Melanoma

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Double isolated limb infusion with cytotoxic agents for recurrent and metastatic limb melanoma

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Metastatic melanoma confined to a limb can be treated effectively using an isolated limb perfusion technique (ILP). A simplified alternative method called 'isolated limb infusion' (ILI) has been developed at the Sydney Melanoma Unit (Australia). This method is essentially a very low flow ILP performed via percutaneously introduced catheters without oxygenation of the limb. Previously we have demonstrated that with ILI similar response rates can been achieved as with ILP using melphalan and actinomycin-D as the cytotoxic drugs. The present study was undertaken to determine if electively performing two ILI procedures increases the frequency and/or duration of responses. Short and long-term results were compared with those for patients who underwent a single ILI in the same period of time. Moreover, the value of a second ILI when progression occurred after a first ILI was assessed.

Between 1992 and 1998, a total of 47 patients underwent a planned double ILI and 14 were treated with a second ILI after a good initial response after ILI. After double ILI 76% of patients experienced Wieberdink Grade III or IV toxicity in the treated limb compared to 52% after a single ILI (p=0.02). Overall response (OR) after the planned double ILI was 88% (complete response (CR) 41%, partial response (PR) 47%, stable disease (SD) 12%, progressive disease (PD) 0%). A CR was demonstrated in 70% of patients who were treated with an interval of 3 weeks or less between the two ILIs, however, this was not statistically higher than in patients with longer infusion intervals (p=0.08). The median duration of response was 18 months (6-60), the median patient survival was 17 months. Response rates after double ILI were similar to those in 81 patients treated with a single ILI (CR 41%, PR 41%, SD 12%, PD 5%). Response duration and patient survival were not significantly different for the two groups of patients. Patients who underwent a second ILI because of progression following their first ILI (n=14) had an OR of 77% (CR 8%, PR69%, SD 23%), with a 5 months (4-11) duration of response.

Since elective double ILI does not increase efficacy but increases toxicity, single ILI is the more appropriate treatment for melanoma confined to the limb. However, a second ILI can be of value if disease recurs or progresses following a previous ILI.

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Safety profile of histamine dihydrochloride administered with interleukin-2 in patients with advanced metastatic malignant melanoma

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Histamine Dihydrochloride (HDC) is being investigated as an adjuvant to interleukin-2 (IL-2) therapy in metastatic melanoma. Safety data were collected for 391 patients (pts) in two multicenter studies of HDC (Ceplene) administered with IL-2 in pts with stage IV malignant melanoma (IL-2 + HDC: n=239; IL-2 only: n=152).

Methods: Pts received IL-2 (9 MIU/m2, bid, sc, days 1-2, weeks 1,3; and 2 MIU/m2, bid, sc, days 1-5, weeks 2,4) with or without histamine (1.0 mg, bid, sc, days 1-5, weeks 1-4) for 4 weeks of a 6-week cycle. All therapy was administered in an outpatient at home setting. Safety and toxicity were assessed according to NCI Common Toxicity Criteria in all pts who received at least one dose of study drug.

Results: Common, anticipated toxicities, including fever, chills, asthenia, nausea, vomiting, anorexia, pain, diarrhea, cough, dyspnea, rash, and injection site pain, were reported with similar frequency and intensity (grade 1-4) among the two treatment groups. The following toxicities were observed with greater frequency in the IL-2/HDC group: vasodilation (94% vs. 36% of pts experiencing at least one episode), headache (59% vs. 35%), hypotension (51% vs. 20%), injection site inflammation (49% vs. 22%), injection site reaction (48% vs. 23%), rhinitis (38% vs. 25%), and dizziness (36% vs. 23%). In addition, clinically significant grade 3 and 4 toxicities occurred with similar frequency among treatment groups, with the lone exception being grade 3 headache, observed in 8% of patients receiving IL-2/HDC vs. 2% in the IL-2 group. Dose reduction (14% vs 14%), interruption (28% vs. 29%), and discontinuation (11% vs. 11%) of study drug administration were comparable among treatment groups. The rate of on-study death, which includes deaths due to progressive disease and deaths occurring within 28 days of final study drug administration, was 11% (27/239) in the IL-2/HDC group, compared to 16% (25/152) in the IL-2-alone group.

Conclusions: This review of IL-2/HDC safety and toxicity data suggests that HDC may be safely administered in conjunction with a regimen of low-dose sc IL-2, and adds low clinically significant grade 3 or 4 toxicity.

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A novel mutation of the mismatch repair gene hMLH1 in a human primary skin melanoma

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Purpose: Cells with a defective mismatch repair system (MRS) display high rates of spontaneous mutations, microsatellite instability and resistance to O6-guanine methylating agents and some other drugs. We identified a human melanoma cell line (PR-Mel) expressing almost undetectable levels of the DNA repair enzyme O6-alkylguanine-DNA alkyltransferase and still highly resistant to the cytotoxic activity of the methylating agent temozolomide. We therefore investigated whether PR-Mel tolerance to temozolomide was associated with a defective MRS.

Methods and Results: PR-Mel cell line was analyzed for mismatch repair activity, expression of MRS proteins and microsatellite instability at 12 different loci. The PR-Mel cell line was completely devoided of mismatch repair activity, did not express the hMLH1 and PMS2 proteins, and possessed high levels of microsatellite instability. We evaluated whether in PR-Mel cell line was present an aberrant hMLH1 promoter methylation or whether this line harbored mutations in hMLH1. No promoter methylation was found. Sequence analysis of all exons of hMLH1 revealed the presence of a G to A transition at position ñ1 of the acceptor splice site of intron 15. The mutation, named 1732-1G->A, alters the correct splicing of hMLH1 pre-mRNA leading to the in-frame skipping of exon 16, as attested by RT-PCR analysis. The somatic mutation 1732-1G->A was also detected in genomic DNA from both the primary skin melanoma and the cutaneous metastasis from which PR-Mel cell line had been established. Immunohistochemistry demonstrated no expression of hMLH1 and PMS2 in the tumor specimens. Both the cell line and biopsies appeared homozygous for the mutation, suggesting that the normal allele was lost. Indeed, cytogenetic analysis of PR-Mel cells revealed a 3p deletion possibly including the hMLH1 gene.

Conclusions: A cluster of hMLH1 mutations has been described in the region encompassing exons 15 and 16. However, mutation 1732-1G->A has not been previously described. Our data therefore expand the repertoire of hMLH1 mutations. To our knowledge, this is the first study in which a mutated MRS gene has been identified in a human melanoma.

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Treatment of chemoresistant metastatic malignant melanoma (MMM) with cationic colloidal BCL-2 antisense ODN (SEVINA-22) and vinorelbine-tartrate induces apoptosis via caspase-3 (CPP32) pathway

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Purpose: Metastatic malignant melanoma (MMM) are highly resistant to conventional cancer treatment including radiation and chemotherapy due to overexpression of bcl-2 oncogene which occurs in over 90% of all melanoma cases and acts as a negative regulator of apoptosis. We aim to induce apoptosis in chemoresistant MMM by combined chemogene treatment consisting of bcl-2 antisense ODN and vinorelbine.

Methods: We obtained melanoma cells by FNA of metastatic lesions in a patient with MMM disease. IHC analysis exhibited bcl-2 overexpression. RT-PCR amplification for bcl-2 mRNA was performed and positive results were obtained. An 18 mer phosphorothioated antisense oligodeoxynucleotide (ODN) containing unmethylated CG-dinucleotides (CpG motifs) against the bcl-2 messenger RNA was entrapped inside pegylated liposomes composed of DOTAP, DOGS, DDAP and DOPE. This colloidal bcl-2 antisense