Abstract. Background: The clinical significance of the carboxyterminal telopeptide of type I (ICTP) as a predictor of prognosis is insufficiently known in ovarian cancer. Therefore, we compared the prognostic accuracy of ICTP and CA 125 and their combination with each other and with the aminoterminal propeptide of type III procollagen (PIIINP) and clinical prognosticators. Patients and Methods: Fifty-five epithelial ovarian cancer patients treated in Oulu University Hospital, Finland, from 1989-1995, were enrolled in this study. Cytoreduction was performed as completely as possible. The preoperative serum concentrations of the biochemical markers were determined using commercial kits. Results: In univariate analysis, each biochemical marker and all conventional prognostic indicators, with the exception of histological subtype, correlated significantly with survival. In multivariate Cox analysis on biochemical and clinical markers, ICTP alone and ICTP + CA 125, but not CA 125 alone, proved useful indicators of overall survival. The combination of ICTP and CA 125, however, added no essential information compared to that obtained with ICTP alone. Conclusion: The ICTP test seems to be a clinically useful predictor of the clinical behavior of ovarian cancer. An increased serum ICTP concentration may reflect the spreading and aggressiveness of invasively growing ovarian cancer.

CA 125 is a widely used biochemical tumor marker in ovarian cancer. Most patients with clinically recognizable ovarian cancer show elevated serum levels of CA 125. It may also be useful in monitoring the course of ovarian cancer (1, 2), but its clinical significance as a prognosticator in this disease is unclear (3, 4).

The serum concentration of trivalently cross-linked carboxyterminal telopeptide of type I collagen (ICTP), an indicator of degradation of mature (trivalent cross-links) type I collagen, reflects changes in the extracellular matrix of the ovarian tumor and the surrounding tissues during cytotoxic chemotherapy and follow-up (5,6). Our preliminary observations suggested that the ICTP test might be a clinically useful indicator of prognostic prospects in ovarian cancer (6). The aminoterminal propeptide of type III procollagen (PIIINP), an indicator of type III collagen metabolism, has also been a useful complement to CA 125 in monitoring the clinical behavior of ovarian cancer (7).

The present study, which is a continuation to our previous investigations on ICTP in ovarian cancer (5, 6, 8-11), was conducted to find out about the prognostic significance of the preoperative serum concentrations of ICTP and CA 125 in ovarian cancer patients followed-up for more than 7 years. These indicators were compared with each other and with PIIINP and several conventional clinical variables.

Patients and Methods

Fifty-five patients with epithelial ovarian cancer treated in Oulu University Hospital, Finland, during the period 1989-1995, were enrolled into this study. Serum samples for ICTP, CA 125 and PIIINP determinations were obtained preoperatively. The clinical records of the patients were reviewed for age, clinical stage, histological type and grade, presence of ascites, peritoneal cytology and size of residual tumor after staging and debulking laparotomy and therapy. The preliminary results with a short follow-up period and without testing the combination of ICTP and CA 125 have been published previously (6). The median age of the patients was 53 years (range 24-73). The Ethics Committee of the University of Oulu approved our study design.

All the tumors were of epithelial origin, the majority being serous (N=32), while the rest were mucinous (N=9), endometrioid (N=6), clear-cell (N=3) or undifferentiated (N=5) tumors. According to the classification of the International Federation of Gynecology and Obstetrics (FIGO), there were 21 stage I, 4 stage II, 24 stage III and 6 stage IV tumors. Twelve tumors were well-
(grade 1), 18 moderately- (grade 2) and 25 poorly- (grade 3) differentiated. The surgery was carried out through a vertical midline incision. Cytoreduction was performed as completely as possible and all palpable enlarged lymph nodes were removed. The operation was radical (no detectable residual disease) in 28 patients. The cytoreduction was optimal (defined as the reduction of all tumor lesions to less than 2.0 cm in diameter) in 10 and suboptimal in 17 patients. Twenty-seven patients had ascitic fluid at primary laparotomy (defined as 100 ml or more fluid in the peritoneal cavity). At least 4 courses of platinum-based (cisplatin 50 mg/m²) combination chemotherapy with cyclophosphamid (500 mg/m²) and epidoxorubicin (50 mg/m²) were given to 53 patients at four-week intervals. Two patients died within 2 months after the operation without any further therapy. None of the patients received radiotherapy. None of the patients had bone metastases at any point of follow-up.
The median follow-up time was 54 (range 1-168) months. At the end of the study, after a mean follow-up period of 7.5 years (median 125, range 90-168 months), 20 patients were alive (18 without any evidence of recurrent disease). Thirty-three had died of ovarian cancer after a median follow-up time of 27 (range 1-94) months, and two had died of concurrent diseases at follow-up times of 41 and 95 months. Follow-up data were available for all of the 55 patients.

The blood samples were centrifuged and the sera were immediately frozen and stored at -20°C until analyzed. The serum ICTP (12) and PIIINP (13) concentrations were determined by equilibrium radioimmunoassays for the human antigens, using commercial kits from Orion Diagnostica (FIN-90460 Oulunsalo, Finland). The upper limits of the reference interval for serum ICTP and PIIINP were 4.6 ìg/l and 4.2 ìg/l, respectively. The intra- and interassay coefficients of variation were less than 6% for all the assays at the concentrations encountered in this study. Serum CA 125 was assayed with kits from Centocor Europe (Tongeren, Belgium). Concentrations higher than 35 U/ml were considered elevated. All assays were performed in duplicate.

For statistical analyses, the patients with stage I and II as well as grade 1 and 2 tumors were combined and tested against those with stage III-IV and grade 3 carcinomas, respectively. For the same purpose, the patients were divided into two histopathological groups: those with serous versus non-serous neoplasms. For the biochemical indicators ICTP, PIIINP and CA 125, the cut-off values used to discriminate between survivors and non-survivors were similar to those reported in our previous paper (6) as follows: 5.7 ìg/l for ICTP, 170 U/ml for CA 125 and 3.9 ìg/l for PIIINP.

Survival was calculated as corrected survival from the date of laparotomy to the date of death or to the closing date of this study. Survival analyses were performed according to the Kaplan-Meier method (14). Univariate analyses were carried out using the log-rank test and multivariate analyses using the Cox model with backward variable elimination and the Wald statistic. The Mann-Whitney U-test was used for continuous variables because of the skewed distribution of the variables and the χ²-test was used for the contingency tables of discrete variables.

### Results

The median (range) ICTP, CA 125 and PIIINP concentrations of the patients who died of ovarian cancer during the follow-up (N=33) were higher than those of the patients who survived (N=20) as follows: ICTP 5.9 ìg/l (1.9-31) vs. 3.4 ìg/l (1.9-11.4) (p=0.030); CA 125 618 U/ml (9-14,800) vs. 109 U/ml (5-3650) (p=0.001) and PIIINP 4.4 ìg/l (1.0-21.5) vs. 2.8 ìg/l (1.0-9.8) (p=0.007).
The ICTP value was higher than 5.7 μg/l in 18 and the CA 125 value was higher than 170 U/ml in 28 of the 33 patients who died of the disease. The corresponding figure for ICTP (> 5.7 μg/l) and CA 125 (> 170 U/ml) combined was 18. Three of the 20 patients who were alive at the end of the follow-up had ICTP values higher than 5.7 μg/l and 7 had CA 125 values higher than 170 U/ml. The corresponding figure for ICTP and CA 125 combined was 2. The sensitivity, specificity and predictive values of positive and negative tests in correlation with the outcomes of the patients are presented in Table I. The combined use of ICTP and CA 125 did not add any essential information to that provided by ICTP alone (Table I).

In the univariate analysis, all biochemical variables were significant prognostic indicators of the patients’ clinical outcome (Table II, Figure 1a-c). The conventional clinical and histopathological prognosticators, apart from the histological subtype, also differentiated between the high-risk and low-risk patients (Table II).

The Cox multivariate stepwise method showed CA 125 and ICTP to be significant prognostic indicators of survival among the three biochemical markers evaluated here (Table III). In the analyses where ICTP alone or combined with CA 125 were associated with the conventional markers, ICTP alone and combined with CA 125 appeared to be significant prognosticators of overall survival together with age and clinical stage. When CA 125 was associated with the conventional indicators of prognosis, it lost its significance as an independent prognosticator of overall survival. In this analysis and in the analysis of conventional markers, residual tumor volume and age remained independent prognostic indicators of survival (Table III).

Discussion

We have previously found that the preoperative serum ICTP concentration gives potential independent information about the prognostic prospects of ovarian cancer patients (5, 6, 11). In the present study, we further tested the reliability of this hypothesis by lengthening the follow-up period up to 7 years or more. The value of the combination of ICTP and CA 125 was also evaluated.

This study provided additional evidence of the clinical value of ICTP as a prognostic indicator in epithelial ovarian cancer. With the present cut-off values, ICTP was better than CA 125 in specificity, while CA 125 was better than ICTP in sensitivity. A high ICTP concentration predicted a fatal outcome with 86% accuracy and a low CA 125 concentration predicted a good prognosis with 72% accuracy. ICTP and CA 125 thus seem to complement each other as indicators of prognosis. However, the combination of ICTP and CA 125 did not essentially improve the predictive value of ICTP as regards poor outcome. Previous studies on the predictive value of preoperative serum CA 125 have yielded controversial results. In advanced (15, 16) but not in early stage (17) disease, serum CA 125 has failed in this function. Serum ICTP and CA 125 originate from different sources; ICTP is a degradation product of the mature type I collagen molecule, and CA 125 a membrane product of the epithelial cells of the coelomic origin of the tumor and mesothelium (18, 19).

In clinical praxis, the information obtainable by serum tumor markers is used together with conventional clinicopathological indicators of prognosis. In a test including such variables, serum ICTP remained in the equation together with clinical stage and the patient’s age.

Table III. Independent and significant prognostic indicators of the survival of 55 ovarian cancer patients as evaluated by the Cox regression model. ICTP is compared with PIIINP and CA 125 (A) and ICTP and CA 125 alone and combined with different clinical and histopathological variables (B). The relative risk of death and its 95% confidence interval and p-value of additional information in the χ²-test are given for each covariate in the equation.

<table>
<thead>
<tr>
<th>Variables entereda</th>
<th>In equation</th>
<th>Exp(B)</th>
<th>p-value</th>
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<tbody>
<tr>
<td>A)</td>
<td></td>
<td></td>
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<tr>
<td>ICTP</td>
<td>CA 125</td>
<td>3.531 (1.283-9.717)</td>
<td>0.015</td>
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<td>CA 125</td>
<td>ICTP</td>
<td>2.201 (1.060-4.571)</td>
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<tr>
<td>ICTP</td>
<td>PIIINP</td>
<td>3.319 (1.658-6.646)</td>
<td>0.001</td>
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<tr>
<td>B)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ICTP</td>
<td>Stage</td>
<td>3.741 (1.532-9.136)</td>
<td>0.004</td>
</tr>
<tr>
<td>Stage</td>
<td>Age</td>
<td>1.044 (1.007-1.083)</td>
<td>0.019</td>
</tr>
<tr>
<td>Grade</td>
<td>ICTP</td>
<td>2.297 (1.111-4.751)</td>
<td>0.025</td>
</tr>
<tr>
<td>Residual tumor</td>
<td>Age (continuous)</td>
<td>3.474 (1.400-8.619)</td>
<td>0.007</td>
</tr>
<tr>
<td>ICTP + CA 125b</td>
<td>Stage</td>
<td>2.478 (1.177-5.217)</td>
<td>0.017</td>
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<tr>
<td>Stage</td>
<td>Age</td>
<td>1.044 (1.007-1.082)</td>
<td>0.020</td>
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<td>Residual tumor</td>
<td>Age (continuous)</td>
<td>4.227 (1.919-9.311)</td>
<td>&lt;0.001</td>
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<td>CA 125</td>
<td>Residual tumor</td>
<td>4.227 (1.919-9.311)</td>
<td>&lt;0.001</td>
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<td>Grade</td>
<td>Age</td>
<td>1.042 (1.006-1.080)</td>
<td>0.022</td>
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<tr>
<td>Residual tumor</td>
<td>Age (continuous)</td>
<td>4.227 (1.919-9.311)</td>
<td>&lt;0.001</td>
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<td>Stage</td>
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<td>0.022</td>
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</tbody>
</table>

aCut-off values for ICTP, PIIINP and CA 125 ≤ 5.7 μg/l, ≤ 3.9 μg/l and ≤ 170 U/ml, respectively
bICTP ≤ 5.7 μg/l and CA 125 ≤ 170 U/ml vs. others
In corresponding multivariate analysis, serum CA 125 dropped out of the equation and thus lost its independent significance as a predictor of prognosis. The strong dependence of serum CA 125 on residual tumor volume (6) may explain this finding, because tumor volume remained as the most powerful prognosticator in this combination. On the other hand, the ICTP concentration reflects the invasive aggressiveness of the tumor rather than the tumor load (6).

This biological property of ICTP is important and predominant, as in multivariate Cox analysis, the combination of ICTP + CA 125 remained in the equation together with clinical stage and age, as also did ICTP alone.

We have previously found the 9-month ICTP test to be an even more precise prognosticator of overall survival than the preoperative ICTP test in patients with epithelial ovarian cancer (11). Irrespective of whether the tumor was at an early or advanced stage at the time of diagnosis, the ICTP test made preoperatively or at 9 months of follow-up was able to differentiate between the survivors and non-survivors with relatively good accuracy. A short half-life of serum CA 125 during postoperative cytotoxic chemotherapy has been found to be a sign of good prognosis in many (20-23), but not all studies (24). The transient decline of the serum CA 125 concentrations in some patients with a poor prognosis diminishes the clinical predictive value of the CA 125 test during the first half of the postoperative year. From 9 months of follow-up onwards, serum CA 125 is quite comparable to ICTP as a predictor of prognosis in this disease.

Wide dissemination of peritoneal implants and extensive malignancy-induced degradative damage of soft tissues in the neighbouring organs (25) are signs of biological aggressiveness of the tumor and a poor prognosis. Irregularly arranged and destructured ICTP-positive fibres are characteristic of poorly-differentiated ovarian cancer tissue (26,27). Large amounts of different collagen metabolites in the ascitic fluid of ovarian cancer patients are also typical of advanced ovarian cancer (28). These results demonstrate that the metabolism of fibrillar type I and type III collagens is significantly enhanced and affected by serious disorders.

The present ICTP assay measures fragments released from trivally cross-linked, mature type I collagen molecules (29). Because the content of mature, trivalently cross-linked, mature type I collagen is low and that of insufficiently matured fibers high in malignant ovarian tumor tissue in contrast to benign soft tissues (27), the excess serum ICTP is unlikely to be derived from the tumor tissue. In the serum of patients with progressive ovarian cancer, it must originate from the soft tissues surrounding the malignant lesions, where the invasive processes cause tissue damage most likely via the matrix metalloproteinase pathway (30,31). The ICTP test actually measures malignancy-specific biological phenomena that are not identifiable with other methods.

In ovarian cancer tissues, irregularly organized PIIINP-positive fibers have been seen close to the cancer cells as well as further away in the stroma in immunohistochemical studies (32). Although the action of cancer upon the stroma is predominantly degradative (33), some synthesis of matrix components, including fibrillar collagens, also occurs (10, 34). Enhanced metabolism of type III collagen is also evidenced by the increased serum concentrations of PIIINP, which correlate with the clinical behavior of the disease (7).

In agreement with our previous observation (6), the preoperative PIIINP level was a prognostic indicator in ovarian cancer in univariate analysis, but its significance was lost in multivariate analyses involving other biochemical prognostic indicators.

In conclusion, our study suggests that the determination of preoperative serum ICTP level is clinically useful, because it gives specific information about the aggressiveness of epithelial ovarian carcinomas and the consequent prospects of survival independently of conventional tumor markers. The combined use of ICTP and CA 125 as a predictor of prognosis provided no further information compared to that available from ICTP alone.

References


