80% of the mutations cause a single amino acid change (V599E). We have subsequently shown that BRAF mutations cause activation of the BRAF kinase activity, are transforming in NIH3T3 cells and often (but not always) render the cell independent of signalling through RAS proteins. The patterns of BRAF mutation and their associated biology have revealed new insights into kinase function, pathway function and have generated a plausible new target for drug development. In the future as systematic genome wide mutational screens progress they will reveal insights into global patterns of mutation that differ between individual cancers and cancer types and will provide information on fundamental parameters of human cancers: how many genes are mutated and implicated in the genesis of a single human cancer and how many different cancer genes are there?

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Predicting breast cancer behaviour by genetic analysis

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In the treatment of breast cancer, patient tailored therapy is becoming increasingly important. Decisions on optimal treatment include the choice between mastectomy and breast conserving treatment; dose of radiotherapy; and decisions on adjuvant chemotherapy and hormonal therapy.

Specific DNA alterations, most notably amplification of oncogenes and inactivation of tumour suppressor genes, will have an influence on tumour cell behaviour and may therefore be clinically useful. The assessment of germline alterations in the BRCA1 and BRCA2 genes are already used to identify women with a genetically determined increased risk to develop breast cancer. It has been shown that breast carcinomas with an amplified HER2 gene respond to optimal dosed anthracyclin based therapy and may be less sensitive to tamoxifen. The unravelling of other associations between genetic alterations and tumour behaviour can be expected to impact on the clinical management of breast cancer patients.

Gene expression profiling by micro-array analysis allows the study of the level of expression of large numbers of mRNA's in a single experiment. Gene expression analysis can be used to subclassify tumors on the basis of hierarchical cluster analysis in specific subgroups; supervised cluster analysis can be used to directly link gene expression profiles to clinical characteristics, including prognosis and response to various forms of treatment.

We have used microarray analysis, first on a series of 117 breast carcinomas and more recently on a series of 295 breast carcinomas.

We have defined a gene expression profile of 70 genes that is predictive for a short interval to distant metastases (<5 yrs) in lymph node negative (LN0) patients. We have validated the prognostic value of this gene expression profile in lymph node negative patients; and also in premenopausal lymph node positive patients. The profile outperforms all currently used clinical parameters in predicting outcome of disease.

At present, we are employing gene expression profiling to identify patients at high risk of local recurrence after breast conserving therapy and to predict the responsiveness of primary and metastatic disease to systemic treatment.

As a result, we expect that in the future, gene expression profiling of breast cancer will be used to guide optimal therapy.

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Expression and CGH analysis in soft tissue sarcomas, bladder cancer, and prostate cancer

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We have performed Expression and Comparative Genomic Hybridization studies onto cDNA arrays for a variety of cancer types including sarcomas, prostate cancer and bladder cancer. Expression profiles were obtained for 37 leiomyosarcomas. The dataset was first filtered to select a set of 335 genes whose expression varied most widely between primary and metastatic tumours. Clustering analysis of non-metastatic tumours using this gene set revealed that the tumours could be divided into two distinct groups. The metatstatic potential of primary tumours in the two groups were dramatically different (log-rank test p=0.001). We concluded that expression profile could predict metastatic potential of human sarcomas and that the ability to metastasis was a bulk property of the tumour. Expression studies have also been performed on primary prostate cancer with the objective of identifying new potential prostate markers. In this study we used microarrays randomly selected from a prostate LNCaP cDNA library. Several novel potential prostate cancer markers have been identified. CGH onto Geneset microarrays studies were performed usingh prostate and bladder DNA to identify regions of genetic gain and loss. Several novel classes of genetic alteration have been identified. Acknowledgements: We thank the National Cancer Research Institute, Cancer Research UK and the Medical Research Council for funding this work.

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Proteomic pattern analysis serum for early detection of ovarian cancer

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Diagnosis and management of cancer requires tools with both high sensitivity and specificity. The minimally invasive cervical smear has demonstrated how such a test can change the public health profile of a cancer from deadly to cured. Neither a robust test, nor reliable or specific early symptoms are available for ovarian cancer and other solid tumors. Current approaches testing one protein or gene at a time will not address this expeditiously. New high throughput cost-efficient technologies are needed. These should focus on available patient resources, blood or urine, or minimally invasive approaches such as cervical smears. Proteomics, the study of the expressed proteins and protein fragments, has been applied experimentally to cancer diagnostics. Ovarian cancer is a rare disease with 1:2500 postmenopausal women affected in their lifetime. It is diagnosed in advanced stage in over 70% of women with similar trends for pancreatic, gastric, and other cancers. A specificity of 99.6% on a background of 100% sensitivity is the target requirement for an ovarian cancer biomarker to yield positive predictive value of 10% but is not powered for the detection of early stage cancer, occurring in 15% of ovarian cancer cases. Early detection of ovarian cancer can increase frequency of long term survival to over 90%. The goal of proteomic monitoring is development of a reliable screen to identify stage I/II disease and to allow rapid and optimal patient intervention. We have applied mass spectroscopy (MS)-based proteomic screening of serum with bioinformatic pattern analysis for ovarian cancer biomarker development under the hypothesis that circulating blood contains information from organ-confined disease. Surface-enhanced or matrix-assisted laser desorption and ionization MS has been used to detect low molecular weight proteins, an untapped information reserve. Small serum samples yield datastreams containing over a hundred thousand features. A protein separation on a solid-phase capture matrix directs the view of the proteome. Advanced bioinformatics algorithms mine the MS datastreams for diagnostic patterns of information. The algorithm is trained with data from known samples to define the signature pattern. This pattern is tested with blinded unknowns for validation. The weak cation exchange matrix analysis yielded a experimental diagnostic signature pattern 99-100% sensitive and 99-100% specific when queried with blinded unknown samples (n=250). All 36 early stage cases were correctly identified as cancer. The features comprising the diagnostic pattern can soon be isolated and identified with newer. We are initiating a large scale prospective blinded study to determine the robustness of these early findings and to form the basis for prospective randomized testing. Application of MS coupled with bioinformatic techniques has promise for identification of discriminating protein signature patterns in the blood of organ-confined ovarian and other cancers.

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Early colorectal cancer - treatment choice

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Early colorectal cancer unfortunately is a seldom diagnosis in symptomatic patients. The proportion of 10% however may be increased to 45%, provided that screening for colorectal cancer (CRC) with fecal occult blood test programs are accepted in average risk persons above 50 years of age and positive stool tests are followed by a complete colonoscopy.

The pT-stage may be defined, when the resection margin is free of tumour tissue. Before treatment it is possible to define the T-stage by intraluminal ultrasound examination with a high accuracy; and local excision (endoscopic polypectomy, peranal excision Transanal Endoscopic Microsurgery) of a TI

tumour cures the patient in more than 90%, provided that no spread has occurred (NO, MO).

The prognosis for TII tumours is not as good (75% cure), and only highly differentiated tumours with no spread as evaluated by MR scanning should be considered possible candidates for local treatment. When resection margins are involved, the tumour is poorly differentiated, and/or vascular or lymphatic invasion has been demonstrated the local treatment should be considered insufficient, and the patient should be offered major surgery immediately. Later major surgery for recurrence (salvage treatment) has a worse prognosis (less than 50% 5 years crude survival).

In patients with severe co-morbidity resulting in a high immediate mortality after major surgery, the above criteria should not be strictly followed, and local treatment, which carries a very low immediate mortality, may be preferred. However, prospective evidence to justify such a policy is scarce.

Adjuvant preoperative radiotherapy may have a place in this context; no large RCT's are available, but selected series have demonstrated 5 years survival up to 75%. It may even be possible to cure some of the rectal cancers by radiotherapy alone.

It has been suggested to give adjuvant chemotherapy for all TII rectal tumours, and high risk TI tumours before or after local excision.

In conclusion, local treatment for early CRC demands a detailed evaluation of stage and clinical status of the patient before embarking on local treatment, and the stage must be revised when the full pathology report is available.

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The therapeutic relevance of the molecular biology of colorectal cancer

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Introduction: The fluoropyrimidine 5-Fluorouracil (5-FU) is widely used in the treatment of colorectal, cancer. Fluoropyrimidines were developed in the 1950s following the observation that rat hepatomas utilized uracil more rapidly than normal tissues, suggesting that uracil metabolism was a potential target for antimetabolite chemotherapy. The mechanism of 5-FU cytotoxicity has been ascribed to the misincorporation of fluoronucleotides into RNA and DNA and to the inhibition of the nucleotide synthetic enzyme thymidylate synthase (TS). While 5-FU in combination with other chemotherapeutic agents improves response rates and survival in breast and head and neck cancers, it is in colorectal cancer that 5-FU has had the greatest impact. It has been demonstrated that 5-FU-based chemotherapy improves overall survival and disease-free survival of patients with resected stage III colorectal cancer. Nonetheless, in the metastatic disease setting, response rates for 5-FU-based chemotherapy as a first-line treatment for advanced colorectal cancer are only 10-20%. Combination of 5-FU with the newer chemotherapies - irinotecan (CPT-11) and oxaliplatin - has improved the response rates of advanced colorectal cancer to 40-50%. However, despite these improvements, new therapeutic strategies are urgently needed. DNA microarray technology has the potential to identify novel genes that may play key roles in mediating resistance to 5-FU-based chemotherapy. Such genes may be therapeutically valuable as predictive biomarkers of 5-FU chemosensitivity and/or provide new molecular targets that overcome drug resistance. This talk reviews how pre-clinical and clinical studies have impacted on the clinical use of 5-FU and discusses how DNA microarray profiling may affect its future clinical application.

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Surgery - radiotherapy: when and how to combine?

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According to meta-analyses covering all randomised trials it is obvious that the best reduction in local recurrences is received if radiotherapy is given preoperatively. In the postoperative setting similar data can be achieved if radiotherapy is combined with chemotherapy. Moreover, data do support that preop radiotherapy is superior to postoperative treatment.

The problem with the preoperative approach is the drawback of treating too many patients in vain. Therefore, good staging is necessary. Based upon the data from all studies, low rectal cancer, tumours growing anteriorly, narrow male pelvis, and obese patients are at risk. Also stage of disease is important to find preoperatively. The problem with this risk analysis is the fact that all data are from trials using "old fashion" surgery. With a more appropriate surgical technique, i.e., the use of TME (Total Mesorectal Excision), the risk calculations might be changed.

When taking staging into consideration it is important to focus on wha part in the multimodal treatment, which is the "weak" one. Also it is importan to identify situations when a combined treatment is superfluous. The "weak part is the way the surgeon can make a curative loco-regional procedure and the best prognostic factor is the circumferential margin. The loca only imaging technique picking up the circumferential margin with a high sensitivity is modern MRI staging. Based upon good MRI imaging it is possible to divide the patients with a rectal cancer into three groups; good bad and ugly. The good cases have a wide circumferential margin and neec no radiotherapy. The bad one has a narrow circumferential margin anc would certainly benefit from preoperative radiotherapy. The ugly looking group has a positive circumferential margin. Those patients will probably dc best if preoperative radiotherapy is combined with chemotherapy.

Conclusion: Radiotherapy should be used selectively based upon the preoperative imaging of the tumour size. In this staging not only the T-stage but also the N-stage is important in terms of identifying the distance to the circumferential margin when the specimen has been retrieved after high quality TME surgery.

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Chemoprevention - a realistic option

Abstract not received.

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Social inequality and risk of cancer - do we have a problem?

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Socio-economic status is known to be associated with the risk of many cancers, though limited data are available on potential variation in these associations between countries and over time within countries. Possible factors responsible for socio-economic differences in cancer risk include lifestyle factors (e.g., smoking, diet, reproductive factors and sexual behaviour), work and living environment, and access to health care services.

In large population-based studies on social class and cancer risk among working-aged Finns during 1971-1985/95 (e.g., [1,2]), the cancers found to be associated with low social class included, e.g., those of the lip, oesophagus, stomach, larynx, cervix uteri and lung (the latter only in men). Cancers of the colon, prostate and breast, and skin melanoma in the trunk and limbs were most common in high social classes. In some cancers the positive social class gradients diminished (e.g., testicular cancer) or disappeared (e.g., cancer of the corpus uteri) during the observation period; the positive gradient seen in female lung cancer in the 1970s reversed to a negative one in the 1980s. Most of the observed patterns can be at least partially explained by the social class distribution of the known risk factors for cancers in Finland. Overall, it was estimated that about 26% of all cancer cases among working-aged Finns during 1971-85 could have been avoided if all social classes had been able to maintain a lifestyle similar to that of the social class with the lowest incidence.

In conclusion, known risk factors for cancer, particularly smoking, appear to account for a substantial proportion of the socio-economic differences in cancer risk, but unknown factors are likely to contribute as well. A current challenge for epidemiologists is thus to uncover what is still hidden behind the socio-economic differences in the risk of certain cancers; that for policy makers and health care practitioners is to continue the fight against smoking and other unhealthy behaviour and to reduce social inequalities (where such exist) in access to health care services.

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