Evaluation of inflammatory response in patients undergoing surgical treatment for early and delayed femoral fractures

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Submitted: 17 July 2016
Accepted: 21 August 2016

Arch Med Sci 2019; 15, 1: 141–145
DOI: https://doi.org/10.5114/aoms.2016.63013

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Abstract

Introduction: It has been shown that long bone fractures are correlated with the inflammatory response. In the initial injury, surgical reduction and fixation of fractures induce the immunoinflammatory response. This study aimed to evaluate serum variation of inflammatory markers in patients undergoing surgical treatment for early and delayed femoral fractures.

Material and methods: This study aimed to evaluate serum variation of inflammatory markers in patients undergoing surgical treatment for early and delayed femoral fractures. The patients were randomly divided into two groups using the method of block randomization including early surgery (within 24 h) and delayed surgery (after 48 h). Serum levels of inflammatory markers in both groups including interleukin (IL)-1, 5, 6, tumor necrosis factor α (TNF-α) and interferon γ (IFN-γ) were determined using specific kits. From each patient 10 ml blood was collected for cytokine assay in their serum.

Results: Our findings suggest that serum levels of IL-8 were markedly decreased from 12 h until 48 h postoperatively ($p < 0.05$). Moreover, the results indicated that serum levels of TNF-α were significantly increased in the early hours, but after 48 h a decreasing trend was detected ($p < 0.05$). Furthermore, serum levels of IL-10, IFN-γ, and IL-6 were significantly increased from 12 h until 48 h postoperatively ($p < 0.05$).

Conclusions: The inflammatory status of the patient may be a useful adjunct in clinical decisions. With an improved understanding of the molecular basis of the inflammatory response, and by identifying relevant clinical markers of inflammation, surgeons can better manage the timing of surgical stabilization.

Key words: inflammation, tumor necrosis factor, interleukin, patient, femoral.

Introduction

The inflammatory response has been recognized as a physiologic reaction to injury. Surgery was shown to be the cause of a systemic response, the extent of which is moderated by different parameters such as the health and nutritional status of the patient, the severity of recent trauma and the presence of any preexisting physiologic derangement, and

the magnitude, duration, and technique of surgery [1–4]. It has been shown that hip fracture and surgery in aged rats induced a systemic inflammatory response and lung injury correlated with increased susceptibility to infection during the acute phase after injury and surgery. It has been shown that long bone fractures are correlated with the development of the systemic inflammatory response syndrome and are strongly associated with multi-organ failure, sepsis, hospital length of stay, and mortality [5–7]. Different components of the immune system have been demonstrated to be involved in this process, such as inflammatory cytokines, leukocyte adhesion molecules, growth factors, nitric oxide, platelet-activating factors, and the activation of local and systemic polymorphonuclear neutrophils (PMNs), lymphocytes, and macrophages. This complex response arises from the interplay between various mediators produced at the site of injury, including cytokines [8]. These mediators can regulate gene transcription, and modify intracellular signaling pathways [9]. In the initial injury, surgical reduction and fixation of fractures induce the immunoinflammatory response [10]. Therefore, modulation of cytokine release has been considered a tempting strategy [11]. This study aimed to evaluate serum variation of inflammatory markers in patients undergoing surgical treatment for early and delayed femoral fractures.

Material and methods

Patients and serum parameters

This study is a randomized clinical trial and all samples were conducted among patients with femoral fractures, between 2014 and 2015 in Rasol Hospital of Tehran. This study was approved by the Ethical Committee for Clinical Research of the Hospital, and informed consent was obtained from all the patients. It is worth noting that the criteria included ages of 20 to 50 years, and patients with femoral shaft fractures without injury in other parts of the body were recruited for our study. The patients were randomly divided into two groups using the method of block randomization including early surgery (within 24 h) and delayed surgery (after 48 h). Serum levels of inflammatory markers in both groups including interleukin (IL)-1, 5, 6, tumor necrosis factor (TNF)-α and interferon (IFN)-γ were determined by specific kits. From each patient 10 ml of blood was collected for cytokine assay in their serum.

Patients with the following criteria were excluded from the study: patients who had chronic inflammatory disease or a history of trauma in the last month, patients who had suffered multiple organ damage in their recent trauma, and patients with complex fractures.

ELISA analysis

Serum was also separated from blood using centrifugation (2000×g for 15 min at 4°C). All samples were frozen at –20°C in sterile tubes until used for cytokine measurements by the ELISA method using commercial kits (BIORBYT).

Statistical analysis

All variables were analyzed using the software SPSS version 16.0 (SPSS Inc, IL, USA). To compare levels of inflammatory markers including IL-1, IL-5, IL-6, TNF-α and IFN-γ the independent *t*-test was used. Differences were considered statistically significant when p was less than 0.05.

Results

Our findings suggest that serum levels of IL-8 were markedly decreased from 12 h until 48 h postoperatively ($p < 0.05$). Moreover, the results indicated that serum levels of TNF-α were significantly increased in the early hours, but after 48 h a decreasing trend was detected ($p < 0.05$). Furthermore, serum levels of IL-10, IFN-γ, and IL-6 were significantly increased from 12 h until 48 h postoperatively ($p < 0.05$) (Table I).

Discussion

The complex inflammatory response arises from the interplay between various mediators produced at the site of injury, including cytokines [8]. These mediators can regulate gene transcription, and modify intracellular signaling pathways [9]. In the initial injury, surgical reduction and fixation of fractures induce the immunoinflammatory response. Therefore, modulation of cytokine release has been considered a tempting strategy [10]. Zhang *et al.* reported that hip fracture and surgery in aged rats induced a systemic inflammatory response and lung injury correlated with increased susceptibility to infection during the acute phase after injury and surgery [11].

In the present study, our findings suggest that serum levels of IL-8 were markedly decreased from 12 h until 48 h postoperatively. Moreover, the results indicated that serum levels of TNF-α were significantly increased in the early hours, but after 48 h a decreasing trend was detected. Furthermore, serum levels of IL-10, IFN-γ, and IL-6 were significantly increased from 12 h until 48 h postoperatively.

Neumaier *et al.* [12] reported that the C-reactive protein (CRP) values were significantly lower in early surgery within 24 h after trauma than in delayed surgery. Moreover, they found that a lower postoperative inflammatory reaction after early surgery of hip fractures provides a better out-

Table I. Comparison of serum levels of inflammatory markers in patients with different times

Number of patient	Gender	Sampling dates and times	IL-8 concentration	TNF-α concentration	IL-6 concentration	IFN-γ [ng/µl]	IL-10 [ng/µl]
1	M	0		0			
2	M	24 h					
3	M	48 h	0				46.439
4	M	0	0				32.874
5	M	24 h		0			
6	M	48 h		0			
7	M	48 h	0	0	88.27	108.474	258.598
8	M	24 h	0	28.871	374.836	0	164.614
9	M	0	0	1.679	56.695	141.466	572.878
10	F	48 h	57.992	135.058	393.762	104.554	395.093
11	F	24 h	0	0	29.466	2.842	130.332
12	F	0	35.064	176.806	314.69	78.371	576.652
13	M	0	0	0	76.802	11.52	118.66
14	M	48 h	0	0	55.636	11.52	86.575
15	M	24 h	0	0	67.127	1.066	167.327
16	M	0	0	37.275	79.316	6.946	580.454
17	M	24 h	0	33.393	64.027	0	416.276
18	M	48 h	0	0	60.903	16.441	13.428
19	M	48 h	0	7.839	48.118	0	492.988
20	M	24 h		22.465	131.388	61.03	159.263
21	M	48 h	0	0	66.612	4.822	23.186
22	M	24 h	0	318.514	128.109	19.013	1479.571
23	M	0	0	357.436	107.241	41.772	1476.714
24	M	0	0	0	28.277		67.388
25	M	48 h	0	0	20.93	54.418	32.107
26	F	24 h	0	1.679	46.482	93.063	1346.595
27	F	0	0	2.549	21.558	96.848	943.885
28	M	24 h	0	0	70.716	4.822	11.433
29	M	24 h	0	0	58.805	6.946	9.475
30	M	0	0	0	612.362	11.52	24.638
31	M	48 h	0	228.051	202.519	9.184	1441
32	M	48 h	0	0	148.101	29.939	35.193
33	M	24 h	0	0	214.574	89.322	94.726
34	M	0	0	0	180.941	74.811	74.866
35	M	24 h	0	28.871	4.063	196.526	85.576
36	M	48 h	0	91.92	185.003	4.822	315.489
37	M	0	0				
38	M	24 h	0	66.171	115.362	64.406	230.739
39	M	0					
40	M	24 h	0	0	72.753	0	15.458
41	M	0 h	0	0	155	0	49.781
42	M	48 h	0	0	350.672	4.822	61.946
43	M	0		0			
44	M	24 h	0	24.823	253.136	74.811	86.575
45	M	24 h	0		117.261	201.428	37.545

Table I. Cont.

Number of patient	Gender	Sampling dates and times	IL-8 concentration	TNF-α concentration	IL-6 concentration	IFN-γ [ng/µl]	IL-10 [ng/µl]
46	M	48 h		0	77.306	74.811	197.418
47	M	24 h	0	0.722	70.205	145.795	237.491
48	M	0	0	1.679	86.787	0	279.141
49	M	0	0	20.067	59.856	0	17.522
50	F	48 h	0	0	127.64	0	24.638
51	F	24 h	0	0	148.562	0	11.433
52	F	0	0	0	38.141	11.52	32.874
53	M	48 h	0	0	123.879	120.506	80.645
54	M	24 h	0	0	66.097	137.182	74.866
55	M	0	0	2.549	46.482	0	37.545
56	M	48 h	0	0	39.27	0	11.433
57	F	24 h	0	0	108.68	0	12.759
58	F	0	0	0	31.234	0	17.522
59	M	24 h	0	0	113.458	16.441	38.335
60	F	24 h	0	20.067	177.323	54.418	253.198
61	F	0	0	11.905	29.466	51.183	55.772
62	M	0	0	19.461	42.069	54.418	35.973
63	M	48 h	0	0	50.285	29.939	21.748
64	F	48 h	0	0	72.753	16.441	19.619

come when treated with arthroplasty. Findings of Harwood *et al*. [13] support the continued use of damage control procedures in severely injured patients and complement data already available, suggesting that a damage control orthopedics (DCO) approach reduces the subsequent inflammatory response. Moreover, they concluded that the inflammatory status of the patient may be important in clinical decision making regarding the timing of conversion to an intramedullary device. In agreement with our study, they found that the pattern of serum IL-6, keratinocyte, IL-10, and IL-1 release was dynamic, but no significant elevation in TNF-α was detected. The early hepatic and pulmonary infiltration of polymorphonuclear cells occurred in the absence of significantly elevated serum cytokine levels, indicating that either early minor changes with an imbalance in inflammatory mediators or locally produced cytokines may initiate this process. Nakamura *et al*. in Japan reported that IL-1 and IL-6 and TNF-α were increased after femoral fractures and that they originated from synovial cells [14]. It has been reported that intramedullary nailing fixation resulted in an increase in the level of inflammatory cytokines in animal models. As a matter of fact, it has more adverse effects on the inflammatory response, system stress, and multiple organs [14].

A previous study found that the serum levels of IL-6 and IL-8 in the cerebrospinal fluid were increased, and it raises the possibility that IL-8, acting in the central nervous system (CNS), plays a role in the postinjury syndrome. The mechanism by which CNS IL-8 is produced in trauma is unclear, but a physiological role is supported by the known ability of the CNS to produce IL-8 and the presence of receptors for its action in the CNS [15, 16].

In an animal model, it has been reported that immune cell performance can be increased, and this phenomenon results in an increase in cytokine secretion levels [17]. A previous study evaluated the change in IL-6 levels perioperatively in patients treated for femoral shaft fracture. It was reported that damage control procedures provoked a significantly smaller increase in IL-6 levels when compared with those observed after primary intramedullary nail (IMN). Furthermore, similar studies on bone fracture were conducted previously by other authors [18–20]. In conclusion, the inflammatory status of the patient may be a useful adjunct in clinical decision making. With an improved understanding of the molecular basis of the inflammatory response, and by identifying relevant clinical markers of inflammation, surgeons can better manage the timing of surgical stabilization.

Conflict of interest

The authors declare no conflict of interest.

References

1. Galle C, De Maertelaer V, Motte S. Early inflammatory response after elective abdominal aortic aneurysm repair: a comparison between endovascular procedure and conventional surgery. J Vasc Surg 2000; 32: 234-46.
2. Munoz M, Garcia-Vallejo JJ, Sempere JM, et al. Acute phase response in patients undergoing lumbar spinal surgery: modulation by perioperative treatment with naproxen and famotidine. Eur Spine J 2004; 13: 467-73.
3. Pape HC, Schmidt RE, Rice J, et al. Biochemical changes after trauma and skeletal surgery of the lower extremity: quantification of the operative burden. Crit Care Med 2000; 28: 3441-8.
4. Wakai A, Wang JH, Winter DC, et al. Tourniquet-induced systemic inflammatory response in extremity surgery. J Trauma 2001; 51: 922-6.
5. Faist E, Baue AE, Dittmer H, Heberer G. Multiple organ failure in polytrauma patients. J Trauma 1983; 23: 775-87.
6. Lane MK, Nahm NJ, Vallier HA. Morbidity and mortality of bilateral femur fractures. Orthopedics 2015; 38: e588-92.
7. Giannoudis PV, Cohen A, Hinsche A, et al. Simultaneous bilateral femoral fractures: systemic complications in 14 cases. Int Orthop 2000; 24: 264-7.
8. Rankin JA. Biological mediators of acute inflammation. AACN Clin Issues 2004; 15: 3-17.
9. Robinson CM. Current concepts of respiratory insufficiency syndromes after fracture. J Bone Joint Surg Br 2001; 83: 781-91.
10. Finnerty CC, Herndon DN, Przkora R, et al. Cytokine expression profile over time in severely burned pediatric patients. Shock 2006; 26: 13Y19.
11. Zhang H, Sun T, Liu Z, Zhang J. Systemic inflammatory responses and lung injury following hip fracture surgery increases susceptibility to infection in aged rats. Mediators Inflamm 2013; 2013: 536435.
12. Neumaier M, Vester H, Martetschläger F, Freude T. Optimaler Zeitpunkt zur prothetischen Versorgung von Schenkelhalsfrakturen 2011; 82: 921-6.
13. Harwood PJ, Giannoudis PV, van Griensven M, Krettek C, Pape HC. Alterations in the systemic inflammatory response after early total care and damage control procedures for femoral shaft fracture in severely injured patients. J Trauma 2005; 58: 446-52.
14. Nakamura H, Yoshino S, Shiga H, Tanaka H, Katsumata S. A case of spontaneous femoral neck fracture associated with multicentric reticulohistiocytosis: oversecretion of interleukin-1beta, interleukin-6, and tumor necrosis factor alpha by affected synovial cells. Arthritis Rheum 1997; 40: 2266-70.
15. Tiansheng S, Xiaobin C, Zhi L, et al. Is damage control orthopedics essential for the management of bilateral femoral fractures associated or complicated with shock? An animal study. J Trauma 2009; 67: 1402-11.
16. Buzdon MM, Napolitano LM, Shi HJ, et al. Femur fracture induces site-specific changes in T-cell immunity. J Surg Res 1999; 82: 201-8.
17. Pape HC, Grimme K, Van Griensven M, et al. Impact of intramedullary instrumentation versus damage control for femoral fractures on immunoinflammatory parameters: prospective randomized analysis by the EPOFF Study Group. J Trauma 2003; 55: 7-13.
18. Bączyk G, Samborski W, Jaracz K. Evaluation of the quality of life of postmenopausal osteoporotic and osteopenic women with or without fractures. Arch Med Sci 2016; 12: 819-27.
19. Dytfeld J, Marcinkowska M, Drwęska-Matelska N, Michalak M, Horst-Sikorska W. Association analysis of the COL1A1 polymorphism with bone mineral density and prevalent fractures in Polish postmenopausal women with osteoporosis. Arch Med Sci 2016; 12: 288-94.
20. Sadr SSH, Javanbakht J, Norouzi Javidan A, Ghaffarpour M, Safoura Khamse S. Descriptive epidemiology: prevalence, incidence, sociodemographic factors, socioeconomic domains, and quality of life of epilepsy: an update and systematic review. Arch Med Sci 2018; 14: 717-24.