HLA-DR and IL-17A serum concentration in pre and post-operative periods among Laparoscopic versus Conventional surgery in Iraqi Patients.

Dr. Manal M. Kadhim[1], Dr. Adel Sh. Al tamimi[2], Shujon H.Jadaan[3]
[1,3] Department of Medical Microbiology, College of Medicine, Al-Qadisiyah University, Diwaniya, Iraq
[2] Department of surgery, College of Medicine, Al-Qadisiyah University, Diwaniya, Iraq,
shujon.hasan@yahoo.com

Abstract

Background: During the past decade, a number of experimental and clinical studies have provided evidence that surgical and trauma injury markedly affects the immune system, including both the specific and the nonspecific immune responses .Aim: the present study was conducted to evaluate the effect of conventional and laparoscopic surgery on HLA-DR lymphocyte and IL-17. Methods: A total of 50 patients were included in the study who were admitted to hospital from the period between March to August 2018 for different types of surgery, and divided into open surgery group (25) patients and laparoscopic surgery group (25) patients. Two blood sample were taken from each patients one before anesthesia and the other sample (12-24) hr. later postoperatively for HLA-DR lymphocyte flowcytometry and serum concentration of IL-17 by ELISA technique. Results: The count of CD8 lymphocytes, in Conventional Surgery group post-operative was significantly less than that of Laparoscopic surgery group, Mean serum concentration of IL- 17 in Conventional Surgery group post-operative was92.89±8.04 versus Laparoscopic Surgery group; 45.02±1.43. Conclusion: The results in the study showed that the HLA-DR expression was depressed in conventional surgery and significantly lower than that in patient with lap. Surgery intervention.

Keyword: surgery, IL-17, HLA-DR. laparoscopic, conventional

1. Introduction

During the past decade, a number of experimental and clinical studies had provided evidence that surgical and trauma injury markedly affected the immune system, including both the specific and the nonspecific immune responses [1].

Major body injury, surgical or accidental, evokes reproducible metabolic, hormonal, hemodynamic, and immunological responses. Although the immunological responses that occur during surgical injury have been widely investigated, there is growing interest in the cytokine factors that mediate aspects of the acute phase response to injury and inflammation. Among the cytokines, tumor necrosis factor alpha, interleukin-1, and interleukin-6 (IL-6) are considered the major regulators of the acute phase response to inflammation and tissue injury [2]. Surgery induced immunosuppression has considerable implications for patients. It is associated with impaired wound healing, delayed post-operative recovery and development of postoperative septic complications and tumor metastasis formation [3]. Cytokines are key modulators of inflammation and play both inflammatory and ant inflammatory roles [4]. Over past decades, cytokines have gained more attention in the understanding of physiological changes after trauma or surgery. Cytokines participate in acute and chronic inflammation in a complex network of interactions [5]. Any degree of surgical trauma leads to changes in the hemodynamic, metabolic, and immune response of patients during the postoperative period [6]. IL-17 is mainly produced by activated T lymphocytes and stimulates fibroblasts, endothelial cells, macrophages and epithelial cells to produce multiple proinflammatory mediators such as IL-1, IL-6, IL-8 and TNF-α, as well as the activation of NOS2, metalloproteinases and chemokines, inducing inflammation and increasing the expression of intercellular adhesion molecule-1 (ICAM-1) [7]. IL-17A is involved in the protection of the organism against extracellular bacteria and fungi due to its capacity of recruiting neutrophils to the sites of infection. A pathological role of this cytokine has been demonstrated in various models of autoimmune diseases such as experimental autoimmune encephalitis and rheumatoid arthritis [8]. Human Leukocyte Antigen-DR (HLA-DR), one of the Major Histocompatibility Complex class-II proteins expressed on monocytes, macrophages and dendritic cells, is a crucial molecule for the presentation of antigen to lymphocytes and the initiation of adaptive immune responses [9]. Reduced levels of HLA-DR expression on blood monocytes following surgery is proposed as a potential biomarker for surgery-induced immunosuppression and the development of postoperative infections [10]. The analysis of HLA-DR expression on monocytes seems to be a useful tool to quantify the immune status of patients. There is a negative correlation between surface expression of HLA-DR on monocytes and the incidence of infectious diseases in trauma patients [11], and a decreased HLA-DR expression in septic shock is associated with in- creased mortality risk [12].

2. Materials and Methods

The current study was carried on 50 patients (18 males, 32 females) age range between 20-60 years from March to August2018. We excluded patients with Parasite, malignancy, regional or local anesthesia, Psychiatric and patients with

doi.org/10.31838/hiv22.02.112 Received: 13.01.22, Revised: 15.05.22, Accepted: 13.08.22

chronic liver diseases or kidney disorders.25 patients with conventional open surgery and 25 patients with laparoscopy surgery. A ten ml of blood samples were collected and withdrawn from each patient within 2 different period including five ml preoperative and five ml post-operative by vein puncture using disposable syringes under aseptic technique 2 ml of each sample were transferred to sterile Gel tube, and allow to clot at room temperature and centrifuge at 2500 rpm for 10 minutes and the separated serum was saved in Eppendorf tubes and immediately frozen at -20 C till further use to avoid repeated thawing and freezing for IL17 ELISA Kit (ElabscienceUSA) test. The remaining (3ml) were transferred into tube with EDTA maintained at room temperature until analysis sample at same day for CD4 lymphocyte count by Flowcytometry kit for HLA-DR Thermo Fisher/ Bioscience™ USA. This study was in agreement with ethics of Al-Diwaniya Teaching Hospital and verbal informed consent was obtained from all participants.

3. Results

Table (1), declared a non-statistical difference in Lap.group and in Con. group regarding the risk factors as p<0.05. Patients in both groups were comparable in mean serum concentration of IL-17A and HLA-DR expression table (2and 3). Mean serum concentration of IL- 17 in Conventional Surgery group post-operative was significantly higher than that of Laparoscopic Surgery group; 92.89±8.04 versus 45.02±1.43 and P-value was (P < 0.0001), and the HLA-DR expression depressed in post-operative period in the Conventional Surgery group more than that of Laparoscopic Surgery group; 16.88±0.58 versus 18.22±0.46 and P-value was (P <0.078). Figure (1) showed the flow cytometric analysis of HLA-DR expression on lymphocyte cells in Laparoscopic. surgical patients before taken anesthesia (A) and after surgery (B), while the figure (2) showed the flowcytometric analysis of HLA-DR expression on lymphocyte cells in conventional surgical patients before taken anesthesia (A) and after surgery (B).

Table 1: Distribution of Laparoscopic and Conventional groups according to risk factors.

Risk Factors	Conventional Surgery group NO. (%)		Laparoscopic Surgery Group NO. (%)	
	Positive	Negative	Positive	Negative
Diabetes mellitus	5(20)	20(80)	4(16)	21(84)
Smoking	7(28)	18(72)	6(24)	19(76)
Hypertension	11(44)	14(56)	4(16)	21(84)
P value	0.17(NS)		0.704(NS)	

Table (2): Serum concentration of IL-17 in Laparoscopy and Conventional surgery groups before and after surgery.

Serum IL17 pg/ml	Laparoscopic Surgery group		Conventional Surgery group		P value
	pre	post	pre	post	
mean±SE	36.32±0.46	45.02±1.43	38.4±1.05	92.89±8.04	0.0001
Range	8.13	31.64	19.28	179.65	(S)
Median	36.08	42.25	37.11	83.35	
Inter-quartile range	4.06	9.83	7.35	43.82	

Table (3): The compassion between the study groups pre-operatively and post operatively regarding HLA-DR expression.

HLA-DR expression	Laparoscopic Surgery group		Conventional Surgery group		P
	pre	post	pre	post	
Mean ±SE	20.49±0.49	18.22±0.46	23.42±0.47	16.88±0.58	0.078
Range	7.93	8.85	7.61	10.33	
Median	20.4	18.17	23.62	17.45	
Inter-quartile range	5.07	3.72	4.02	4.77	

4. Discussion

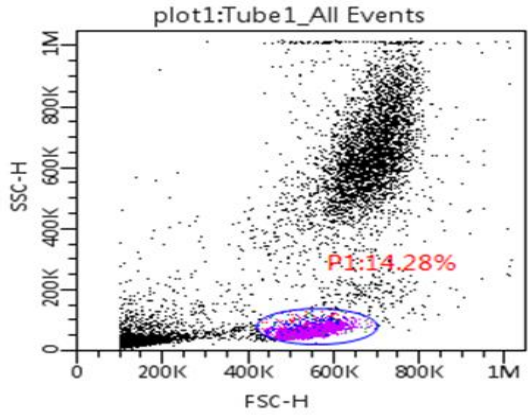
Significant difference in surgery and risk factors were observed in this study, which agree with Simpson et al. [13] they founded no statistical significant difference between smoking, hypertension and diabetic with surgery. Any surgical procedure is a controlled tissue injury. Traumatic tissue injury leads to release of intracellular substances, which act as endogenous triggers of the immune system, often referred to as alarmins or damage-associated molecular patterns (DAMP) [14].in this study, the IL-17A highly increase in open surgery than Lap.surgery, this results agree with Borges et al. [7] how studied 40 women with cholelithiasis. The patients were divided into two groups submitted to conventional laparoscopic cholecystectomy and to single-port laparoscopic cholecystectomy. Significantly higher postoperative expression of IL-17A was detected in patients submitted to single-port laparoscopic cholecystectomy IL-17A was (0.28±0.03) when compared to preoperative levels (0.27±0.03), in conventional laparoscopic cholecystectomy the levels IL-17A post-operative was (0.24±0.02) while preoperative was (0.26±0.02) Significant postoperative expression of IL-17 was observed in the group submitted to single-port laparoscopic cholecystectomy when compared to preoperative levels, indicating that surgical stress in this group was higher compared to the conventional laparoscopic cholecystectomy.

Both laparoscopic and open cholecystectomy procedures altered the inflammatory milieu of patients in the postoperative period, and inflammation caused by the

laparoscopic procedure was significantly less [6], however, the inflammatory response to the same type of trauma can vary from patient to patient [15]. Serum cytokine levels are related to the magnitude of operative trauma and therefore, they can be used as objective biochemical markers that reflect the surgical and tissue trauma [16, 17]. So the levels of interleukins are increased and significantly higher in patients undergoing open procedures demonstrating that laparoscopic method displays less inflammatory response compared to open technique [18].The results of current study in concordance with Lachmann et al. [19] when they analyzed HLA-DR by flowcytometry in patient with elective esophageal or pancreatic resection before and after surgery they found significant reduction in HLA-DR postsurgical patients. Handy et al. [20] they Studied cytometric analysis for HLA-DR in patients have open abdominal surgery and other patients with laparoscopic bariatric procedures, depression of HLA-DR expression appeared in both group after surgery but open abdominal surgery showed greater reductions in HLA-DR expression than laparoscopic gastric banding, decline was observed as early as 60min after surgical incision and accentuated 24h after surgery monocyte HLA-DR expression as an early biomarker in predicting postoperative complications. While decreased monocyte HLA-DR expression can be associated with increased infection rates or poor outcome in major surgery or trauma [10].

5. Conclusions

Any surgical trauma causes depression in immune system but Conventional surgery causes immune depression more than Laparoscopic surgery. The beneficial effects of laparoscopic surgery may therefore relate, in part, to less surgical stress in the immediate post-operative period. In this study we found a reduced inflammatory response after laparoscopic surgery compared with open, represented by decreased levels of IL- 17. In the open surgery group we observed a post-operative decrease in the HLA-DR of peripheral monocyte. Patients who undergone LC showed a normal level of HLA-DR expression.



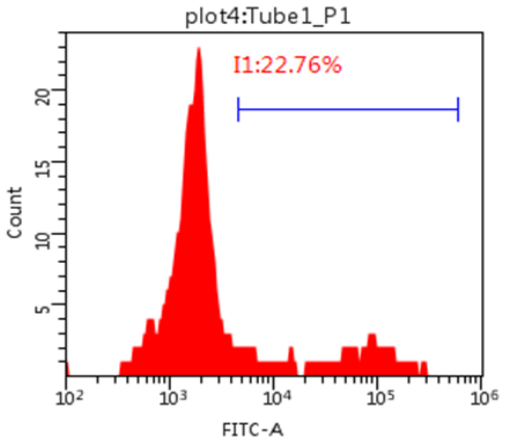


Figure (1): Flow cytometric analysis of HLA-DR expression on lymphocyte cells in Laparoscopic .surgical patients before taken anesthesia (A) and after surgery (B).

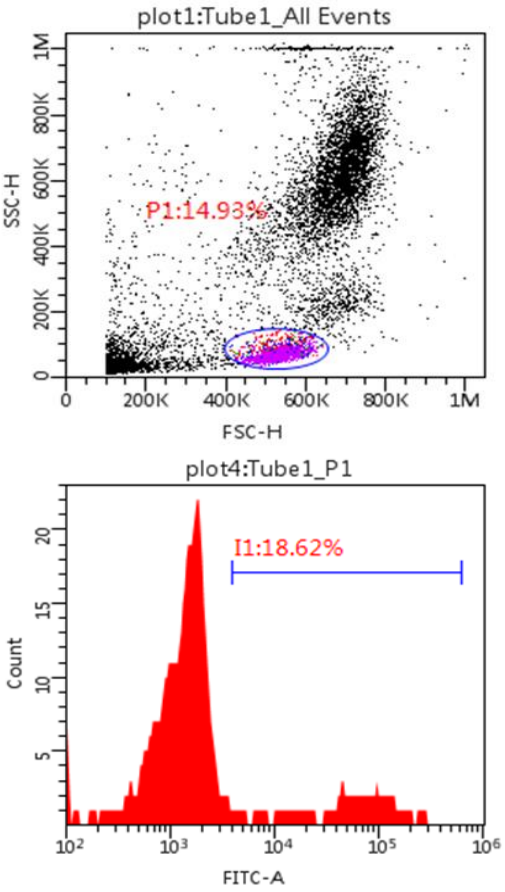
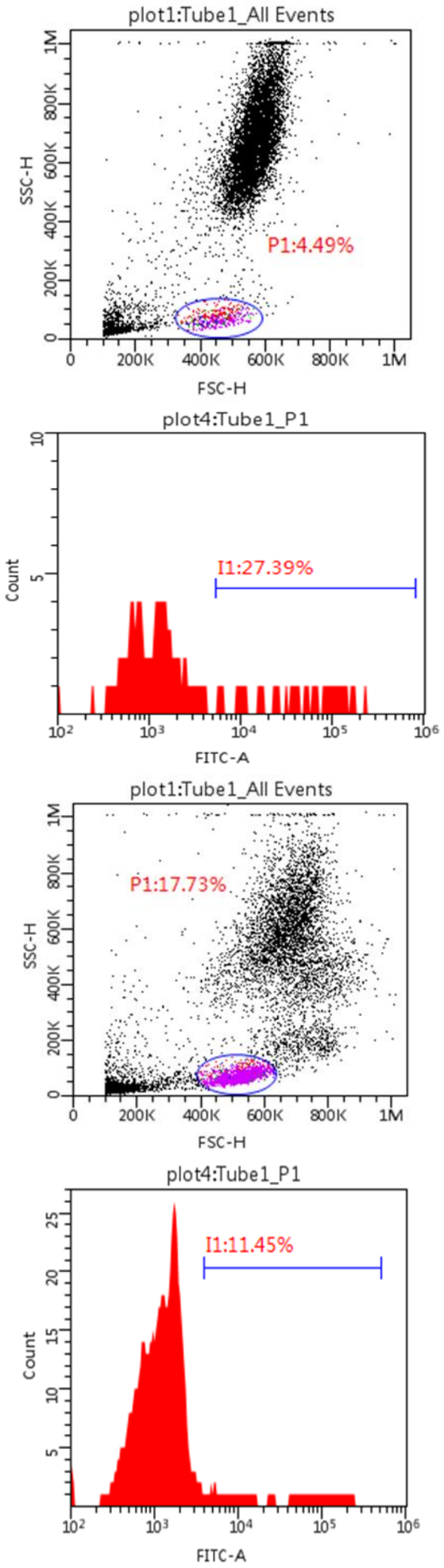


Figure (2): Flow cytometric analysis of HLA-DR expression on lymphocyte cells in conventional surgical patients before taken anesthesia (A) and after surgery (B).



Reference

1. Kimura F, Shimizu H, Yoshidome H, et al. Immunosuppression following surgical and traumatic injury. Surgery Today. 2010;40(9):793-808. https://doi.org/10.1007/s00595-010-4323-z.
2. Sari R, Sevinc A. The effects of laparascopic cholecystectomy operation on C-reactive protein, hormones, and cytokines. Journal of Endocrinological Investigation. 2004;27(2):106-10. https://doi.org/10.1007/BF03346253.
3. Hogan BV, Peter MB, Shenoy HG, et al. Surgery induced immunosuppression. Surgeon. 2011;9(1):38-43. https://doi.org/10.1016/j.surge.2010.07.011.
4. Lenz A, Franklin GA, Cheadle WG. Systemic inflammation after trauma. Injury. 2007;38(12):1336-45. https://doi.org/10.1016/j.injury.2007.10.003.
5. Hsing C-H, Wang J-J. Clinical implication of perioperative inflammatory cytokine alteration. Acta Anaesthesiologica Taiwanica. 2015;53(1):23-8. https://doi.org/10.1016/j.aat.2015.03.002.
6. Sayyed E, Hussain N, Najmul I, et al. < A> prospective study of altered inflammatory response and its clinical outcome following laparoscopic and open cholecystectomy. 2017;42(4):347-53. https://pesquisa.bvsalud.org/portal/resource/pt/emr-191161
7. Borges MdC, Takeuti TD, Terra GA, et al. Comparative analysis of immunological profiles in women undergoing conventional and single-port laparoscopic cholecystectomy. ABCD Arquivos Brasileiros de Cirurgia Digestiva (São Paulo). 2016;29(3):164-9. https://doi.org/10.1590/0102-6720201600030009.
8. Normanton M, Marti LC. Current data on IL-17 and Th17 cells and implications for graft versus host disease. Einstein (Sao Paulo). 2013;11:237-46. https://doi.org/10.1590/S1679-45082013000200019.
9. Oczenski W, Krenn H, Jilch R, et al. HLA-DR as a marker for increased risk for systemic inflammation and septic complications after cardiac surgery. Intensive Care Medicine. 2003;29(8):1253-7. https://doi.org/10.1007/s00134-003-1826-8.
10. Haveman JW, van den Berg AP, Verhoeven ELG, et al. HLA-DR expression on monocytes and systemic inflammation in patients with ruptured abdominal aortic aneurysms. Critical Care. 2006;10(4):R119. https://doi.org/10.1186/cc5017.
11. Menges P, Kessler W, Kloecker C, et al. Surgical trauma and postoperative immune dysfunction. European Surgical Research. 2012;48(4):180-6. https://doi.org/10.1159/000338196.
12. Cavaillon J-M, Adib-Conquy M. Determining the degree of immunodysregulation in sepsis. Acute Kidney Injury. 2007;156:101-11. https://doi.org/10.1159/000102075.
13. Simpson AM, Donato DP, Kwok AC, et al. Predictors of complications following breast reduction surgery: A National Surgical Quality Improvement Program study of 16,812 cases. Journal of Plastic, Reconstructive & Aesthetic Surgery. 2019;72(1):43-51.

https://doi.org/10.1016/j.bjps.2018.09.002.
14. Stoecklein VM, Osuka A, Lederer JA. Trauma equals danger—damage control by the immune system. Journal of leukocyte biology. 2012;92(3):539-51. https://doi.org/10.1189/jlb.0212072.
15. Boo YJ, Kim WB, Kim J, et al. Systemic immune response after open versus laparoscopic cholecystectomy in acute cholecystitis: a prospective randomized study. Scandinavian Journal of Clinical and Laboratory Investigation. 2007;67(2):207-14. https://doi.org/10.1080/00365510601011585.
16. Grande M, Tucci GF, Adorisio O, et al. Systemic acute-phase response after laparoscopic and open cholecystectomy. Surgical Endoscopy And Other Interventional Techniques. 2002;16(2):313-6. https://doi.org/10.1007/s00464-001-9042-5.
17. Hildebrandt U, Kessler K, Plusczyk T, et al. Comparison of surgical stress between laparoscopic and open colonic resections. Surgical Endoscopy And Other Interventional Techniques. 2003;17(2):242-6. https://doi.org/10.1007/s00464-001-9148-9.
18. Mijatovic S, Stefanovic B, Jeremic V, et al. Evaluation of serum cytokines levels in patients undergoing laparoscopic versus conventional cholecystectomy. The Journal of Pharmacy and Pharmacology. 2016;4:10-6. https://doi.org/10.17265/2328-2150/2016.01.003.
19. Lachmann G, von Haefen C, Kurth J, et al. Innate immunity recovers earlier than acquired immunity during severe postoperative immunosuppression. Int J Med Sci. 2018;15(1):1-9. https://doi.org/10.7150/ijms.21433.
20. Handy J, Scott A, Cross A, et al. HLA-DR expression and differential trafficking of monocyte subsets following low to intermediate risk surgery. Anaesthesia. 2010;65(1):27-35. https://doi.org/10.1111/j.1365-2044.2009.06161.x.