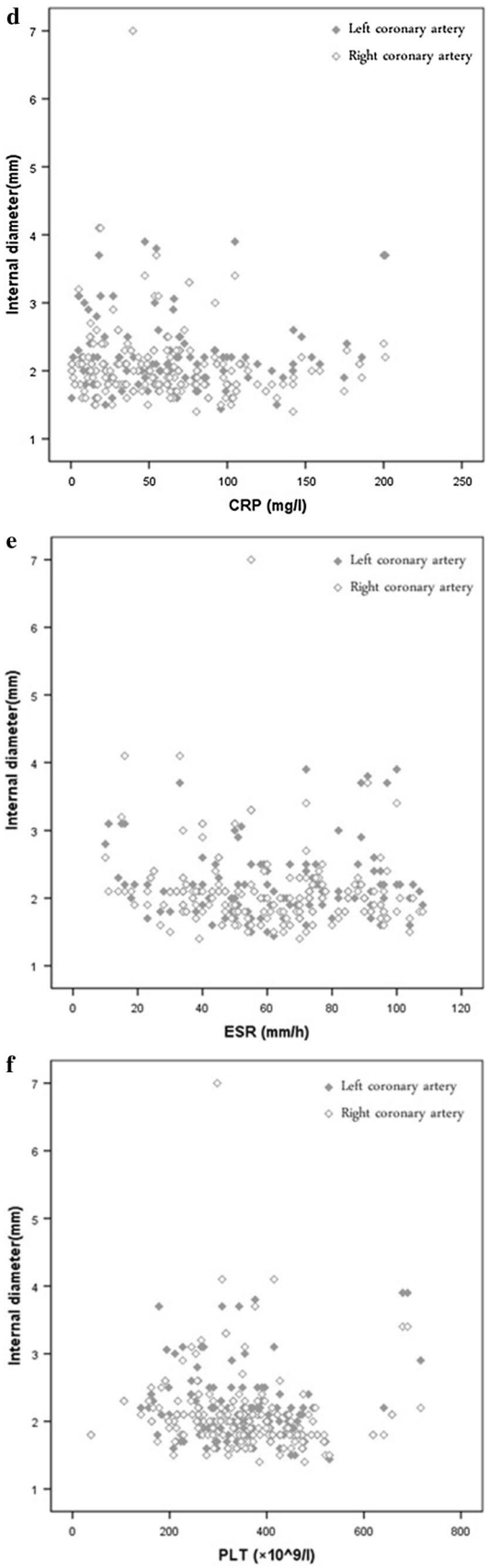

Fig. 3 (continued)

Conclusions and perspectives

According to the findings of our study, IL-6 is prone to be a candidate biomarker for predicting incomplete and IVIG nonresponsive KD rather than CAA. We hypothesized that tocilizumab may advance a novel therapeutic strategy for KD. In 2017, Nozawa et al. [40] administered a single infusion of tocilizumab (8 mg/kg) to four patients with IVIG-nonresponsive KD and found that all patients had rapid improvement in clinical and laboratory features within 48 h, except that only one patient had a persistent giant CAA. However, the therapeutic effect of tocilizumab on KD should be confirmed by a randomized controlled trial in the near future.

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Compliance with ethical standards

Conflict of interest The authors have no conflicts of interest relevant to this article to disclose.

Ethical approval Approval for this research was acquired from the Medical Ethic Committee of the First Affiliated Hospital of Anhui Medical University.

Informed consent Consent was obtained from parents.

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