Prognostic Applications of the Epidermal Growth Factor Receptor and Its Ligand, Transforming Growth Factor-α

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Because growth factors and their receptors have a central role in regulating developmental and neoplastic processes, the expression and action of various growth factor receptors and their ligands in human neoplasia have been extensively investigated. During the past two decades, several investigators have described the amplification of the epidermal growth factor receptor (EGFR) at the genotypic level and overexpression of the EGFR protein at the phenotypic level in many tumors (1–16). However, great heterogeneity in the incidence of overexpression, in the intensity and localization of receptor deposition, and of prognostic significance has been observed both within and between various tumor types. EGFR protein overexpression has been reported to occur and to be of prognostic value for predicting either shorter disease-free survival or shorter overall survival in endometrial carcinoma [49% incidence (1)], esophageal carcinoma [43% incidence (2)], bladder carcinoma [35% incidence (3)], and prostatic carcinoma (4). In several other tumor types where multiple studies have been reported, the results have been mixed. Overexpression of EGFR has been frequently reported in non-small-cell lung carcinoma (5–8); however, the incidence of EGFR overexpression ranges from 12% (21 of 169 cases) to 37.5% (12 of 32 cases), with no demonstrated association between disease-free or overall survival. Similar heterogeneity has been reported in ovarian cancer with observed incidences of 19% (9) to 50% (10); both studies claimed an association of EGFR with either advanced stage (10) or poorer prognosis (9). Verbeek et al. (11) reported overexpression of EGFR in 20%–30% of breast cancer cases investigated by immunostaining and claimed an association with poorer prognosis; however, by use of either conventional (12) or quantitative (13) radioimmunohistochemistry, two studies have suggested that the expression level of EGFR is significantly lower in malignant breast tissue than in benign and normal breast epithelium. Although Iwase et al. (14) reported a 38.8% incidence of EGFR positivity among tumor tissue samples in 80 breast carcinoma cases, EGFR expression was inferior to lymph node involvement or tumor size as a prognostic indicator. Similarly, conflicting reports of quantitative analysis of lower than normal tissue levels of EGFR in cervical carcinoma (15) versus increased levels in patients with invasive cervical carcinoma (16) without prognostic significance have been reported. As summarized by Robertson et al. (13), a great deal of this heterogeneity may be attributable to a general lack of standardization between laboratories for EGFR assay procedures. Quantitative methodologies, whether by labeled ligand or by messenger RNA (mRNA) or DNA analyses, are performed on preparations derived from tumor biopsy specimens containing malignant cells and non-tumor-cell populations and thus do not necessarily represent the neoplastic cell population; immunohistochemical analysis, while subjectively capable of discriminating between normal and neoplastic elements, is not readily quantifiable. Conflicting series observations will need to be clarified by carefully controlled studies comparing quantitative and qualitative determinations of frequency and identity of EGFR-expressing cells and examination of sufficiently large patient cohorts to determine potential prognostic significance.

In this issue of the Journal, Rubin Grandis et al. (17) present the fifth paper in a series of studies of EGFR protein expression in head and neck carcinomas and report on the potential of these marker levels for predicting disease-free and overall survival. Previous studies of EGFR expression in head and neck cancer (18–20) have generally concurred that EGFR overexpression was associated with shorter relapse-free intervals and overall survival; a single study in 68 patients with laryngeal carcinoma (21), while identifying a 42.6% incidence of EGFR protein overexpression, found no association of EGFR overexpression with survival or recurrence rates. The study by Rubin Grandis et al. (17) is itself noteworthy for the strength of the demonstrated association between EGFR and transforming growth factor-α.

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Note

Editor's note: B. L. Weber is a member of the Clinical Advisory Board and holds stock options with Myriad Genetics, Inc., that performs BRCA1 and BRCA2 genetic testing.
(TGF-α) levels and survival. This convincing association is predicated on the preceding studies that have largely circumvented the methodologic problems cited above and provided a firm foundation for the retrospective analysis presented here. Meaningful stratification of the patients into the tertiles required for determination of survival curves, median survival times, and resultant multivariate analysis by logrank tests presented here requires a standardized and reliable determination of sample positivity or negativity; this criterion was met by the authors' careful introduction of computerized image analysis of staining intensity (22) and comparison with standard immunohistochemical evaluation (22) and quantitative northern blot analysis of EGFR and TGF-α mRNA levels (23). This group demonstrated that EGFR and TGF-α protein and mRNA levels were greater in tumors and surrounding histologically normal mucosa in samples from patients with head and neck cancers as compared with normal mucosa by agreement of both quantitative and qualitative methods (22,23). Parallel in vitro experiments with tumor-derived cell lines established the growth inhibition of these cell lines by either TGF-α down-modulation (i.e., decreasing TGF-α expression with antisense oligonucleotides) or EGFR inhibition (24,25), which provided a basis for a proposed autocrine growth loop in head and neck cancer, resulting in decreased survival.

The investigation of TGF-α and EGFR coexpression within the same tumor sample is certainly not unique to this study; the presence of both markers has been found to be prognostically significant in esophageal cancer (2) and in Barrett’s-associated esophageal cancer (26). In a study of 97 patients with non-small-cell carcinoma of the lung, Rusch et al. (6,7) reported that, while levels of TGF-α and EGFR expression were elevated in 37/97 (38%) cases, there was no association, either independently or jointly, with either disease-free or overall survival. However, the authors postulated that since the overexpression of both ligand and receptor occurred in all stages of overt lung cancer, they might be acting in an autocrine fashion “to promote local tumor growth without having an impact on tumor progression or metastasis” (7). A similar conclusion was reached by Cohen et al. (27) and by Prewett et al. (4) in studies of prostatic carcinoma; these authors proposed that the paracrine loop in normal and benign prostatic tissues (TGF-α expression by stromal cells and EGFR expression in epithelial cells) changes to an autocrine loop with neoplastic progression, as suggested by the coexpression of both markers by prostatic adenocarcinoma cells in 22/46 (48%) patients (27). In previous studies of coexpression of TGF-α and EGFR in patients with head and neck cancer, disparate results have been reported. Issing et al. (20) demonstrated coincident expression of TGF-α in only three patients; these three patients had the highest levels of EGFR expression. The association between dual marker presence and decreased survival in this group is suggestive, but it is compromised by the small number of observations. Conversely, in an immunohistochemical evaluation of 68 patients with laryngeal carcinoma, Wen et al. (21) found that neither EGFR levels of overexpression (42.6%) nor TGF-α levels of overexpression (55.9%) were associated, independently or jointly, with overall survival rate; those patients with TGF-α overexpression did exhibit a significantly increased recurrence rate.

The data presented here by Rubin Grandis et al. (17) convincingly demonstrate the applicability of TGF-α and EGFR protein expression as prognostic markers in head and neck cancer patients. The protein expression data upon which their statistical analyses were based were extensively and thoroughly established by previous studies. In the 91 patients studied by Rubin Grandis et al., TGF-α and EGFR protein expression levels were each significantly associated with both decreased disease-free and cause-specific overall survival. The effect of these markers on overall cause-specific survival was the same across lymph node stage categories and consistently predicted clinical outcome independently of cervical lymph node status. By multivariate analysis, Rubin Grandis et al. have established that a combination of either increased TGF-α or increased EGFR protein expression with greater lymph node stage provided the strongest predictor of adverse outcome. This study, in conjunction with the previous publications by this group, provides a solid basis for the use of TGF-α and EGFR protein expression determination as a prognostic tool following initial resection in head and neck cancer patients.

The ability to identify high-risk groups requires use of more aggressive or novel therapeutic modalities for those patients. As Rubin Grandis et al. (17) suggest, the use of EGFR- or TGF-α-targeting monoclonal antibodies or immunotoxins has been introduced in some therapeutic trials; however, because the normal EGFR is ubiquitous in normal human tissue (most notably liver), its value as a cytotoxic or cytostatic targeting agent is compromised. As the observation of EGFR gene rearrangement as a frequent accompaniment to EGFR gene amplification has led to the identification of tumor-associated EGFR variant forms that provide the tumor target specificity required, the demonstration of EGFR gene amplification or rearrangement in 6/47 (13%) patients with head and neck cancer of the most advanced stage (28) suggests that immunotherapeutic targeting of variant EGFR molecules may be appropriate for high-risk head and neck cancer patients.

References


