The Diagnostic and Prognostic Value of Tumor Markers (CEA, SCC, CYFRA 21-1, TPS) in Head and Neck Cancer Patients

VIVIAN BARAK1, AMICHAY MEIROVITZ1, VERA LEIBOVICI2, JACOB RACHMUT3, TAMAR PERETZ1, RON ELIASHAR4 and MENACHEM GROSS4

Departments of 1Oncology, 2Dermatology, 3Surgery and 4Otolaryngology-Head and Neck Surgery, Hadassah–Hebrew University Medical Center, Jerusalem, Israel

Abstract. Background/Aim: Establishing prognostic factors is very important in the management of cancer patients. Our aim was to evaluate the clinical significance of a panel of tumor markers, including CEA (Carcino Embryonic Antigen), SCC (Squamous Cell Carcinoma Antigen), TPS (Tissue Polypeptide Specific Antigen) and CYFRA 21-1 in head and neck cancer patients, for assessing treatment response and prognosis of patients. Patients and Methods: We evaluated 312 blood samples from 143 head and neck cancer patients, from several sub-groups: 82 Larynx Carcinoma pre- and 38 post-therapy, 46 Oral Cavity pre and 29 post-therapy, 12 nasopharynx, 16 parotid and other salivary gland patients. Blood tumor markers levels were evaluated by conventional ELISA assays. Correlations of marker levels to stage of disease, lymph node involvement and therapy, were performed. Results: Serum levels of all four tumor markers were higher before therapy and decreased thereafter in all patients. The decrease in TPS level following therapy was significant (p=0.03). Significantly higher levels of TPS and similarly higher levels of the other tumor markers were demonstrated in advanced disease (stages III and IV) patients, as opposed to early disease (stages I and II) patients (p=0.012). Node positive patients had significantly higher TPS levels as compared to node negative (p=0.02). The same trend was shown by the other markers as well, but did not reach statistical significance. TPS was best correlated to survival of patients; those having low levels had the best clinical outcome and longer survival. Conclusion: CEA, SCC, TPS and CYFRA 21-1 can all serve as useful tumor markers in HNC patients. They assessed response to therapy and were prognostic for recurrence. TPS proved to be the most sensitive predictor of advanced disease and poor prognosis.

Over 40,000 new cases of head and neck cancer (HNC) are diagnosed every year in the USA, and about 12,000 patients will die from the disease, yearly. About 90% of the HNC are of the squamous cell carcinoma (SCC) type. The main etiologies of HNC are smoking and alcohol consumption. In many cases, second malignancies were described in treated HNC patients (1, 2). Prognosis of the patients is according to the stage of the disease, lymph nodes involvement and response to therapy. The main problem remained that approximately two-thirds of the HNC patients present with loco-regionally advanced (stage III and IV) disease. This requires a multimodality therapy, including surgery, radiation, and/or chemotherapy. During recent years the combination of Chemo-Radio therapy (CRT) as part of organ preservation protocols, is the more common. However, despite recent advances in treatments for HNC, survival of patients did not improve and accounts for about 50% after 5 years (3, 4).

Tumor markers are important tools to mainly predict and assess response to therapy in those patients. Early detection of the disease and of recurrence by tumor markers, should improve the clinical outcome. The most used tumor marker over the years was Carcino Embryonic Antigen (CEA); however, additional markers were studied in various configurations over the years (5). We have previously demonstrated that cytokeratin tumor markers such as Tissue Polypeptide Antigen (TPA), Tissue Polypeptide Specific Antigen (TPS) and cytokeratin-19 fragments (CYFRA 21-1) are very sensitive markers and account for tumor activity and response to therapy, in various types of cancer (6, 7). Other studies also pointed out the additive information by using a

Correspondence to: Prof. Vivian Barak, Head, Immunology Laboratory for Tumor Diagnosis, Department of Oncology, Hadassah-Hebrew University Medical Center, Jerusalem, Israel. Tel: +972 26776764, Fax: +972 26435308, e-mail: barakvivi@hadassah.org.il, barakvivi@gmail.com

Key Words: Head and neck cancer, tumor markers, clinical utility.

0250-7005/2015 $2.00+.40
panel of markers such as CEA combined with a cytokeratin marker in various carcinomas, such as breast, ovary, bladder, and prostate cancers (8-10). Based on our and other former studies, we evaluated in the present study a panel of tumor markers including cytokeratins, as to their clinical value in HNC patients.

Patients and Methods

The aims of the present study were to investigate the clinical role and significance of a panel of tumor markers: CEA, SCC, CYFRA 21-1 and TPS in head and neck cancer patients, following various treatments.

Patients and methods. Serum (received after patients' blood centrifugation) levels of the tumor markers CEA, SCC, CYFRA 21-1, TPS were evaluated by conventional ELISA assays: TPS for the quantitative measurement of the M3 epitope of soluble fragments of human cytokeratin 18 was purchased from IMMULITE SIEMENS, UK. For the other markers, CEA, SCC and CYFRA 21-1, ELISA kits were purchased from BRAHMS, Hennigsdorf 25, Germany. The following groups of 143 HNC patients (from them we received 312 blood samples) were evaluated: larynx n=82, nasopharynx n=12, oral cavity n=33, parotid and other salivary gland malignancies n=16. Of these patients, 120 were evaluated both prior to and after therapy (surgery, radiation, chemotherapy or combined modalities) and levels of the samples, as indicated in figures, were correlated to clinical status, stage, lymph nodes and response to treatment. Overall survival was estimated according to initial levels of the tumor markers.

Statistical analysis. Statistical analyses were performed using SPSS software and included non parametric tests as Wilcoxon matched-pairs signed rank test, Mann-Whitney U-test and survival analysis (Kaplan Meyer method and the log rank test). A p-value less than 0.05 was considered statistically significant.

Results

All marker levels were higher in advanced disease (stages III and IV) compared to early disease (stages I and II), while TPS was significant, \( p=0.012 \), as shown in Figure 1. This finding shows a good correlation between tumor mass and levels of serum markers.

Comparing serum tumor marker levels before and following therapy, demonstrated higher levels of the markers CEA, SCC and CYFRA 21-1, in all patients, as opposed to decreased levels post therapy (TPS, \( p=0.03 \)), as shown in Figure 1.
Figure 2. This finding shows an association of marker levels with tumor mass, nodal status, activity of the disease, correlating to response to therapy.

Patients with node positive disease had significantly higher TPS levels, compared to those with node negative disease ($p=0.02$), as shown in Figure 3. The other three markers showed a similar trend, but did not reach statistical significance. When correlated to irradiation therapy, only SCC levels were significantly higher prior therapy than following irradiation therapy ($p=0.05$), as shown in Figure 4. Rest of the markers showed a similar trend, however, with no statistical significance.

We also compared marker levels in the subgroups of HNC patients. TPS levels were significantly higher in the group of Parotid and other salivary gland patients ($317+120$) than in Larynx ($76+10$), Nasopharynx ($66+13$), and Oral cavity ($86+18$) patients, which were in the same range ($p=0.05$). Similarly, CYFRA 21-1 levels were also significantly higher in the Parotid and other salivary gland patients ($15.211+9.93$) than in Larynx ($1.13+0.09$), Nasopharynx ($1.95+0.44$), Oral cavity ($1.49+0.23$) patients ($p=0.02$). The other two markers did not differ significantly between those groups of patients.

A kinetic evaluation of an individual patient followed up every 2 months with TPS, is shown in Figure 5. The significant increases in TPS levels reflected earlier disease recurrence, shown later by a computed tomographic scan of this patient (see raw on the graph).

Overall survival correlated best to TPS levels: of patients with high TPS levels, 57% were alive after 2 years and 46%
after 5 years, while of patients with low TPS levels, 90% of them were alive after 2 years and 75% after 5y. The most sensitive marker in the panel of tumor markers we tested, was the TPS.

**Discussion**

To achieve personalized treatment for cancer, markers for determining prognosis, predicting response to therapy and predicting severe toxicity related to treatment, are urgently required and expected to increase survival, due to their earlier treatment (5).

CEA has been evaluated for use as a prognostic parameter in patient outcomes. However, this marker is related mostly to tumor mass and does not reflect proliferative activity. In contrast, cytokeratin tumor markers reflect proliferative activity, which is one of the most important phenotypic characteristics of tumor aggressiveness and may thus be more beneficial as a prognostic indicator.

Cytokeratins are well recognized biomarkers in various types of cancer, as we and others have demonstrated (6, 7). In Lung cancer, tumor markers including cytokeratins are recommended for use in differential diagnosis, prognosis and therapy monitoring, particularly of non-small cell lung cancer (NSCLC), as we and other colleagues have shown previously (8-17). Beyond lung cancer, there are an increasing number of studies showing a remarkable sensitivity of cytokeratin markers, such as TPA and TPS, mostly in breast, as we have previously demonstrated (6, 19), but also in bladder, ovarian, and colorectal cancer (14-16). This might be explained by a higher general cytokeratin release during cell death, that might more frequently occur in tumor disease due to a high cellular turnover in some situations (17).

In our present study, only TPS levels were statistically significant higher in advanced stage (stage III and IV) patients than in early stage (stage I and II) patients. TPS was also significantly higher in node positive patients, indicating a significant correlation between the level of markers and tumor burden or proliferative activity of the patients. We have also previously shown in Breast Cancer the significance of using TPS in addition to tumor mass markers as CEA and CA15-3, for the early detection of recurrence (6, 19), suggesting earlier treatment and longer survival, similar to the results in HNC patients of this study.

In the present study we compared the tumor marker levels in subgroups of HNC patients. TPS levels were significantly higher in patients with Parotid and other salivary gland malignancies compared to other sites such as larynx, nasopharynx and oral cavity. To our knowledge, this finding has not been described in the literature.

During recent years there have been many efforts to identify new markers for HNC, using different methods, from HNC cell lines to proteomics (20-26). Thus, PLAU and IGFBp7 were found significantly increased in HNC patients relative to controls (22). In addition, CD147 was suggested to be a novel marker for the diagnosis of Oral Cancer. In a recent review, summarizing prognostic markers for HNC from the literature, the cytokine IL-6 was identified as the most significant predictor of patient's outcome (24). Additional markers for HNC were suggested as well, such as MMPs1,2,9 (26). Although these results are promising (22-26), a significant validation in a high number of patients followed up adequately for a longer period is required, in order to introduce them into the clinics as routine.

In conclusion, we demonstrated in the present study that SCC (in Squamous type Cancers), CEA, TPS and CYFRA 21-1, are sensitive and useful tumor markers in HNC patients. Higher levels of the markers were associated with active disease, higher stages and node positive patients. Significant associations were demonstrated between response to therapy, such as first surgery and radiation and decreases in marker levels. Decreases in all 4 marker levels

---

Figure 4. SCC levels (mean±SE) according to radiation therapy in HCN patients.

Figure 5. Kinetic evaluation of TPS levels in a HNC patient.
demonstrated positive therapy effects and a longer survival. Increases in TPS were the most sensitive predictors of advanced disease and poor prognosis, best correlated to overall survival. Those results are in good concordance with our previous data on TPS and its significance in Breast cancer patients (27-30). We therefore suggest, introducing into routine a panel of tumor markers including cytokeratins, for the useful follow up of HNC patients, assessment of their response to therapy and early detection of recurrence for an improved survival.

Acknowledgements

The Authors appreciate and thank Mrs. Kalichman I and Dr. Nisman B for their technical assistance. They also thank Mr. Aharonovich M for the statistical workup.

References


