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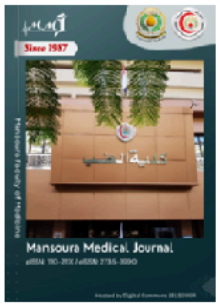

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STUDY OF CHANGES IN THE SERUM LEVELS OF GRANULOCYTE MACROPHAGE COLONY STIMULATING FACTOR (GMCSF) AND INTERLEUKIN ONE BETA (IL-I B) IN COLONIC CANCER

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ABSTRACT

It is well known that GMCSF and IL-IB are important factors that modulate host immune response against cancer and consequently, they are expected to be high in patients with colonic carcinoma. The aim of this study was to herald the changes that occur in the above mentioned factors in those patients one month after surgery which is the optimum time of relief of the immune system from operative trauma. In our study, thirty patients were submitted to surgery after being diagnosed as colonic carcinoma using the following methods : careful history taking, complete clinical examination, laboratory and radiological investigations, Finally, colonoscopy and biopsy was done. Assay of GMCSF & IL-IB in patients sera was done by using solid phase Enzyme Amplified Sensitivity Immune Assay (EASIA) performed on micro-titer plate (MEDGENIX) (Belgium).. Results revealed that both levels of GMCSF and IL-IB were found to be higher than normal in our patients and also there was highly significant increase in both factors after surgery. GMCSF (P = 0. 0008), IL.1B (P = 0. 0001).

Conclusion : surgery remains the gold standard in treatment of colonic carcinoma based on immunological background. The more radical the surgery, the better the immune response .

INTRODUCTION

Lymphocytes, which form about 20% of the white cell population, are principally of two forms, T and B cells (1). The functional classification of lymphocytes depends heavily on the detection of cell surface markers, which are identified using specific monoclonal antibodies (2). T cells which differentiate in the thymus (at least in the fetus) have a number of functions, including the regulation of the level of the immune response (3). Activated T cells carry class II major histocompatibility complex (MHC) molecules and have receptors for the cytokine interleukin-2 (IL. -2). Class II MHC and IL-2 receptors are not present on resting cells (4). B cells develop in the bone marrow or fetal liver. Activated B cells proliferate and mature under the influence of T cells (5). Also B cells produce immunoglobulins (or antibodies) in the presence of antigen specific T. cells (6). Null cells are distinct group of cells that show neither T nor B cell characteristics. They carry a mixture of surface markers including CD-16 and CD-56 (7). Natural killer (NK) cells are able to recognize determinants on some tumor cells and destroy them without the assistance of antibody. This killing is also MHC, Restricted (8). Killer (K) cells are in many ways similar to NK and are responsible for antibody dependent cytotoxicity (ADCC)(9). Cytokines are water soluble low molecular weight peptides, which act on or are produced by lymphocytes. More recent concepts see the cytokine as an important short-range messenger communicating between cells of the immune system (10). These compounds are thus called interleukins (IL-I to IL-3), interferons, macrophage colony stimulating factor (M-C-SF), and granulocyte macrophage colony stimulating factor (GM.CSF) (11). (IL-I Beta) may also stimulate B cells and induce macrophages to release prostaglandin E2 and tumor necrosis factor(TNF) (12). The colony stimulating factors are manufactured by a number of cells including stromal cells, fibroblasts , endothelium and lymphocytes (13). The role of immune system in the surveillance for and the destruction of tumor cells is now widely accepted specially in colonic carcinoma (14). This work aimed at telescoping of the changes that may occur in the serum levels of GMCSF and IL-IB in our patients due to colonic cancer, and the effect of surgery on the serum levels of those two important natural anti-cancer factors. The out come of this study will

either support or minimize the role of surgery in management of this aggressive type of cancer .

PATIENTS & METHODS

This study was done in Department of Surgery and Clinical immunology unit, Mansoura university hospital during the period from March 1999 till March 2000 inclusive . In our study, thirty patients with colonic cancer were diagnosed after using:

-careful history taking, thorough and complete clinical examination .

-Laboratory investigations that included : complete blood picture, serum creatinine, occult blood in stool, urine analysis, liver functions, blood sugar (fasting & 2 hours post prandial) .

- Radiological investigations included : plain x-ray chest, bone survey, abdominal ultrasound, intravenous urography, barium enema, abdominal C.T., MRI was done in selected cases when needed .

- Cystoscopy was done . Colonoscopy and biopsy was the claw for final diagnosis .

- In this study, our patients were (12) females and (18) males. Their ages ranged from (45-65) with mean age (50) years. According to the clinical presentation they were classified into 4 groups:

Group (1) : cachexia, anemia, anorexia (9 patients) .

Group (2) : occult blood in stool (8 patients) .

Group (3) : bleeding per rectum (8 patients) .

Group (4) : acute intestinal obstruction (5 patients).

Regarding metastatic work up, the evident distant metastases were present in (10 patients). Most type of metastases were present in the liver (7 patients), whereas bony metastases were present in (1 patient). Both liver and central lymph node metastases (para aortic, superior mesenteric or inferior mesenteric nodes) were present in (3 patients) and proved more during surgery. In our study, we considered central lymph nodes enlargement as distant metastasis equal to stage D Duke's classification. Pre-operative preparation of the colon was done using mechanical method (repeated enema) and chemical method (metronidazole infusion & oral neomycin). Intravenous fluids started 3 days before elective surgery. ECG and complete blood picture were done for all patients preop-

eratively . Fifteen of our patients received blood transfusion (after cross matching) pre-operatively, during surgery and immediately 24 hours after the end of surgery. The average amount of blood transfused to patients was (1-3 units) according to the severity of bleeding during heroic surgery and the degree of anemia . In patients presented with acute intestinal obstruction (5 Patients), mechanical and chemical preparation of the colon was not available and emergency surgery was done with on table lavage technique, also blood transfusion was given to them when needed. Regarding the surgical effort, generous abdominal mid line incision (upper & lower) passing through the umbilicus was done in all cases (30 patients). Radical surgery was done in (15 patients) in the form of complete resection of the tumor with safety margin (5 cm and below the tumor) and adequate lymph node resection up to the central nodes if enlarged . Primary anastomosis of the mechanically and chemically prepared colon was done . Surgical removal of localized focal lesion in the liver was done in (2 patients). The resected tumors were classified according to Duke's classification into A,B,C, and D. Colostomy was done in (10) patients, (5) covering colostomy and (5) were palliative. Post-operative intravenous fluids was continued for (5) days . Removal of Ryle's tube on 6th post-operative day. Removal of the intra abdominal rubber drains on 7th post-operative day after the patient started oral feeding and no evidence of fistula formation . Abdominal stitches were removed after (12-14 days) from operation. We intended to delay stitch removal due to long incision, critical state of the patients, heroic surgery and expected hypoproteinaemia. All patients were referred to medical oncology unit after removal of stitches to complete chemotherapy and/or radiotherapy for either adjuvant or palliative treatment .

- Regarding the immunological side of this research, assay of the serum level of GMCSF & IL-IB was done for all patients twice, once after reaching the final diagnosis & one month after the end of surgery as it was the optimum time of release of the immune system & body defense mechanisms from the surgical trauma. Assay of IL-I B and GMCSF was done on principles of the MEDGENIX EASIA, which is a solid phase Enzyme Amplified Sensitivity Immune assay performed on micro-titer plate. The sample

obtained from patients of non heparinized blood & the assay was 5 ml. was done by utilizing the patient's serum.

Results

Table (1) : Immunological data before and after surgery in all patients.

	Interleukin I-B	Granulocyte Macrophage CSF
Before surgery		
Mean	94.9	254
SE	22.8	68.1
After surgery		
Mean	371.7	1193.1
SE	62.8	217.5
P	< 0.0001	< 0.0001

Table (2) : Immunological data before and after surgery in females group.

	Interleukin I-B	Granulocyte Macrophage CSF
Before surgery		
Mean	105.8	272.4
SE	31	93.6
After surgery		
Mean	393	1137.6
SE	81.3	292.3
P	< 0.0001	< 0.0013

Table (3) : Immunological data before and after surgery in males group .

	Interleukin I-B	Granulocyte Macrophage CSF
Before surgery		
Mean	69.6	211.2
SE	23.9	68.3
After surgery		
Mean	322.2	1322.6
SE	92.7	265.1
P	< 0.0117	< 0.0109

Table (4) : Immunological data before and after surgery in cases without distant metastasis .

	Interleukin I-B	Granulocyte Macrophage CSF
Before surgery		
Mean	72.9	232.7
SE	20.6	77.5
After surgery		
Mean	349.3	922.4
SE	70.4	140.3
P	< 0.0001	< 0.0002

Figure (1) Correlation between age and Granulocyte Macrophage CSF before surgery

Figure (2) Correlation between age and Granulocyte Macrophage CSF after surgery

Figure (3) Granulocyte Macrophage CSF before and after surgery in cases with and without distant metastasis .

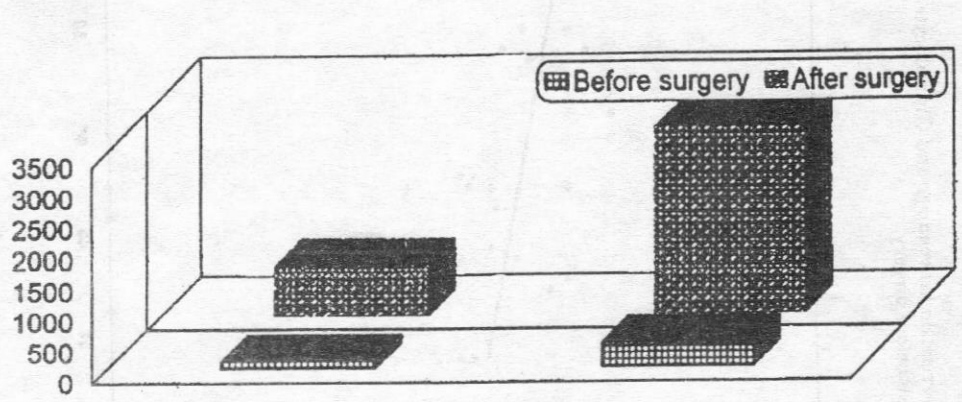


Figure (4) Granulocyte Macrophage CSF before and after surgery in different pathological stages

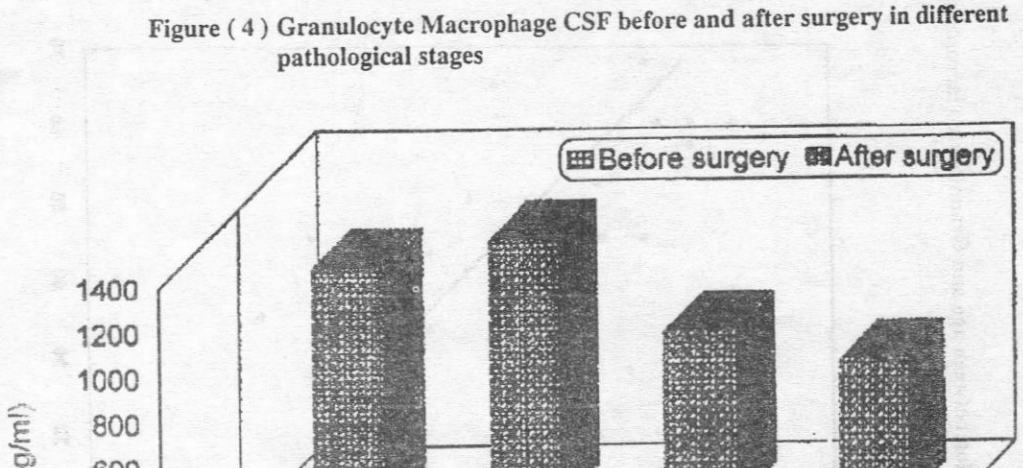


Figure (5) Interleukin 1-B before and after surgery in different pathological stages

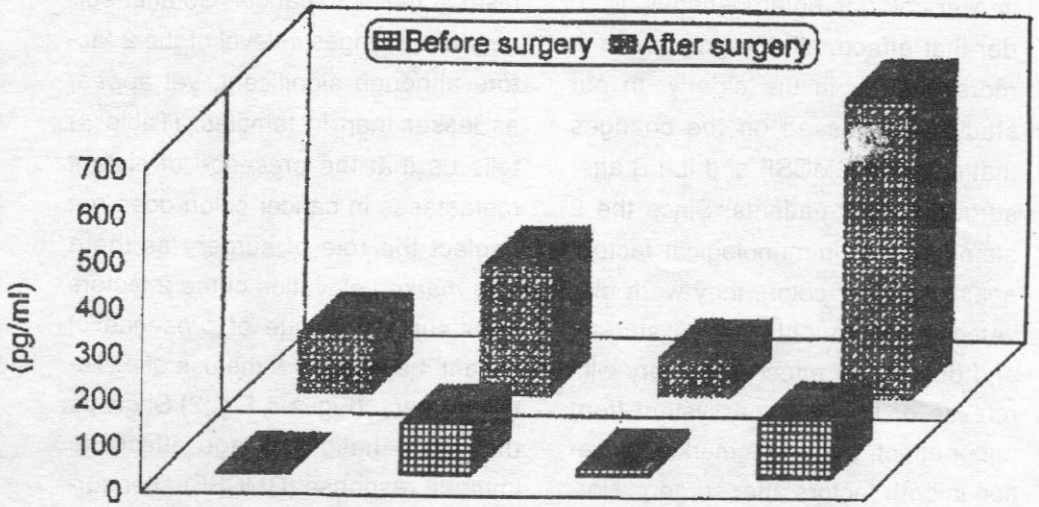
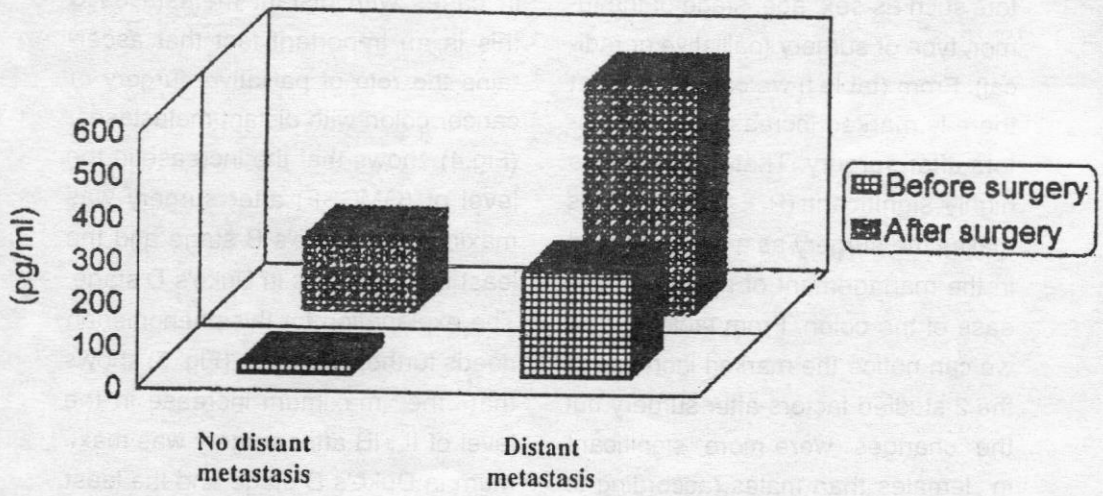


Figure (6) Interleukin -1-B before and after surgery in cases with and without distant metastasis



DISCUSSION

Schwartz (1986) described that cancer colon is heterogeneous disorder that affects all age groups but is more serious in the elderly. In our study we stressed on the changes that occur in GMCSF and IL-I B after surgery in our patients. Since the 2 elements are immunological factors against cancer colon, they were elevated in all patients before surgery and due to the effect of surgery with release of the immune system from tumor effect, there was marked elevation in both factors after surgery. Normal level GMCSF (0-2 pgm/ml). Normal level of IL-I B (0-5 pgm/ml) (MEDGENIX). The variation in the level of increase of GMCSF & IL-I B was variable depending on many factors such as sex, age, stage of the tumor, type of surgery (palliative or radical). From (table I) we can decide that there is marked increase of both factors after surgery. That increase was highly significant ($P < 0.0001$). This makes the surgery as a gold standard in the management of malignant disease of the colon. From table (2 & 3) we can notice the marked increase in the 2 studied factors after surgery but the changes were more significant in females than males (according to P value). The explanation of this is due to better immune response to tumors & better host defense mechanism in males to cancer. So after surgery the changes in level of the 2 factors although significant, yet appear as lesser than in females. (Table 4) tells us that the presence of distant metastases in cancer colon does not neglect the role of surgery as there was marked elevation of the 2 factors after surgery inspite of presence of distant metastases & the use of palliative surgery. (Fig. 1 & Fig. 2) Show us that the variations of age affect the immune response (GMCSF) after surgery. The more advanced the age of the patients, the better the immune response before and after surgery. (Fig.3) Shows us that the better immune response (GMCSF) is obtained in cases with distant metastases & this is an important fact that ascertains the role of palliative surgery of cancer colon with distant metastases. (Fig.4) shows that the increase in the level of (GMCSF) after surgery was maximum in Duke's B stage and the least increase was in Duke's D stage. The explanation for this phenomenon needs further research. (Fig. 5) shows that the maximum increase in the level of IL. IB after surgery was maximum in Duke's D stage and the least increase was in Duke s C stage. Also

the explanation of this phenomenon needs further research. (Fig.6) shows us the increase in the level of IL. IB after surgery is more in cases with distant metastasis and this means again that surgery remains the gold standard in management of cancer colon and even in advanced cases we can do the possible palliative surgery.

REFERENCES

(1) **Goding J .w . (1996) :** Monoclonal Antibodies: principles and practice. Academic press, London.

(2) **Lennex E.S . (Ed .) (1994) :** Clinical applications of monoclonal antibodies. Brit. Med. Bull. Churchill livingstone, UK.

(3) **Mayforth R.D & Quintans J. (1990) :** Designer and catalytic antibodies. New Eng. J. med. 323,173.

(4) **Mitchison NA. & O Malley C. (1987) :** Three cell type clusters of T cells with antigen presenting cell best explain the epitope linkage and non cognate requirements of the in vivo cytolytic response Euro. J. Immunol., 17. 1579.

(5) **Herberman R.B., Reynolds C.W & Ortaldo J. (1996) :** Mechanisms of cyto toxicity by NK cells. Ann. Rev. Immunology, 4,651.

(6) **Law S.K.A. & Reid K.B.M. (1998) :** Complement. Infocusseries. D.K. Male (ed.) IRL press, oxford.

(7) **Rother k. & Till G.O. (eds.) (1989) :** The complement system. Springer. verlage, Berlin.

(8) **Herbert W.J., Wikinson . P.C. & stott. D. I. (ed) (1995) :** Dictionary of Immunology, 3 rd ed. Black well scientific publications, oxford.

(9) **Male K., champion B. & Cooke A. (1997) :** Advancedmmunology. Gower Medical publishing, London.

(10) **Playfair J.H.L. (1992) :** Immunology at a glance, 4 th, ed. Blackwell scientific publications, oxford.

(11) Fiott I.M., Brosto FF J. & Male D.K (1998) : Immunology 2 nd ed. Gower Medical publishing, London.

(12) States D. P., Stab J. D . & Wells J.V. (1996) : basic and clinical immunology 6 th, ed. Lang Medical publications, California.

(13) Getzoff E.D., Tainer, J. A . Lemer R.A. & Geysen H.M. (1998) : 1 th. The chemistry and mechanism of antibody binding to protein antigens. Adv. Immunol, 43,1.

(14) Townsend A.R.M. & Bodmer B. (1998) : Antigen recog nition by class I. Restricted. T. lymphocytes. Ann. Rev. Immunology, 7, 601.

(15) Schwartz. T. B. (1986) : Metastasis from cancer colon. Lancet 733.