Pathological complete response was used as a surrogate measure of chemosensitivity.
One hundred and twenty-five ER negative tumours (55 pCR) were tested: 66 in the FEC arm (28 pCR) and 59 in the T → ET arm (27 pCR). RNA was prepared from sections of frozen biopsies taken at diagnosis and hybridized to Affymetrix X3P microarrays. In vitro single agent drug sensitivity signatures were combined to obtain FEC and T → ET regimen-specific signatures.
The regimen-specific signatures significantly predicted pCR in patients treated in the appropriate arm ($p < 0.0001$). The FEC predictor had a PPV of 68% (27/40, 52−80%) and NPV of 96% (25/26, 81−99%). The T → ET predictor had a PPV of 71% (25/35, 55−84%) and NPV of 92% (22/24, 74−98%). Analysis of tumour size, grade, nodal status, age and the regimen-specific signatures showed that the genomic signatures were the only independent variables predicting pCR at $p < 0.01$.
Conclusions: We have validated the use of regimen-specific drug sensitivity signatures in the context of a multicentre randomised trial. Selection of patients with these signatures would increase the pCR rate from 44% to around 70% in the patients tested here. The high NPV of both signatures (the NPV for each regimen-specific genomic signature is over 90%) indicates the potential to select patients who should be considered for trials with new agents. Organising clinical trials on this basis would have important implications for the subsequent use of the new agents tested.

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High throughput expression studies in primary cutaneous melanoma

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Main Message: Genes identified in a validated and reproducible signature prognosticating metastases or death are mainly associated with replication or DNA repair. For replication, two pathways are over-represented: the replication origins firing genes (ROF) and the separation of sister-chromatids by securin. Poor prognostic melanomas are characterized by a global overexpression of ROF-related genes. MCM-4 and MCM-6 immunoexpression is strongly correlated with metastasis free survival and OS. This prognostic value is maintained when age, sex, location of the primary tumor, thickness and ulceration are introduced in the multivariate model. The whole ROF system is locked by geminin that complexes CDT1 and CDC6. When CDT1 and CDC6 are released, they can recruit MCMs at the replication origins. When this interaction is altered, for instance when BRCA1-IRIS relieves geminin-CDC6 interaction, the helicases cascade becomes overactive leading to replication increase. hPTTG gene, coding for securin, is among the top genes of the prognostic signature. Securin has three known activities: it blocks the sister-chromatids separation in stabilizing separase, it stimulates angiogenesis and it decreases p53 transcription. Securin acts as an oncogene and provides a positive growth advantage as it downregulates sister-chromatids separation and therefore avoids the cells to enter into aneuploidy. P53 transcription inhibition leads to a decrease in p53-mediated apoptosis. Securin immunohistochemical expression is observed in vertical growth phase whereas melanomas in radial growth phase do not express securin.
Overexpression of DNA-repair genes is associated with metastases or death. Increase in DNA repair capacity could explain spontaneous resistance of most melanomas toward radiotherapy and alkylating agents. In this DNA repair genes list, NER and BER family genes are not represented. On the contrary, most of the DNA repair genes present in this group are involved in post-replicative repair of DNA lesions. This is in accordance with the hypothesis that aggressive melanomas need a fast and effective replication, and need to repair mistakes induced during replication. One of the genes of which overexpression is most evidently associated with poor prognosis is topoisomerase 2A (Top2A). Top2A codes for an enzyme that is essential for replication and chromosomal segregation. Actually Top2A expression seems to be a consequence rather than a cause of cell proliferation. However, cells that overexpress Top2A, and PCNA with which it interacts, are much more resistant toward alkylating agents.
In parallel with expression genomics studies, CGH array studies have provided important informations to refine the melanoma classification. The expression signature associated with B-Raf mutations reveals a strong association between B-Raf mutation and CD63 overexpression ($p = 10^{-14}$!). These results and their consequences will be discussed during the presentation as well as the intergration of expression and aCGH data.