Estimation of C-reactive Protein Associated with Mandibular Fracture

D. N. Kiran · Rajendra Desai

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Abstract

Background The aim of the present study is to evaluate the C-reactive protein (CRP) levels pre-operatively and post-operatively following a surgical intervention of mandibular fracture with osteosynthesis by rigid fixation using AO/ASIF principles and to try and correlate the prognosis of the convalescent period.

Methods Twenty five patients with trauma were surgically treated. The blood samples are collected pre-operatively, immediate post-operatively, after 24 h and on the seventh post-operative day.

Results The CRP levels were high pre operatively due to body's initial response to trauma. An increase was noticed immediately after the surgery (mean value 1.96 ± 0.56 mg/dl). After 24 h of surgery, CRP levels were raised markedly (mean value of 2.3 ± 0.58 mg/dl). On the seventh day after the surgery CRP levels were significantly decreased to attain normal level (mean value of 1.58 ± 0.52 mg/dl), indicating normal healing at the surgical site.

Conclusion In cases of patients with mandibular fracture the CRP concentration increases directly after the trauma and the surgical procedure. Then it undergoes a gradual normalisation which ensures non complicated healing post operatively.

Keywords C-reactive protein · Trauma · Acute phase protein · Mandibular fractures · Infection · CRP

Introduction

Maxillofacial surgeons routinely encounter patients with facial trauma. In maxillofacial trauma, mandibular fractures alone accounts to 61% of all fractures. The sequel of a mandibular fracture will invariably lead to swelling which may be just an inflammatory response or as a result of infection, prior to treatment or post treatment. This makes it difficult for a surgeon to differentiate between the two. There are many laboratory investigations to evaluate the prognosis of healing. Routinely inflammatory markers are measured in frozen serum using standardized assays like interleukin (IL-6), Tumor necrosis factor (TNF), C-reactive protein (CRP), and soluble receptors (IL-2 sR, IL-6 sR, TNF sR1 and TNF sR2). Bacterial markers including white blood cell (WBC) count, absolute neutrophil count (ANC) and CRP estimations are considered. But off late, CRP is gaining importance.

C-reactive protein is an acute-phase protein synthesized by the liver in response to a number of stimuli that involve tissue damage. Trace amount of CRP is present in healthy people in the blood serum [1, 2]. Stimulation of macrophage caused by tissue damage is necessary for CRP synthesis [3–5]. Interleukin 6 (IL-6) is the most important factor that stimulates CRP synthesis as well as all other acute phase proteins [5, 6]. Interleukin 1 (IL-1) and tumour necrosis factor (TNF- α) stimulate macrophages, monocytes, fibroblasts, endothelium cells and others to produce IL-6 [6–8]. CRP in the organism performs a defensive and reparative role [2, 3, 9, 10].

Bacterial infection is a particularly potent stimulus with marked elevation in serum CRP levels occurring within a

D. N. Kiran (✉)
Department of Oral and Maxillofacial Surgery,
M M College of Dental Sciences & Research,
M M University, Mullana, Ambala, Haryana, India
e-mail: kdn30673@gmail.com

R. Desai
Department of Oral and Maxillofacial Surgery,
College of Dental Sciences, Davangere, Karnataka, India

few hours. Infection elicits a powerful inflammatory response, both locally and systemically, with chemotactic cytokine release into the circulation.

C-reactive protein appears in the plasma as early as 2 h after the trauma and it reaches its peak 48–72 h after the injury. In non-complicated cases it returns to normal after 6–7 days [7, 9]. Its value decreases gradually; it rises, however, when the healing is interrupted by a bacterial infection. Fractures of the facial skeleton are in danger of infections with bacterial flora present in the oral cavity. The anti-bacterial prophylaxis which is routinely administered may be terminated when plasma CRP level normalizes. This avoids prolonged antibiotics therapy and its side-effects [11].

The aim of the present study is to evaluate the CRP levels pre-operatively and post-operatively following a surgical intervention of mandibular fracture with osteosynthesis by rigid fixation using AO/ASIF principles and to correlate the prognosis of the convalescent period.

Methods

Twenty five patients were treated for mandibular fracture due to trauma with osteosynthesis by rigid fixation using AO/ASIF principles. With the clearance from the ethical committee, CRP concentration was determined pre operatively, immediate post operative, after 24 h and on the seventh post operative day. The patients were in the age group of 12–50 years. During the hospitalization period all patients received antibiotics as prophylaxis, mainly from the cephalosporin group. Patients with malnutrition, endocrinological, immunological ailments and with hepatocellular or cardiovascular system damage were excluded from the measurements. The time interval between the occurrence of trauma and the surgery ranged from a period of 2 to 38 days. In these, surgical procedures conducted were either intra-oral or extra-oral. Intra-orally, 19 cases and extra-orally, 5 cases were carried out. In one case both approaches were utilized, but this is considered under the extra-oral procedures in our analysis. The duration of surgery ranged from 15 to 150 min.

The criteria for the diagnosis of post-operative wound infection were those used by the National Research Council [12] of USA who defined POWI as “the presence of pus in a wound which has either discharged spontaneously or has to be released by the removal of sutures or re-opening the incision” [13–17].

Materials Used

- Reagent (Dr. Reddy's Laboratory reagents)

- C-reactive protein antibody
Solution containing goat anti human CRP (0.9 mg/ml) and sodium azide (0.09%) pH 7.0.
- C-reactive protein buffer
Solution containing phosphate buffer (120 mmol/l) and sodium azide (0.09%) pH 7.5.
- C-reactive protein standard
Serum (Human) containing sodium azide (0.1%).

- Auto analyzer equipment.

The cubital fossa was cleansed with antiseptic solution and venipuncture was done. A 23 gauge needle was used to draw 3 cc of blood without tourniquet stasis. Contracted clot is centrifuged at 4000 rpm for 20 min at 4°C and serum was gently pipetted off into a clean tube using a glass pasteur pipette. Samples were analyzed by auto analyzer method. Serum collected was mixed with buffer and antibody. CRP in the sample combines specifically with anti-human CRP in the reagent to yield an insoluble aggregate that causes increased turbidity in the solution. The degree of turbidity of the solution was measured optically which is proportional to the amount of CRP in the patient's sample.

Results

In our study, the patients were in the age group of 12–50 years, with a mean age of 29.6 ± 9.5 years. The time interval between the occurrence of trauma and the surgery ranged from a period of 2 to 38 days, with a mean of 9 ± 8 days. The duration of surgery ranged from 15 to 150 min, with a mean range of 40 ± 32 min.

The CRP levels were slightly high when the patients were hospitalised. An increase was noticed immediately after the surgery to 1.96 ± 0.56 mg/dl, which is considered to be normal mechanism of the body. Twenty four hours after the surgery CRP levels were raised markedly (mean value of 2.3 ± 0.58 mg/dl). On the seventh day after surgery, CRP levels were significantly decreased to attain normal levels (1.58 ± 0.52 mg/dl), indicating abolition of inflammation and normal healing at the surgical site (Tables 1, 2, 3; Fig. 1).

Discussion

C-reactive protein was discovered in 1930 by Tillett and Francis [18], as they were investigating serological reactions in pneumonia with various extracts of pneumococci and observed a non-specific somatic polysaccharide fraction, which they designated as “Fraction C”, which were found to be precipitated by the sera of acutely ill patients.

Table 1 Frequency distribution with corresponding C-reactive protein levels at different times

CRP (mg/dl)	Pre operative	Post operative, same day	After 24 h	7th day
0.1–0.5	2	–	1	2
0.5–1.0	1	2	–	1
1.0–1.5	3	3	1	6
1.5–2.0	12	4	4	13
2.0–2.5	7	12	8	2
2.5–3.0	–	4	10	1
3.0–3.5	–	–	1	–
Total	25	25	25	25
Mean CRP $\pm$ SD	1.66 $\pm$ 0.53	1.96 $\pm$ 0.56	2.30 $\pm$ 0.58	1.58 $\pm$ 0.52

Table 2 Post-operative changes in C-reactive protein levels

Time interval	CRP levels (mg/dl)				Difference from pre-op	Significance of difference*	
	Min	Max	Mean	SD		t-value	P-value
Pre-operative	0.3	2.2	1.66	0.53	–	–	–
Post-operative	0.6	2.6	1.96	0.56	0.30 $\pm$ 0.13	11.33	0.001, HS
After 24 h	0.8	3.2	2.30	0.58	0.64 $\pm$ 0.26	12.34	0.001, HS
7th day	0.4	2.6	1.58	0.52	(–) 0.08 $\pm$ 0.29	1.51	0.14, NS

* Paired *t*-test

(–) Sign indicates decrease in C-reactive protein levels

Table 3 Comparison of changes in C-reactive protein level between the intraoral and extraoral approaches

Time interval	Intraoral (n = 19)		Extraoral (n = 6)		Intraoral vs. extraoral
	Mean $\pm$ SD	Difference from pre-operative	Mean $\pm$ SD	Difference from pre-operative	
Pre-operative	1.60 $\pm$ 0.59	–	1.87 $\pm$ 0.16	–	–
Post-operative	1.88 $\pm$ 0.61	0.28 $\pm$ 0.13	2.20 $\pm$ 0.18	0.33 $\pm$ 0.15	NS
After 24 h	2.22 $\pm$ 0.64	0.62 $\pm$ 0.28	2.53 $\pm$ 0.23	0.67 $\pm$ 0.20	NS
7th day	1.53 $\pm$ 0.58	(–) 0.07 $\pm$ 0.33*	1.72 $\pm$ 0.18	(–) 0.15 $\pm$ 0.05	NS

* Unpaired *t*-test. Not significant **Fig. 1** Graph showing mean changes in C-reactive protein level

After the crisis the capacity of the patient's sera to precipitate C-polysaccharide (CPS) rapidly disappeared and the C-reactive material was not found in sera of normal healthy individuals.

Avery et al., characterized the C-reactive material as a protein which required calcium ions for its reaction with CPS and introduced the term 'acute phase protein' to refer to sera from patients acutely ill, with infectious diseases, which contains the CRP.

Iizuka [9], studied 80 patients, who underwent treatment for mandibular fractures with osteosynthesis by rigid fixation using AO/ASIF principles noticed the pre-operative CRP values of 28.5 mg/l and it reached maximum of 73.2 mg/l on the second day of the surgery.

In our study, CRP levels were increased at the time of hospitalization (mean value 1.56 mg/dl). This may be due to trauma or both trauma and infection. Just before the surgery, CRP levels were comparatively decreased (mean value 0.3 mg/dl), which may be attributed to administration of antibiotics. Post operatively, after 24 h of surgery, CRP levels were raised markedly (mean value 2.30 ± 0.58 mg/dl) even though administration of antibiotics was maintained which may be due to surgical trauma. On the seventh day, CRP levels decreased indicating normal healing at the surgical site. These results correlated with the study of Iizuka [9].

Also, Werner [19] demonstrated that the levels of β globulins, including CRP, were raised in serum after initial trauma. He also reported that elevated levels of β globulins and CRP were due to surgery. Our study concurs with this study of Mario Werner where in CRP levels increased in response to trauma as well as surgery.

It is difficult to differentiate a post-surgical inflammatory and infectious condition, even though clinical signs are the same. So CRP as a prognostic tool is a reliable exponent of the regularity of ossification and reparative processes. In the conditions of correct bone fusion its value is low; it rises, however, when the healing is interrupted by a bacterial infection. Fractures of the facial skeleton are in danger of infections with saprophytes and pathological bacterial flora present in the oral cavity. The perioperative anti-inflammatory prophylaxis which is routinely administered may be terminated when plasma CRP level normalizes, thus preventing prolonged antibiotic therapy and its side-effects [11].

Schentag et al. [20], noted that patient with abdominal sepsis who could not be treated by antibiotics alone and needed surgery and antibiotics had a higher CRP level pre-operatively indicating infection. CRP levels rose after surgery and reduced to normal after the seventh post-operative day indicating normal healing at the surgical site. In the present study, CRP levels rose after surgery due to surgical trauma. Normal CRP levels after the seventh day post-operatively indicating normal healing at the surgical site.

Mayer et al. [21], stated that CRP levels predicted exacerbation and severity of acute pancreatitis before appearance of clinical signs and symptoms. In the present study, since we did not come across pre operative or post operative infection in the study population, hence we could not estimate the CRP levels in infected cases. So, further research is needed in infected cases.

The CRP is one of the most important defensive proteins that take part in the nonspecific immunity mechanism as a result of the acute phase response. In the course of the acute phase response in patients with mandibular fractures, the CRP concentration increases directly after the trauma and the surgical procedure, but only to values slightly

higher than the normal CRP level in blood serum. Then it undergoes a gradual normalization in the course of non-complicated healing.

The above study gives an impression that an estimation of CRP levels helps us to confirm the normal healing pattern and thereby enable us to alter the further treatment protocol of the patient if necessary, so as to prevent the occurrence of the post operative complications.

The estimation of CRP level though cannot be said as the only parameter for assessing the prognosis of the disease, but it certainly gives an indication of healing. Also as the CRP levels normalize, administration of antibiotics can be terminated, which prevents prolonged usage of antibiotics and its side effects.

References

1. Macy EM et al (1997) Variability in the measurement of C-reactive protein in healthy subjects: implications for reference intervals and epidemiological applications. *Clin Chem* 43:52–58
2. Black S et al (2004) C-reactive protein. *J Biol Chem* 279(47):48487–48490
3. Mackiewicz S et al (1993) Nowe aspekty odpowiedzi ostrej fazy. *Post Nauk Med* 6:147–153
4. Mortensen RF (2001) C-reactive protein, inflammation, and innate immunity. *Immunol Res* 104:1192–1196
5. Nerkowska M (1998) Przydatność białka C-reaktywnego we współczesnej diagnostyce. *Klin Chor Zak Zak Szpit* 2:69–73
6. Stahl W (1987) Acute phase protein response to tissue injury. *Crit Care Med* 15(6):545–550
7. James K (2002) Cellular and humoral mediators of inflammation. In: McClatchey KD (ed) *Clinical laboratory medicine*. Lippincott Williams & Wilkins, Philadelphia
8. Stein MP (2000) C-reactive protein binding to murine leukocytes requires Fc γ receptors. *J Immunol* 164:1514–1520
9. Iizuka T, Lindqvist C (1991) Changes in C-reactive protein associated with surgical treatment of mandibular fractures. *J Oral Maxillofac Surg* 49:464–467
10. Kallio P et al (1990) C-reactive protein in tibial fractures, natural response to injury & operative treatment. *J Bone Joint Surg Br* 72(4): 615–617
11. Niskanen R et al (1996) Serum C-reactive protein levels after total hip and knee arthroplasty. *J Bone Joint Surg Br* 78:431–433
12. National Academy of Sciences, National Research Council, Division of Medical Sciences, Ad Hoc Committee on Trauma (1964) Post-operative wound infection: the influence of ultraviolet irradiation on the operating room and of various other factors. *Ann Surg* 160(2):1–192
13. Olson M, O'Connor M, Schwartz ML (1984) Surgical wound infection: a five year prospective study of 20,193 wounds at Minneapolis V.A. Medical Centre. *Ann Surg* 199:253–259
14. Efem SEE, Aja A, Inyang U (1986) Surgical wound infection rate in Calabar University Teaching Hospital, Calabar. *West Afr J Med* 5:61–68
15. Ojiegbe GC, Njoku-obi AN, Ojukwu JO (1990) Incidence and parametric determinants of post-operative wound infections in a University Teaching Hospital. *Cent Afr J Med* 36:63–67
16. Cruse PJE, Foord R (1980) The epidemiology of wound infection: a ten-year prospective study of 62,939 wounds. *Surg Clin North Am* 60:27–40

17. Moylan AJ, Kennedy VB (1980) The importance of gown and drape barriers in the prevention of wound infection. *Surg Gynecol Obstet* 151:465–470
18. Tillett WS, Francis T (1930) Serological reaction in pneumonia with non protein somatic function of pneumococcus. *J Exp Med* 52:561
19. Werner M (1969) Serum protein changes during the acute phase reaction. *Clin Chem Acta* 25:299–305
20. Schentag JJ, O'Keiffe D, Marmion M et al (1984) C-reactive protein as an indicator of infection relapse in patients with abdominal sepsis. *Arch Surg* 119(3):300–304
21. Mayer AD, Mc Mahon MJ, Bowen M et al (1984) C-reactive protein; an aid to assessment and monitoring of acute pancreatitis. *J Clin Pathol* 37:207