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Levels of Circulating Intercellular Adhesion Molecule-1 in Patients with Metastatic Cancer of the Prostate and Benign Prostatic Hyperplasia

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Current reports suggest a role for intercellular adhesion molecule-1 (ICAM-1) in the progression of malignancy. The availability of a new antibody makes it possible to measure circulating ICAM-1 (cICAM-1) in human body fluids including serum; this might help in monitoring tumour burden and in providing additional prognostic information. In this study, serum levels of cICAM-1 were measured by an ELISA assay in patients with benign prostatic hyperplasia (BPH; $n=20$) and metastatic cancer of the prostate (CaP; $n=25$). Serum ICAM-1 concentrations were also measured in a group of healthy men ($n=8$). The mean ± S.E.M. cICAM-1 level for BPH was 339.52 ± 15.30 ng/ml compared with 263.55 ± 18.54 ng/ml for CaP. Even though the difference between the two groups was significant ($P<0.005$), there was a marked overlap between the individual values in both groups, thus minimising the prognostic value of these measurements in prostate cancer. Endocrine therapy had no notable effect on the serum levels of cICAM-1. The mean ± S.E.M. cICAM-1 concentrations in serum from a younger group of healthy volunteers was 204.1 ± 10.38 ng/ml, and this value was significantly lower than that measured in serum from either BPH or CaP. We also undertook some immunohistochemical studies to examine the distribution of ICAM-1 in prostate tissue. We observed focal epithelial cell membrane staining which was exceedingly patchy in both the BPH and cancer specimens. On the basis of these studies, we suggest that cICAM-1 levels do not provide additional information on patients with metastatic CaP.

Key words: ICAM-1, BPH, prostate cancer
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INTRODUCTION

RELEASE OF membrane surface antigens appears to be a common feature of malignant cells [1]. These antigens have been detected in culture media of active growing cell lines *in vitro* [2] and in body fluids of humans and animals with various malignant tumours *in vivo* [3]. In cancer patients, the levels of these antigens seem to correlate with the clinical stage of the tumour [4]. One of these membrane surface antigens is the intercellular adhesion molecule-1 (ICAM-1) which is an early marker of immune activation and response. Immunohistological staining of tissues suggested that ICAM-1 may have important implications for the investigation and monitoring of various inflammatory, neoplastic and immune disorders [5]. The availability of a new antibody, which recognises the binding site on the ICAM-1 antigen to the lymphocyte function associated antigen (LFA-1), makes it possible to detect soluble ICAM-1 antigen. Evidence from the literature suggests that serum ICAM-1 levels may be a useful marker to monitor tumour burden in cancer patients, not being specific for a particular tumour [6]. Whether this marker is elevated in patients with benign prostatic hyperplasia (BPH) and malignant diseases of the prostate such as metastatic cancer of the prostate (CaP) is not yet known.

MATERIALS AND METHODS

Patients

Blood samples were obtained from 20 patients with histologically confirmed BPH between 48 and 78 years (age mean = 67). In addition, a consecutive series of 25 patients with untreated and advanced cancer of the prostate between 50 and 88 years (age mean = 74) were also entered in this study. Blood was taken from the CaP patients before and after a 12 month course of endocrine therapy. Blood was also drawn from a younger group (under 40) of eight healthy volunteers. Blood specimens were allowed to clot, and the sera were stored at −20°C until analysis.

Prostate tissue was obtained at the time of transurethral resections from 6 patients with BPH and 6 with prostate cancer. Serial sections of the prostate specimens were cut and alternative sections were stained by haematoxylin and eosin to confirm the histopathology. The remainder of the sections were used for immunohistochemical staining.

ICAM-1 ELISA assay

Levels of cICAM-1 were measured employing a commercially available ELISA-Kit (British Biotechnology Products, Oxford, U.K.). This assay utilises an immunoenzymatic (streptavidin/Biotin) technique for quantification of cICAM-1 in human serum, with photometric quantification of the coloured reaction product against standardised purified soluble form of recombinant ICAM-1 allowing cICAM-1 levels to be calculated. The sensitivity limit of the assay was 2.5 ng/ml.

Immunohistochemistry

Immunohistochemistry was performed on frozen sections employing monoclonal antibodies to cICAM-1 (Genzyme Corporation, West Malling, Kent, U.K.) at a dilution of 1/600 (v/v), and following the procedures routinely used in this laboratory [7]. For each staining experiment, a "negative" control section was included in which the primary antibody was omitted. A "positive" control employing lymphatic tissue was also used. The staining of the specimens was analysed by an independent pathologist.

Statistical analysis

Data were analysed using a non-paired, two-tailed student *t*-test to obtain statistical comparisons.

RESULTS

The concentrations of cICAM-1 in the blood specimens obtained from the younger healthy volunteers and patient groups are illustrated in Figure 1. The mean ± S.E.M. values for the BPH patients were 339.52 ± 15.3 ng/ml (range: 240–458.7 ng/ml). This was significantly higher than the mean concentrations measured for the CaP patients before (263.55 ± 18.54 ng/ml, range: 105.73–508.35 ng/ml; $P<0.005$) and after (266.84 ± 22.14 ng/ml, range: 85.67–608.5 ng/ml; $P<0.05$) treatment even though there was a marked overlap between the different groups. Our study also demonstrated that endocrine therapy had little effect on the serum concentrations of cICAM-1 in CaP patients since there was no difference in their levels before and after treatment. Interestingly, the mean cICAM-1 concentrations in blood specimens obtained from healthy volunteers (204.1 ± 10.38 ng/ml, range 137–226.7 ng/ml) were significantly lower than the concentrations measured in both the BPH ($P<0.001$) and CaP ($P<0.01$) patient groups.

Immunohistochemically, the bulk of the staining occurred along the epithelial cell membranes, but this was very patchy and exceedingly weak (results not shown). Additionally, there was no change in the intensity of the staining as one progressed from BPH to cancer.

DISCUSSION

ICAM-1 was first described by Rothlein and associates [8] as an adhesion molecule that participates in lymphocyte function-association antigen (LFA-1) adherence in leucocytes, and was subsequently identified as the counter-receptor for LFA-1 [9]. The LFA-1/ICAM-1 complex is important in a number of leucocyte adhesive activities, including the conjugate formation between cytolytic T-cells and their target [10] and natural killer (NK) cell-mediated cytolysis [11]. It is generally recognised that

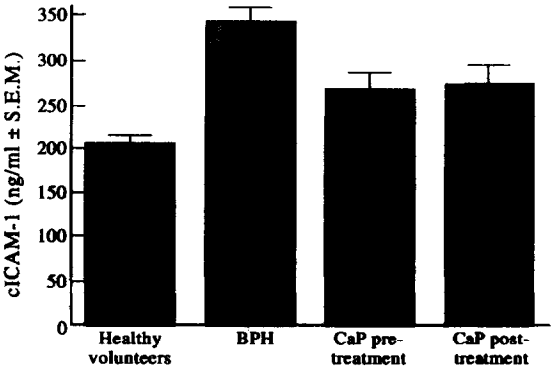

Figure 1. Measurement of soluble ICAM-1 concentrations in sera from normal healthy volunteers and patients with BPH and prostate cancer (CaP) before and following endocrine therapy. Values are expressed as mean ± S.E.M.

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NK cells are involved in the surveillance and regulation of tumour metastasis [12]. ICAM-1 has a wide cellular distribution and has been found in endothelial as well as epithelial cells [13].

Elevated levels of soluble ICAM-1 have been found in the blood of cancer patients [6,14–16]. Soluble ICAM-1 present in the sera of cancer patients is though to be, in part, derived from tumour tissue, as witnessed by the release of soluble ICAM-1 into the serum of nude mice by human melanoma cells [17]. However, interpretation of the clinical and biological significance of these data is complicated by the fact that mixed groups of patients, such as those investigated by Banks and colleagues [16], exhibited varying stages of disease, and some were on either active chemotherapy or on biological therapy. Therefore, the question of whether elevated cICAM-1 levels provide additional prognostic information in cancer patients still remains unanswered as no clear and universal pattern of cICAM-1 levels in human tumours has been found. Furthermore, ICAM-1 expression has been reported to be increased by cytokine therapy [17, 18], and raised cICAM-1 levels do not appear to be specific for neoplasms, but may be found in various inflammatory and immune disorders [5, 19–21].

In the current study, we found serum levels of cICAM-1 to be significantly elevated in patients with benign and malignant prostatic disease compared with a healthy younger group of volunteers. However, since the healthy group is of a significantly younger age, it is difficult to ascertain whether the increase in cICAM-1 concentrations in the patient groups is in any way related to the abnormal growth of the prostate or mainly reflects an age-dependent change. Additionally, cICAM-1 concentrations fail to reflect the malignant state of the cancer on an individual basis due to a considerable overlap in cICAM-1 concentrations in patients with BPH and CaP. Whilst we have noted some faint staining for the adhesion molecule in BPH and prostate cancer specimens, it is difficult to ascribe the elevation in serum cICAM-1 concentrations to the prostate cancer proper or indeed to the surrounding tissue even though BPH is associated with lymphocytic infiltration [22]. Furthermore, cICAM-1 blood profiles in prostate patients do not appear to be influenced by endocrine manipulation since no major changes were observed following a 12 month period of treatment. However, in an earlier study of a group of 6 patients with metastatic CaP, receiving interferon alpha as a second line of treatment, Pummer and associates [23] showed a decrease in cICAM-1 concentrations in two patients who responded subjectively to the treatment whereas the remaining four patients with no response demonstrated an increase in cICAM-1 levels. Nonetheless, because both BPH and CaP frequently co-exist in the prostate, the measurement of cICAM-1 levels in patients with CaP is unlikely to provide any additional information which could be useful for the management of prostate cancer.

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