evaluation of MAPK signaling inhibitors, VEGF signaling inhibitors, survival kinase inhibitors, and cyclin-dependent kinase inhibitors in melanoma. An overview of melanoma biology and established targets, followed by a summary of completed and ongoing early phase clinical trials will highlight the failure of the first generation of targeted therapies to improve outcomes as single agents. In contrast, early hints of improved outcomes have been generated by clinical trials testing the combination of sorafenib and chemotherapy. The potential of targeted therapies in combination with chemotherapy or regimens consisting of multiple targeted therapies will be explored as increasing evidence suggests that combination therapeutics could finally impact the outcome of metastatic melanoma.

175 INVITED

Target discovery in melanoma

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Tumor metastasis to regional (sentinel) lymph nodes represents the first step of tumor dissemination in most melanomas and serves as a major prognostic indicator for disease progression. However, little is known about the mechanisms how tumor cells gain entry into the lymphatic system. In this respect, we have previously shown that tumors can actively induce the formation of lymphatic vessels (leading to the new concept of tumor lymphangiogenesis) and that tumor lymphangiogenesis was correlated with lymph node metastasis in an orthotopic breast cancer model. Our studies in human cutaneous malignant melanomas demonstrated the presence of both intratumoral and peritumoral lymphangiogenesis in cutaneous melanoma. They also showed that primary melanomas that later metastasized were characterized by increased lymphangiogenesis – as compared to non-metastatic tumors – and that the degree of tumor lymphangiogenesis can serve as a novel predictor of lymph node metastasis and overall patient survival, independently of tumor thickness. Moreover, we found that the extent of lymphatic vessel growth in primary human cutaneous melanomas was the most sensitive parameter for predicting whether these tumors had already metastasized to the sentinel (draining) lymph node at the time of surgery. Importantly, we have recently found - for the first time - that metastastic tumor cells can induce lymphatic vessel growth within lymph nodes, furthering their metastatic spread. This has led to the new concept of lymph node lymphangiogenesis. Surprisingly, we found that tumor cells can induce lymph node lymphangiogenesis already before they metastasize, giving a new twist to the seed-and-soil hypothesis and suggesting that tumors can prepare lymph nodes for their future arrival. We have characterized the transcriptional profile of normal and of tumor-associated lymphatic and blood vessels by laser capture microdissection. This has enabled us to identify a number of new targets for anti-(lymph)angiogenic cancer therapy. Taken together, tumor lymphangiogenesis has emerged as a novel prognostic parameter for the metastatic risk of human melanomas, and inhibition of tumor-associated lymphangiogenesis appears to represent an exciting new strategy to inhibit cancer progression.

Symposium (Wed, 26 Sep, 14:45−16:45)
New drugs and new tools in the treatment of patients with myeloma

176 INVITED

New insights in the biology of multiple myeloma: basis for novel therapies

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Multiple myeloma (MM) remains as an incurable disease; therefore, new treatment strategies are needed in order to improve the outcome of these MM patients. The increase knowledge in MM biology is already contributing to a more specific drug design, and we have recently learned that in the pathogenesis of MM, as important as the malignant cells themselves, is their interaction with the microenvironment.
Multiple myeloma requires a multistep transformation process that implies the sequential generation of primary Ig translocations, chromosomal instability (including mutations – RAS s– and deletions − RB), as well as secondary translocations. Most Primary immunoglobulin gene translocations occur early in the pathogenesis of MM. These translocations, which are mediated by errors in immunoglobulin heavy-chain switch recombination, result in the juxtaposing of an oncogene and an immunoglobulin enhancer. On the basis of Ig H translocations MM patients can be divided into 5 subgroups: (1) D-type cyclins: Cyclin D1 on 11q23, Cyclin D3 on 6p21 and Cyclin D2 on 12p13 (25% of cases); (2) MMSET/FGFR3 proteins (4p16.3) (15% of cases); (3) B-zip transcription factors: c-maf on 16q23 and maf B on 20q11 (15% of cases); (4) other Ig H translocations (20% of cases); and (5) No Ig H translocations (25% of cases). Secondary oncogenic events may involve both genes different from Ig locus, as well as the 14q32 region, as occur in the c-myc translocations.
Some of these molecular events represent potential therapeutic targets. Thus t(4;14) translocation generate a constitutive activation of the oncogenic receptor tyrosine kinase FGFR3 with subsequent phosphorylation of the antiapoptotic STAT3 signaling pathway. Therefore, the use of Kinase inhibitors of FGFR3 tyrosine kinase as well as Kinase inhibitors of cyclin dependent kinases would be attractive therapeutic targets. Similarly C-maf, that is over expressed in MM patients with t(14;16) as well as in some MM cases lacking this translocation, also represent a potential target.
The second area of MM pathogenesis that may have important implications for treatment intervention is the interaction between the malignant cell and the bone marrow microenvironment. MM cells adhere to the extra cellular matrix (ECM) proteins and bone marrow stromal cells (BMSC) through a series of adhesion molecules, such as the β1-integrin family (VLA-4, VLA-5 and VLA-6 or CD49d, e and f, respectively) as well as ICAM-1 and VCAM-1. Adhesion of myeloma cells to BM microenvironment induces a CAM-DR phenotype (cell-adhesion-mediated drug resistance). Interruption by downregulating the interactions between the tumor cell and its microenvironment can potentially halt cell growth and proliferation, and be of benefit to patients with MM. The binding of MM cell to BM microenvironment it also induces the transcription and secretion of cytokines (TNFa, IL-6, IGF-1, IL-21, SDF1a, VEGF), by both the PC and BMSC, which triggers signalling pathways (such as the RAF/MEK/MAPK, PI3K/AKT, and JAK/STAT pathways), that promote cell proliferation and prevent apoptosis. These pathways are also potential targets for therapeutic intervention.

177 INVITED

New drugs

M. Boccadoro. *Italy*

Abstract not received.

178 INVITED

Autologous and allogeneic transplantation in multiple myeloma

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In patients with multiple myeloma (MM) high-dose therapy/stem cell transplant (HDT/SCT) can be applied in different clinical settings and by using different approaches. In patients with relapsed/refractory disease, HDT/SCT is of no benefit. In contrast, patients with sensitive relapse are the most likely to benefit. Concerning newly diagnosed patients, two randomized trials showed that autologous transplant resulted in higher response rate as well as in longer progression-free and overall survival when compared with standard chemotherapy; however, other three randomized trials failed to show a significant survival advantage in favour of high-dose therapy. In any event, autologous transplantation in currently considered as part of the up-front therapy in younger myeloma patients. Double autologous (tandem) seems to be of benefit for patients not achieving complete remission or very good partial response with a single procedure. Allogeneic transplant with conventional conditioning results in a high response rate and cure in about 20% of patients. However, the transplant-related mortality (TRM) is between 30 and 50%. For this reason, the so-called “mini-allogeneic” or reduced-intensity conditioning allogeneic transplant (allo-RIC) is currently used in most institutions. The TRM with allo-RIC is about 20%; however, the relapse rate is higher than with conventional conditioning, this resulting in a similar long-term outcome with the two allogeneic approaches. In patients with advanced disease the allo-RIC seems to be of no benefit. In MM, HDT/SCT constitutes an important tool for tumour mass decrease. In the current era of novel agents and more effective treatment combinations, the additional tumour mass reduction achieved with HDT/SCT will hopefully result in an improved long-term outcome for patients with multiple myeloma.

179 INVITED

Waldenström macroglobulinaemia

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Waldenström’s macroglobulinemia (WM) results from the clonal proliferation of lymphocytes that produce monoclonal immunoglobulin M (IgM) and always involves the bone marrow. The normal counterpart of WM malignant cell is believed to be a post-germinal center B cell. WM cells do not bare

IgH rearrangements and the main recurrent cytogenetic abnormality is 6q deletion. Gene-expression studies showed that WM has a gene expression profile similar to CLL and normal B cells; IL6 and MAPK signaling pathway associated genes are the most significantly up-regulated genes.
Clinical manifestations and laboratory abnormalities in WM are related to direct tumor infiltration and the amount and specific properties of the monoclonal IgM. Weakness and fatigue are usually related to anemia secondary to marrow infiltration; B symptoms are common. Hepatomegaly, splenomegaly and lymphadenopathy occur in 15−30% of patients. Peripheral neuropathy, symptoms and signs of cryoglobulinemia, cold agglutinin disease or amyloidosis may predominate. Hyperviscosity syndrome occurs in 10−30% of patients at diagnosis.
Diagnosis of WM should be confined to patients with a lymphoplasmatcytoid lymphoma with compatible immunophenotype involving the bone marrow with demonstrable IgM monoclonal protein. Differential diagnosis of WM includes other B-cell lymphoproliferative disorders including splenic marginal zone lymphoma, CLL and IgM-Multiple myeloma. Immunophenotyping and clinical criteria are helpful for an accurate diagnosis. IgM-MGUS is far more common than WM and is characterized by the absence of morphologic evidence of marrow infiltration. Patients with monoclonal IgM and overt manifestations such as peripheral neuropathy, cryoglobulins, cold agglutinin disease, AL amyloidosis or other rare manifestations, without evidence of marrow infiltration, should be regarded as IgM-related disorders.
Median survival of patients with WM ranges between 5 and 10 years. Therapy is indicated for patients presenting with symptoms and signs due to malignant infiltration of organs or tissues or due to circulating or deposited IgM. Age, hemoglobin concentration, serum albumin and ?2-microglobulin have all been identified as significant prognostic factors while IgM levels have no prognostic value. Consensus criteria have been proposed for the evaluation of response to treatment and have been updated recently. Oral alkylating agents such as chlorambucil, had been the standard of care for many years. Nucleoside analogues (fludarabine, cladribine) have been effective in patients who failed primary treatment as well as in newly diagnosed patients. Combination chemotherapy has also been used. Rituximab induced responses in both previously treated and untreated patients and has been used in combination with dexamethasone, cyclophosphamide or nucleoside analogues resulting in high response rates and long remissions. Bortezomib is also active in both pretreated and untreated patients. High dose therapy with autologous stem cell transplantation and reduced-intensity allogeneic transplantation may have a role in the management of selected patients.

Symposium (Wed, 26 Sep, 14:45−16:45)
Molecular biology in paediatric tumours

180 INVITED

Paediatric brain tumours: exploiting genomics to improve therapies

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Background: Brain tumours represent the most common cause of cancer-related death in childhood. Current therapies fail to cure a significant proportion of cases, and are associated with long-term sequelae. Moreover, disease-risk is difficult to predict based on current clinical and histopathological criteria. Major challenges therefore exist in the individualisation and optimisation of current therapies, alongside a clear need for the development of novel therapeutic approaches. Focussing on medulloblastoma, the most common malignant brain tumour of childhood, this lecture will review our current understanding of the molecular events that underlie medulloblastoma pathogenesis, and how this knowledge is being exploited for potential therapeutic benefit.
Results: Major insights to the genetic basis of medulloblastoma have emerged from rare familial cancer syndromes. The observation of medulloblastoma as a feature of Gorlin, Turcot and Li-Fraumeni syndromes has led to the demonstration of somatic mutations of the genes responsible for these syndromes (ie. the PTCH, APC and P53 tumour suppressor genes) in significant subsets of sporadic cases. Studies in the human disease and transgenic mouse models have since established critical roles in normal cerebellar development for the sonic hedgehog (SHH) and wnt/wingless (Wnt) cell signalling pathways, in which these genes reside, and for their aberrant activation in medulloblastoma. Using contemporary genomics approaches, we have recently identified SHH and Wnt pathway mRNA expression signatures which characterise distinct sub-groups of medulloblastomas, into which genetic mutations affecting the respective pathways cluster. Moreover, genetic mapping analysis revealed tumours within these sub-groups are further defined by unique patterns of chromosomal aberrations. Together, these data are enabling the development of a robust classification of medulloblastoma molecular sub-groups, and the identification of markers which are independently predictive of poor (eg. 17p loss, MYC amplification) and favourable (eg. Wnt pathway activation) prognosis. These markers offer utility for improved disease-risk stratification, and are currently under assessment for this purpose in Europe-wide clinical trials. Finally, our developing understanding of medulloblastoma biological pathways is facilitating the selection, pre-clinical and early clinical assessment of new generations of molecularly targeted agents (eg. SHH antagonists) in this disease.
Conclusions: Recent advances highlight the potential translational impact of a detailed characterisation of the biological basis of medulloblastoma. Advances in medulloblastoma provide a 'roadmap' for translational research strategies in other paediatric brain tumour types, with the overall goal of delivering an improved outlook for children with these diseases.

181 INVITED

International consensus for neuroblastoma molecular diagnostics: report from the international neuroblastoma risk grouping (INRG) biology committee

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Background: Neuroblastoma serves as the paradigm for utilizing tumour genomic data for patient prognosis and treatment allocation. However, there is no worldwide consensus on markers, methodology or data interpretation, inhibiting translational research efforts.
Methods: The Biology subcommittee of the INRG working group (International Neuroblastoam Risk Grouping) was charged with developing an international consensus on all aspects of neuroblastoma molecular genomic diagnostics, including future directions. Consensus was achieved at the September 2005 conference in Whistler, Canada.
Results: A common protocol for specimen acquisition, preparation and banking was approved that focuses on tight quality control, since samples will be used for patient care as well as preservation of high quality research reagents. The working group defined MYCN amplification as >4-fold MYCN signals compared to chromosome 2q reference-probe (FISH method preferred). Whereas MYCN remains the main genomic factor currently used for treatment stratification, the INRG working group has also identified 11q23 allelic status and ploidy as independent markers of survival in certain patient subgroups. Common data elements to be obtained by all groups include these markers as well as allelic status of chromosome band 1p36 and 17q23−25, which are also related to a high-risk phenotype. Pan/multi-genomic methodologies are preferable for collecting DNA copy number data. Thus, genetic characterization of neuroblastomas according to INRG guidelines will require at least 10^7 tumour cells, information regarding the tumour cell content, and certified reference laboratories with expertise in the genetic assays described.
Conclusions: Neuroblastoma treatment planning is closely related to tumour cell genomic features, and it is likely that a panel of DNA-based biomarkers will be used in future risk assignment algorithms. Consensus on methodology and interpretation of these increasingly complex assays is essential and depends on continuous cooperation amongst international cooperative groups as proposed in the INRG.

182 INVITED

Molecular biology of anaplastic large-cell lymphoma

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Anaplastic large-cell lymphoma (ALCL) accounts for approximately 10−15% of all non Hodgkin lymphomas of childhood. It is characterized by a typical morphological appearance and by a peculiar immunophenotype. The great majority of the cases express the chimeric NPM-ALK protein, originating from the t(2;5)(p23;q35).
The availability of monoclonal antibodies reacting against the ALK moiety of NPM-ALK (and other fusion proteins) has permitted the identification of