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Adhesion and invasion of follicular thyroid cancer is regulated by vascular endothelial growth factor (VEGF) *in vitro*

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Tumor progression and metastasis are angiogenic dependent

Methods: We studied the effects of VEGF on invasion and adhesion of 3 metastatic follicular thyroid cancer cell lines including a primary (FTC 133) and two metastatic cell lines (FTC236: lymph node-; FTC238: lung metastasis) from the same patient. We tested *invasion* through an 8 μm pore membrane coated with Matrigel by the MTT assay and analyzed *adhesion* of FTC to major components of the extracellular matrix (ECM) (collagen I + IV, fibronectin, laminin, Matrigel).

Results: Compared to the primary tumor, both metastases had a greater basal invasion and a smaller basal adhesion. VEGF (1–100 ng/ml) dose-dependently stimulated invasion of all FTC, but the effect was greatest in the primary tumor. At 50 ng/ml invasion of FTC133 was enhanced by 35% (FTC236: 24%; FTC238: 19%; $p < 0.01$). Basal adhesion of FTC133 was enhanced by 25%, by 34% in collagen IV and by 31% in fibronectin ($p < 0.01$). Again, both metastases were less sensitive to the modulating effects of VEGF.

Conclusions: These data suggest that vascular endothelial growth factor stimulates invasion of follicular thyroid cancer cells through and adhesion to the extracellular matrix.

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Detection of circulating vascular endothelial growth factor (VEGF), matrix metalloproteinase-3 (MMP-3) and -9 (MMP-9) in gastrointestinal cancer

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Purpose: VEGF, MMP-3 and MMP-9 overexpressed in cancer tissues are implicated in tumor-associated angiogenesis and invasion. We measured plasma levels of those circulating molecules to clarify the clinical significance in gastrointestinal cancers.

Methods: Plasma samples were collected from 20 healthy controls, 12 patients with benign tumors (colon adenoma; 10, gastric adenoma; 2), 14 advanced cancer patients without metastasis (gastric; 9, colorectal; 5), and 40 cancer patients with metastasis (gastric; 25, colorectal; 15). Plasma levels of VEGF, MMP-3 and MMP-9 were determined by ELISA (R&D systems).

Results: Plasma VEGF and MMP-9 levels increased in accordance with the disease progression (healthy controls; VEGF 26 ± 8.2 pg/ml, benign tumors; VEGF 53 ± 33, MMP-9 22 ± 6.8 ng/ml, non-metastatic cancers; VEGF 41 ± 29, MMP-9 26 ± 8.2, and metastatic cancers; VEGF 148 ± 101, MMP-9 44 ± 32). But such relation was not noticed in MMP-3. When the discrimination level of VEGF was set at over 100 pg/ml, the sensitivity and specificity on detecting metastatic patients were 53% (21/40) and 95% (21/22), respectively. These were superior than those of MMP-3 and MMP-9. Plasma VEGF levels were correlated with MMP-9 ($r = 0.35$, $p = 0.010$) and CEA ($r = 0.40$, $p = 0.0049$) levels, but not with MMP-3 ($P = 0.588$) and CA19-9 ($P = 0.16$) levels.

Conclusion: Circulating VEGF in gastrointestinal cancers was suggested to be an useful diagnostic marker for tumor progression, especially metastasis.

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Determination of changes of tissue-specific genes expression in human brain tumors

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Purpose: Characterization of genes which expression is activated or repressed in human brain tumors, should provide new data for understanding of the mechanisms of tumors arising and progression. New genetic markers can be used in tumor diagnostics. The isolation of genes, differentially expressed in human glial tumors, is the objective of this study.

Methods: Differential hybridization of organized human fetal brain cDNA library (provided as high density arrays of clones, by H. Lehrach, ICRF, London) with cDNA probes synthesized on the mRNAs of normal human fetal brain, human meningioma and human anaplastic astrocytoma. The cDNAs which gave evident increase or decrease of the hybridization signal with tumor-specific probes were amplified and characterized by restriction-hybridization analysis and sequencing.

Results: The comparison of Southern hybridization patterns of DNA from isolated clones with normal human brain cDNA, astrocytoma cDNA and glioblastoma cDNA probes showed a visible decreasing of unknown mRNAs H2345, C063, 0073, and J2448 content in both tumors. The increasing mRNA content was observed for unknown clones C1134, H1352, L1033, for mRNA encoding valosin containing protein involved in signal transduction pathway and J1041 mRNA homologous to mouse TSC-22 mRNA.

Conclusion: Differential hybridization of robotically spotted high density arrays of human brain cDNA clones with cDNA probes synthesized on the mRNAs from different tumors showed characteristic pictures, specific for given tumor. The nucleotide sequences of isolated genes are under investigation.

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Cyotokines and surrogate markers in HIV-related opportunistic malignancies

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Purpose: To compare the levels of IL6, TNF-α, ferritin, C-reactive protein (Crp), β2-microglobulin (B2m), and CD4+ lymphocyte counts in HIV-infected patients with Kaposi's sarcoma (KS) and opportunistic infections (OI) vs. HIV patients with non-Hodgkin's lymphoma (NHL) and OI.

Methods: Eight patients were enrolled: 3 NHL (B cell, 2 low and 1 intermediate grade, stage IV, chemotreated) plus OI (2 extrapulmonary TB and 1 CMV infection), 5 KS (end-stage, viscerocutaneous, chemotreated) plus OI (4 CMV + mycobacteriosis, 1 CMV + CNS toxoplasmosis). All patients were severely ill and died shortly thereafter.

Results:

N.V.	IL6 (3.0–6.5 pg/ml)	TNF-α (3.0–20 pg/ml)	Ferritin (15–250 ng/ml)	Crp (<1 mg/dl)	B2m (0.9–3.0 mg/l)	CD4+ (504–1224/ /mmc	Mean age (years)
KS + OI	60.4	55.4	2351	0.9	4.5	25	38.8
NHL + OI	57.6	78	1404	1.6	4.5	152	44.3

Conclusion: Although preliminary, our data seem to underscore that: the combination of KS + OI appears to be lethal quite later than the simultaneous occurrence of NHL + OI: patients belonging in the former group died with lower CD4 cell counts and higher levels of ferritin, both roughly consistent with an "older" infection and with a more impaired immune function. IL6 and B2m behaved quite similarly in the two groups, while TNF-α and Crp showed only minor differences. This study is going on to confirm these preliminary findings.

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Detection of tumor progression in NSCLC stage IIIB/IV patients by serial measurement of CYFRA 21-1, TPA-M, TPS, and CEA

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Purpose: The most common application of tumor markers in lung cancer is disease monitoring. In a prospective study we have investigated the value of serially assessed tumor markers CEA and cytokeratin fragments CYFRA 21-1, TPA-M, and TPS to detect tumor progression in NSCLC stage IIIb/IV patients.

Methods: Tumormarker concentrations were measured using commercially available enzyme immuno assays (CEA: EIA Roche, Basel; CYFRA 21-1: EIA Boehringer, Mannheim; TPA-M: IRMA, Sangtec Medical, Bromma, Sweden; TPS: IRMA, Beki, Bromma, Sweden). Clinical response to therapy was evaluated according to standard criteria of the WHO. For the assessment of response to therapy by changing in the marker levels the difference between 2 consecutive levels must exceed 30%. This value is based on the formula: $\text{Diff} = 2\sqrt{2} \times V_K$ (V_K: interassay coefficient of variation, i.e. <10%)

Results: Tumor progression according to WHO criteria was recorded in 15 patients monitored by CYFRA 21-1 (TPA-M: 22, TPS: 21, CEA: 14). For CYFRA 21-1 67% (TPA-M: 55%, TPS: 67%, CEA: 43%) of the evaluations by rising marker levels were concordant with the clinical assessment. Most