Human Transcription Factors RT2 Profiler PCR Array (PAHS-075A − SABiosciences − Qiagen) was used to evaluate the profile of the expression of 84 transcription factors using real time PCR. The PCR array was validated through analysis of the expression levels of 8 genes by quantitative real time PCR using EVA Green dye.
Results: We analyzed transcription factors downstream of signaling from cytokines, chemokines and growth factors, signaling from androgen, B-cell, G-protein, T-cell and Toll-like receptors, and of signal transduction pathways like JAK/STAT, JNK, MAPK, NFkB, Notch and WNT. The SET profile showed 74 down-regulated and 7 up-regulated genes. Eight genes were validated (6 down-regulated and 2 up-regulated) in HEK293 cells overexpressing SET and confirmed in HN13 cells after SET knockdown.
Conclusions: SET accumulation in OSCC (previously validated by us) may control the expression of many transcription factors fundamental in the cell signaling and this could have implications in cancer progression and treatment.

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DNA methylation: an epigenetic pathway to cancer and a promising target for anticancer therapy in breast cancer

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Objective: To determine whether Estrogen Receptor 1(ESR1) (+) and ESR1(−) status relates to epigenetic changes in breast cancer-related genes and to correlate with molecular breast cancer subtypes.
Methods: Since January/02 to June/05,we quantified methylation levels ERS1 gene in serum of 92 pts breast cancer.A PCR quantitative technique was used to analyze levels of methylation gene.We also examined and correlationed the expression of ESR1 in tumors by immunohistochemistry with molecular phenotype.
Results: Median age was 58 years (32−88); 69% were postmenopausal women. Nodal involvement (N0; 63%, N1; 30%, N2; 7%),tumor size (T1; 58%, T2; 35%, T3; 4%,T4; 4%) and grade (G1;20%,G2;37%,G3;30%).Of the cases, 37pts (40%) were Luminal A (LA), 32 pts (33%) Luminal B (LB), 14 pts (15%) Triple-negative (TN) and 9 pts (10%) HER2+. The methylated ESR1 in serum was significantly associated with ESR1(−) in breast tumors >80%($p = 0.0179$). Methylation ESR1 was preferably associated with TN(80%) and HER2+(60%) subtype. Nevertheless unmethylation ESR1 was found more frequently in LA(71%) and LB(59%) phenotype. With a median follow up of 5 years, we found worse overall survival (OS) with more frequent ESR1 methylation gene($p > 0.05$), Luminal A;ESR1 Methylation OS at 5 years 81% vs 93% when was ESR1 Unmethylation. Luminal B;ESR1 Methylation 86% SG at 5 years vs 92% in Unmethylation ESR1. Triple negative; ESR1 Methylation SG at 5 years 75% vs 80% in unmethylation ESR1. HER2; ESR1 Methylation SG at 5 years was 66.7% vs 75% in unmethylation ESR1.
Conclusions: Gene promoter region hypermethylation is a significant event in primary breast cancer. However, its impact on tumor progression and potential predictive implications remain relatively unknown. Our study identifies the presence of variations in global levels of methylation promoters ESR1 genes in breast cancer with different phenotype classes and shows that these differences have clinical significance. Although numerous issues remain to be resolved,quantitative measurement of circulating methylated DNA may be of significance in the assessment and search of targeted therapy resistance related to ESR1 and HER2 status by epigenetic or transcriptional cancer therapy.

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Expression of methylthioadenosine phosphorylase (MTAP) in malignant pleural mesothelioma (MPM) and its implication for pemetrexed-based chemotherapy

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Introduction: The *MTAP* gene encodes for a key enzyme in the methionine salvage pathway. This gene is located at chromosomal locus 9p21, 100kb telomeric to *p16*, which is frequently deleted in malignant pleural mesothelioma (MPM). MTAP-deficient tumors are dependent on the *de novo* purine synthesis pathway, which is inhibited by drugs such as pemetrexed. This study investigates the MTAP expression in MPM and its relationship to pemetrexed-based therapy.
Material & Methods: DNA was extracted from tissue sections of paraffin-embedded tumor samples from MPM patients treated with pemetrexed-based therapy. Gene copy variation (GCV) of MTAP was determined using multiplex ligated PCR assay (MLPA), which included target probes for *MTAP* and *p16*. In addition, immunohisto- chemistry (IHC) was used to detect MTAP protein, using a validated monoclonal antibody. IHC was graded by two independent assessors using a composite score system which consisted of percentage score for positively stained areas multiplied by the intensity of staining, resulting in a score of 0–300. These findings were then correlated to tumor histopathology and clinical outcome, including disease control rates (DCR), median time to treatment failure (TTF) and overall survival (OS).
Results: Data for 52 MLPA and 59 IHC specimens were available for analysis. MTAP GCV was noted in 55% of samples. In all samples with MTAP deletion, there was co-deletion of *p16*. IHC score of 0–100 (null or minimal staining) and 101–300 (moderate to strong staining) were found in 55% and 45% of samples respectively. An association was observed between loss/minimal expression of MTAP protein and early disease stage ($p = 0.04$). The loss of MTAP gene or protein however, showed no significant association with DCR, TTF or OS.
Conclusion: Loss of MTAP protein expression was associated with early stage disease, but neither its gene copy status nor protein expression were predictive for clinical outcome in MPM patients treated with pemetrexed-based therapy.

	MTAP MLPA			MTAP IHC		
	+	−	p	+	−	p
DCR (%)	51	57	0.2	60	71	0.4
TTF (mths)	5.3	7.4	0.7	5.1	7.4	0.5
OS (mths)	6.2	10.3	0.9	10.0	10.1	0.7

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Promoter methylation of the RGC32 gene in non-small cell lung cancer and its clinical implications

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Background: Lung cancer is the leading cause of cancer-related deaths worldwide. Epigenetic inactivation of certain genes by aberrant promoter methylation is recognized as a crucial component in the initiation and progression of lung cancer. Response gene to complement 32 (*RGC32*) is formerly identified as a cell cycle regulator induced by activation of complements; however, its role in carcinogenesis is still controversial.
Methods: We have examined the methylation status in the promoter region of *RGC32* gene in non-small cell lung cancers (NSCLCs) using a methylation-specific PCR, and correlated the results with clinicopathological features.
Results: *RGC32* methylation was found in 45 of 173 NSCLCs (26.0%) and was related to the gene expression. *RGC32* methylation was more frequent in females than in males ($P < 0.05$). *RGC32* methylation was not significantly associated with the prognosis of patients; however, when the patients were categorized by *TP53* mutational status, the effect of *RGC32* methylation on prognosis was significantly different between those with and without *TP53* mutations ($P = 0.005$ [test for homogeneity]); specifically, *RGC32* methylation was associated with a significantly worse survival in the cases with wild-type *TP53*, whereas it exhibited a better survival outcome in the cases with *TP53* mutations.
Conclusion: The current findings suggest that methylation-associated down-regulation of *RGC32* plays an important role in the pathogenesis of NSCLC, particularly in females. However, further studies with a large number of cases are needed to confirm our findings.

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Immunomodulatory activity of SGI-110, a 5-aza-2′-deoxycytidine-containing demethylating dinucleotide

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Background: We have recently reported that aberrant DNA hypermethylation down-regulates the expression of components of the "tumor recognition