Perioperative levels of Interleukin-1β and Interleukin-6 in women with breast cancer

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Summary

Objective: Proinflammatory cytokines, such as Interleukin-1β and Interleukin-6, are known to play an important biological role in trauma, sepsis and malignant disease. Surgery can modulate the immune system, especially in patients with malignant diseases, by influencing the serum levels of Interleukin-1β and Interleukin-6. It is known that cytokine levels depend on the grade of tissue injury. We have investigated the serum levels of Interleukin-1β and Interleukin-6 during the perioperative period in women with breast cancer undergoing simple mastectomy or segmental resection. The aim of the study was to analyse whether breast cancer surgery influences cytokine expression in the peripheral blood and whether the concentrations of the measured interleukines differ according to the surgical method.

Methods: Blood samples of 45 women with breast cancer (Stage I and II) undergoing simple mastectomy (n = 16) or segmental resection (n = 29) were collected at six different times: before (T1), during (T2), three hours after (T3), one day after (T4), three days (T5) and five days after (T6) surgery. The serum levels of Interleukin-1β and Interleukin-6 were measured by ELISA.

Results: In both groups Interleukin-1β serum concentrations ($p < 0.05$) increased significantly on the first day after surgery. On the third day after surgery the concentrations of Interleukin-1b decreased to the preoperative level. A significant difference in Interleukin-1β concentrations as a consequence of the surgical method was not detectable. Changes in the serum levels of Interleukin-6 within the measurement period were not observed.

Conclusions: Surgery in patients with breast cancer leads to increased Interleukin-1β serum levels on the first postoperative day. It has been shown that elevated Interleukin-1β and Interleukin-6 levels are correlated with a high rate of recurrence. Therefore, this may be of consequence to patients with malignant diseases.

The method of surgery for both types, however, had no influence on the peripheral cytokine expression. Therefore, a nearly equal influence on the immune system can be stated.

Key words: Interleukin-1β; Interleukin-6; Breast cancer.

Introduction

It is generally known that cancer in early stages can be cured by surgery, as long as the malignant disease is still a local one. Breast cancer, however, is often already a systemic disease at the time of detection. Radical surgery attempts, as performed in the past, showed no benefit in comparison to modified surgery [1]. Also, prospective studies of prostate cancer showed no difference between radical surgery and mere observation [2]. It seems to be evident that besides the obvious benefits of surgical treatment, such as tumor reduction, there appears to exist a negative influence on the immune system due to surgical therapy in malignant disease. Studies have shown, that surgery can cause intraoperative shedding of tumor cells into the peripheral blood [3]. In addition, long-time anesthesia and blood transfusion contribute to immune suppression, which is known to enhance cancer growth [4].

Another important factor represents tissue damage and the process of wound healing. A stimulating effect on the growth of metastases within the proximity of fresh wounds was observed and related to the highly cellular and proliferating connective tissue [5]. Especially cytokines and growth factors, induced by trauma, seem to be involved in this process [6].

Cytokines are peptides produced by cells of the immune system that act as mediators of both the immune response and the response of other tissues in the body to injury. Interleukin-1β (IL-1β) and Interleukin-6 (IL-6) are mainly expressed by macrophages and monocytes [7, 8]. They belong to so-called proinflammatory cytokines. Infectious diseases and tissue damage lead to an enhanced production of these interleukines. Cytokines are bioactive at very low concentrations of less than 10^{-11} and have beneficial properties in injury and the healing of wounds, such as stimulation of antimicrobial function, myelostimulation and mobilisation of substrates.

On the other hand, aberrant secretion of cytokines is thought to be responsible for enhanced tumor growth and immune suppression. Surgery with a high rate of tissue damage initially causes local expression of IL-1β and IL-6 which afterwards spreads into the peripheral blood. We measured the perioperative levels of IL-1β and IL-6 in the peripheral blood in patients with breast cancer to investigate the effects of surgery on the immune system and compared these within the operative method.

Patients and Methods

Blood samples of 45 women treated for breast cancer at the Department of Gynaecology and Obstetrics, University Clinic Schleswig-Holstein, Campus Kiel (Germany) were collected

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six times during the perioperative period. All patients had tumors up to 5 cm in diameter without metastasis of lymph nodes or organs and were thus classified for breast cancer Stage I or II. The ages ranged between 30 and 86 years. Twenty-nine of these women were treated by lumpectomy. Lumpectomy or segmental resection means tumor excision, provided that the margins of the resected specimens are free of tumor. Sixteen patients underwent simple mastectomy. This means, that the breast tissue had been totally removed up to the fascia pectoralis. Both types of surgery were combined with axillary lymph node dissection.

The blood samples were obtained from peripheral veins at the following times: A preoperative blood sample was taken after induction of anesthesia. Thereafter, patients were sampled after incision at two and five hours and one, three and five days.

The serum samples were centrifuged, immediately frozen and stored at -81°C until assay. IL-1β and IL-6 were measured using commercially available "sandwich" enzyme-linked immunosorbent assays from BIOSOURCE INTERNATIONAL. The minimum sensitivity of the assays was as follows: IL-1β less than 0.083 pg/ml and IL-6 less than 2 pg/ml.

Statistical data are expressed as medians and interquartile ranges in boxplots. Time dependent changes within each group were analysed with the Friedman test. The differences between the two groups were tested with the Mann-Whitney-Wilcoxon U test; p values less than 0.05 were considered significant.

Results

Due to complications during surgery, technical faults or blood transfusions, blood samples of only 27 women could be sampled completely. Group 1 included 16 patients who underwent segmental resection, group 2 included 11 patients who underwent simple mastectomy.

In both groups a significant increase of IL-1β on the first postoperative day in comparison to the preoperative baseline occurred. On day 3 IL-1β values were back to the initial levels (see Figures 1 and 2). Between both groups no significant difference in IL-1β occurred. We observed no significant changes in IL-6 levels during and after surgery in either group and there were no significant differences between the groups (data not shown).

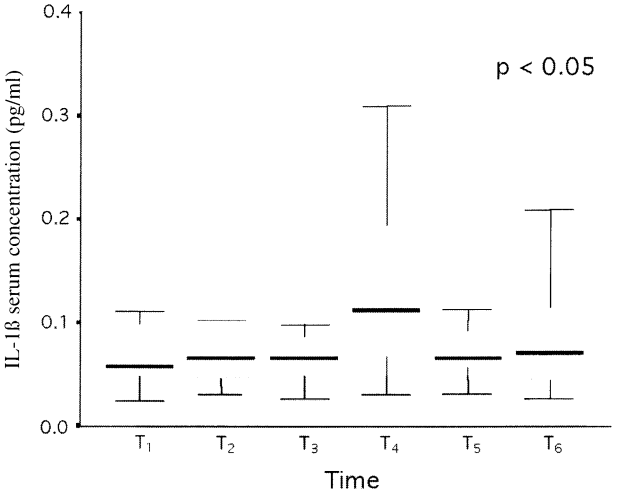

Figure 1. — IL-β serum levels of 16 women who underwent segmental resection for breast cancer.

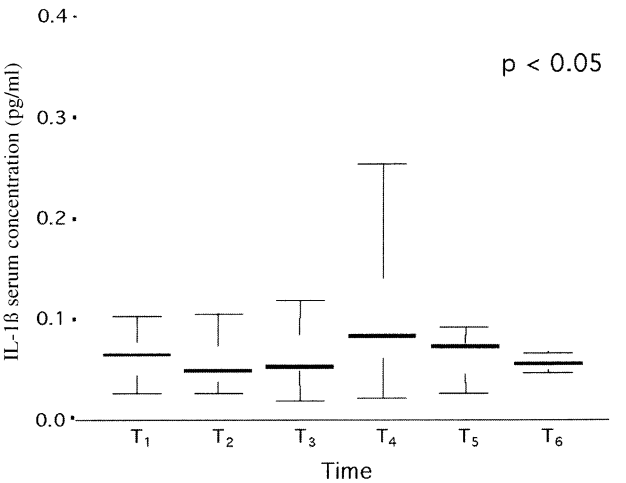

Figure 2. — IL-β serum levels of 11 women who underwent simple mastectomy for breast cancer.

Discussion

Former studies have shown that various surgical procedures influence IL-1β and IL-6 levels in the peripheral blood. In most of the observed surgical procedures a significant increase in serum levels occurred on the first day following surgery. In cases of uncomplicated surgery the increased levels lasted for one or two days. The systemic cytokine response correlated positively with the severity of the surgical measures. Complications such as sepsis caused higher and prolonged elevated interleukine levels [9]. Patients with malignant diseases showed significantly higher cytokine serum levels than patients with benign diseases [10].

The proinflammatory cytokines IL-1β and IL-6 have widespread and partly adverse effects on the immune system. They are bioactive at low concentrations (less than 10^{-11}mol/l) and have important autocrine, paracrine and endocrine functions. IL-1β is synthesised predominantly by tissue macrophages and blood monocytes. IL-6 is secreted by monocytes and fibroblasts. Cell death lysis and the microenvironment of wounds lead to an exaggerated production of these cytokines. They initiate beneficial properties such as stimulating immune defence and healing of wounds, whereas, on the other hand, adverse effects are described in cases of septic shock, chronic and malignant diseases [11].

Surgical procedures in patients with malignant diseases resulted in a stronger postoperative increase of proinflammatory cytokines than surgery in patients with benign diseases. It has been shown that this phenomenon is correlated with depressed cellular immune response and higher rate of postoperative recurrence. These cytokines are thought to have a growth promoting effect on tumor cells [12]. IL-1β and IL-6 are highly likely to have an influence on tumor growth by activating Nuclear Factor-kappaB (NF-κB). IL-1β and IL-6 activate NF-κB,

which has been proven to be constitutive in a subset of breast cancers [13].

In addition, modulation of cell surface antigen expression of cancer cells by proinflammatory cytokines has been proven in cell culture [14]. In contrast to these findings, a merely discrete growth promoting effect of proinflammatory cytokines and an anti-cancer effect on tumor lines in vitro has been documented [15, 16].

We examined whether breast cancer surgery influences peripheral blood cytokines. Within our set up we observed IL-1β and IL-6 levels in two different surgical procedures, simple mastectomy and segmental resection. Similar to the studies mentioned above, we observed a short peak in IL-1β levels in both groups on the first day following surgery. In contrast to other findings, changes in IL-6 levels were not observed for either group in our study. This missing increase in IL-6 levels may have been due to low intraoperative trauma, the absence of postoperative infections and the low grade tumor levels that were treated.

The elevated IL-1β levels demonstrate an activation of the immune system that was similar in both groups. It is known that proinflammatory cytokine levels correlate with the grade of surgical trauma. Simple mastectomy causes larger wounds than lumpectomy and therefore higher cytokine levels could have been expected. On the other hand, lumpectomy requires mobilisation of residual breast tissue, to achieve a cosmetically well formed breast, again increasing the area of traumatised tissue. This, in turn, may have resulted in a similar increase in IL-1β levels for both groups. In addition, it is conceivable, that a larger pool of data may reveal discrete differences.

We conclude that breast cancer surgery in Stages I and II influences IL-1β serum levels significantly. The effects on the immune system, especially in cancer patients, is not yet fully understood. Detailed knowledge of cytokines and their interactions with the immune system and tumor cells offer potential therapy. Conceivably, IL-1β-receptor antagonists could be used locally or systemically to block negative regulatory mechanisms. Further studies are required to investigate the interactions of proinflammatory cytokines and the immune system in patients with cancer.

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