ORIGINAL ARTICLE

Predicting Disease Progression After Regional Therapy for In-Transit Melanoma

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Importance: Although approximately 30% to 50% of patients experience a complete response after regional chemotherapy for in-transit melanoma, a subset of patients will develop rapidly progressive disease. In the current era of an expanding armamentarium, including both regional and systemic options for treating advanced melanoma, identifying perioperative factors that predict disease progression may obviate unnecessary morbidity associated with regional therapy and avoid delays in systemic therapy.

Objective: To identify patient-related clinical and pathological variables, as well as procedural factors, that correlate with disease progression.

Design: Using a prospectively maintained database, we identified patients who either underwent first-time melphalan-based isolated limb infusion (ILI) or first-time hyperthermic isolated limb perfusion (HILP) for in-transit melanoma. Response was defined using modified Response Evaluation Criteria in Solid Tumors for cutaneous disease at 3 months after treatment. Survival analyses were performed using the Kaplan-Meier method, with the differences in survival curves compared using a log-rank test. Potential preoperative and procedural predictors of in-field progressive disease were analyzed using logistic regression.

Participants: Of the 258 patients included in the database, 215 were identified as having undergone first-time regional therapy. Of these 215 patients, 134 underwent ILI, and 81 underwent HILP.

Exposure: Regional therapy (ILI or HILP).

Main Outcomes and Measures: Complete response or progressive disease.

Results: Of 134 patients who underwent ILI, 43 (32.1%) experienced in-field progressive disease. Of 81 patients who underwent HILP, 9 (11.1%) experienced in-field progressive disease. The median survival for patients with in-field progressive disease was 20.3 months for the ILI cohort and 15.0 months for the HILP cohort. In general, patients with progressive disease were younger, with advanced-stage melanoma and increased tumor burden. Compared with patients who experienced a complete response, patients with in-field progressive disease after ILI were younger (odds ratio, 1.06 [95% CI, 0.90-0.98]; $P=.002$). For patients who underwent HILP, no clinically relevant preoperative predictors of in-field progressive disease were identified. Procedural variables, including chemotherapeutic dosing, degree of acidosis or base deficit achieved, and peak temperature attained, were not predictors of in-field progressive disease after ILI or HILP.

Conclusions and Relevance: Patient, clinical, and procedural factors are unreliable predictors of in-field progressive disease after regional therapy in patients with in-transit melanoma. Defining the potential utility of molecular markers in predicting response or failure of regional therapy should be the focus of future research efforts.

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THE INCIDENCE OF MELANOMA is increasing faster than that of any other malignant disease in the United States, with more than 70 000 new cases expected in 2012.[1] Of these cases, 50% will represent disease limited to an extremity, and even after appropriate therapy, up to 10% of these cases will develop into subdermal lymphatic metastases occurring between the primary tumor and its draining lymph node basin.[2,3] This pattern of advanced disease of the extremities represents in-transit melanoma and is classified by the American Joint Committee on Cancer as either stage IIIB or stage IIIC, depending on regional lymph node involvement.[4]

Patients presenting with unresectable in-transit melanoma are often treated with regional therapy in the form of hyperthermic isolated limb perfusion (HILP) and isolated limb infusion (ILI), both of which allow for the circulation of chemotherapy at doses much higher than can be

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administered systemically. Although the complete response rate to regional therapy at our institution is approximately 55%, up to one-third of patients will develop a rapidly progressive disease.[5] In the current era of an expanding armamentarium, including both regional and sys-

Table 1. Response of Patients Who Underwent Regional Therapy in the Form of First-Time ILI or First-Time HILP

Type of Response	Patients, No. (%) ILI (n = 134)	HILP (n = 81)
Complete response	40 (29.9)	36 (44.4)
Partial response	17 (12.7)	21 (25.9)
Stable disease	16 (11.9)	7 (8.6)
Progressive disease	43 (32.1)	9 (11.1)
Not evaluated	18 (13.4)	8 (9.9)

Abbreviations: HILP, hyperthermic isolated limb perfusions; ILI, isolated limb infusions.

temic options for treating advanced melanoma, it is important to identify patients likely to experience progression so as to obviate the potential morbidity associated with regional therapy and, perhaps more importantly, to avoid delays in systemic therapy. Therefore, the purpose of our study was to identify patient-related clinical and pathological variables, as well as procedural factors, that correlate with disease progression.

METHODS

With institutional review board approval, our prospectively maintained regional therapy database, including records from 258 patients treated between 1995 and 2010, was reviewed. All patients were classified as having stage IIIB or IIIC and stage IV cancers based on the American Joint Committee on Cancer classification.[4] Response to therapy was evaluated 3 months after treatment as defined by the modified Response Evaluation Criteria in Solid Tumors for cutaneous lesions.[6] Only patients undergoing first-time regional therapy were included for analysis, focusing on in-field disease, defined as lesions distal to tourniquet placement. Toxicity was evaluated according to Common Terminology Criteria for Adverse Events version 3.[7]

REGIONAL CHEMOTHERAPY PROCEDURES

Isolated limb infusions were performed as previously described by our group.[8] In brief, after placement of percutaneous catheters by interventional radiology into the contralateral limb such that the catheter tips terminate in the middle of the diseased extremity, chemotherapy (using melphalan with or without dactinomycin) was rapidly infused into the arterial portion of the circuit and circulated for 30 minutes after the extremity was warmed to 37.0°C. Melphalan was dosed at 7.5 mg/L for the lower extremity and at 10 mg/L for the upper extremity; dactinomycin was dosed at 75 and 100 μg/L for the lower and upper extremities, respectively.[9] After the 30-minute circulation of chemotherapy, 0.5 to 1 L of isotonic crystalloid solution was flushed through the circuit for manual washout. Limb volume was calculated for both ILI and HILP by integrating the measured extremity circumference at 1.5-cm intervals up to the level of anticipated tourniquet placement, and chemotherapy dosing was typically corrected for ideal body weight (IBW) based on evidence that such modification reduces severe toxicity rates without altering response rates.[10] Of the 134 patients undergoing ILI, 117 (87.3%) received melphalan dosing based on IBW.

Hyperthermic isolated limb perfusions were performed as previously described.[11] In brief, the vessels to be cannulated were surgically isolated and then subsequently connected to a cardiopulmonary bypass circuit. A concurrent lymphadenectomy was often performed prior to vessel cannulation depending on the clinical scenario. Once extremity temperatures reached 38.5°C, chemotherapy was perfused through the circuit for 60 minutes, followed by a 15-minute washout with isotonic crystalloid solution. The HILP was performed using melphalan (10 mg/L for the lower extremity and 13 mg/L for the upper extremity). Of the 81 patients undergoing HILP, 22 (27.2%) received melphalan dosing based on IBW.

The concentration of circulating melphalan was obtained for a small number of patients. Concentrations were determined in the extremity circuit beginning at time zero and at 5-minute intervals for 30 minutes. The systemic melphalan concentration was also determined after 30 minutes of regional therapy to evaluate for tourniquet leak.

METHODS OF SURVEILLANCE

Rapidly progressive disease was defined as a 20% increase in the size of a preexisting lesion or the occurrence of a new lesion either in-field or out-of-field within 12 weeks of regional therapy.[6] For the purposes of statistical clarity, only patients with in-field progression were included for analysis. Both a physical examination to evaluate suspicious lesions and palpable lymphadenopathy and imaging were used to survey for response. After the first 3-month evaluation, patients were initially followed up every 3 months for 1 year and then every 6 months thereafter to determine progression-free survival.

Positron emission tomography/computed tomography was used as a routine surveillance tool to assess disease status prior to therapy and to detect local and systemic progression beginning approximately in 2004. Pathological confirmation via punch biopsies, fine-needle aspirations, computed tomography–guided biopsies, or surgical resections was performed when necessary. In the absence of a physical examination or positron emission tomographic/computed tomographic findings, we also began to routinely perform sentinel lymph node biopsies on patients with in-transit disease, mapping the in-transit disease starting in 2010.

STATISTICAL ANALYSIS

For the purposes of identifying predictors of rapidly progressive disease, these patients were compared with those experiencing a complete response. Survival analyses were performed using the Kaplan-Meier method, with the differences in the survival curves compared using a log-rank test.[12] Potential preoperative and procedural predictors of in-field progressive disease were analyzed in a univariate fashion, with statistical significance set at $P<.05$. All statistical analyses were performed using the commercially available software Stata version 11.1 (StataCorp).

RESULTS

Of the 258 patients included in the database, 215 were identified as having undergone first-time regional therapy. Of these 215 patients, 134 underwent regional therapy in the form of ILI, and 81 underwent regional therapy in the form of HILP. Response to regional therapy defined 3 months after treatment is included in **Table 1**. For the purposes of our study, 43 patients (32.1%) in the ILI cohort had progressive disease and were compared with 40 patients (29.9%) who experienced a complete response in the same cohort. Similarly, 9 patients (11.1%) in the HILP cohort progressed within 3 months and were

Table 2. Clinical, Pathological, and Procedural Variables for Patients Who Underwent ILI

Variable	Patients Who Underwent ILI, No. (%)		*P* Value
	CR (n = 40)	PD (n = 43)	
Age, median (range), y	70 (63-78)	60 (50-69)	<.001
Male sex	17 (42.5)	19 (44.2)	.88
Lower limb melanoma	36 (90.0)	36 (83.7)	.40
AJCC stage			
IIIB	22 (55.0)	19 (44.2)	.44
IIIC	15 (37.5)	22 (51.2)	
IV	3 (7.5)	2 (4.7)	
Disease burden			
Low	23 (57.5)	22 (51.2)	.40
High	15 (37.5)	21 (48.8)	
Unknown	2 (5.0)	0 (0.0)	
Melphalan dose, median (range), mg/L	43.2 (33.6-54.6)	48.5 (41-63.5)	.94
BE at 30 min, median (range), mEq/L	−9.3 (−11.8 to −8.0)	−9.4 (−12.2 to −8.0)	.58
pH at 30 min, median (range)	7.14 (7.09-7.205)	7.14 (7.08-7.19)	.40
Peak temperature, median (range), °C	38.7 (38.3-39.2)	38.4 (37.9-39.1)	.11
Toxicity[a]	5 (12.5)	11 (25.6)	.16

Abbreviations: AJCC, American Joint Committee on Cancer; BE, base excess; CR, complete response; ILI, isolated limb infusion; PD, progressive disease.
[a] Evaluated according to Common Terminology Criteria for Adverse Events version 3.[7]

Table 3. Clinical, Pathological, and Procedural Variables for Patients Who Underwent HILP

Variable	Patients Who Underwent HILP, No. (%)		*P* Value
	CR (n = 36)	PD (n = 9)	
Age, median (range), y	58 (48-65)	56 (54-57)	.67
Male sex	14 (38.9)	3 (33.3)	.76
Lower limb melanoma	6 (16.7)	8 (88.9)	.69
AJCC stage			
IIIB	13 (36.1)	3 (33.3)	.69
IIIC	19 (52.8)	6 (66.7)	
IV	4 (11.1)	0 (0.0)	
Disease burden			
Low	12 (33.3)	2 (22.2)	.21
High	5 (13.9)	3 (33.3)	
Unknown	19 (52.8)	4 (44.4)	
Melphalan dose, median (range), mg/L	110 (100-130)	94.2 (72.1-115.0)	.12
BE at 30 min, median (range), mEq/L	Not observed	Not observed	
pH at 30 min, median (range)	Not observed	Not observed	
Peak temperature, median (range), °C	40.3	40.0	
Toxicity[a]	9 (25.0)	5 (55.6)	.08

Abbreviations: AJCC, American Joint Committee on Cancer; BE, base excess; CR, complete response; HILP, hyperthermic isolated limb perfusion; PD, progressive disease.
[a] Evaluated according to Common Terminology Criteria for Adverse Events version 3.[7]

Table 4. Univariate Analysis of Perioperative Predictors of Progressive Disease After Isolated Limb Infusion

Predictor	OR (95% CI)	*P* Value
Age	0.94 (0.90-0.98)	.002
Male sex	1.08 (0.45-2.55)	.88
Lower limb melanoma	0.57 (0.15-2.12)	.40
AJCC stage		
IIIB	1 [Reference]	1 [Reference]
IIIC	1.70 (0.69-4.17)	.25
IV	0.77 (0.12-5.18)	.79
Disease burden		
Low	1 [Reference]	1 [Reference]
High	1.46 (0.60-3.54)	.40
Melphalan dose	1.02 (0.99-1.05)	.11
BE at 30 min	1.01 (0.89-1.15)	.84
pH at 30 min	0.50 (0.003-85.70)	.79
Peak temperature	0.69 (0.38-1.24)	.21
Toxicity[a]	2.27 (0.71-7.26)	.17

Abbreviations: AJCC, American Joint Committee on Cancer; BE, base excess; OR, odds ratio.
[a] Evaluated according to Common Terminology Criteria for Adverse Events version 3.[7]

compared with 36 patients (44.4%) who experienced a complete response to HILP. Patients found to have stable disease or a partial response to regional therapy were not included in this analysis.

Clinical and pathological variables, as well as procedural factors, were evaluated to identify differences between patients with progressive disease and patients who experienced a complete response, in both the ILI (**Table 2**) and HILP (**Table 3**) cohorts. In the ILI cohort, the only variable differing between patients with progressive disease and patients who experienced a complete response was age, with the patients who experienced progressive disease being younger (60 years vs 70 years; $P < .001$). The stage of disease, disease burden, and melphalan dosing were similar between groups, and although not statistically significant, the proportion of patients with progressive disease who had melphalan toxicity was twice that of patients who experienced a complete response (25.6% vs 13.2%; $P = .16$) in the ILI cohort. There were no differences in perioperative variables between patients who experienced a complete response and patients with progressive disease after HILP. In addition, the trend in toxicity was similar in the HILP cohort, with twice the proportion of patients with progressive disease who had melphalan toxicity compared with patients who experienced a complete response (55.6% vs 25.0%). More patients who underwent ILI (117 of 134 [87.3%]) had a melphalan dose corrected for IBW compared with patients who underwent HILP (22 of 81 [27.2%]). Logistic regression models were used to identify potential predictors of progressive disease after ILI (**Table 4**) and after HILP (**Table 5**). Of the variables tested on univariate analysis, only younger age (odds ra-

Table 5. Univariate Analysis of Perioperative Predictors of Progressive Disease After Hyperthermic Isolated Limb Perfusion

Predictor	OR (95% CI)	*P* Value
Age	1.01 (0.95-1.08)	.65
Male sex	0.79 (0.17-3.66)	.76
Lower limb melanoma	1.60 (0.17-15.30)	.68
AJCC stage		
IIIB	1 [Reference]	1 [Reference]
IIIC	1.37 (0.29-6.48)	.69
IV	Not observed	Not observed
Disease burden		
Low	1 [Reference]	1 [Reference]
High	3.60 (0.45-28.60)	.23
Melphalan dose	0.98 (0.96-1.01)	.24

Abbreviations: AJCC, American Joint Committee on Cancer; OR, odds ratio.

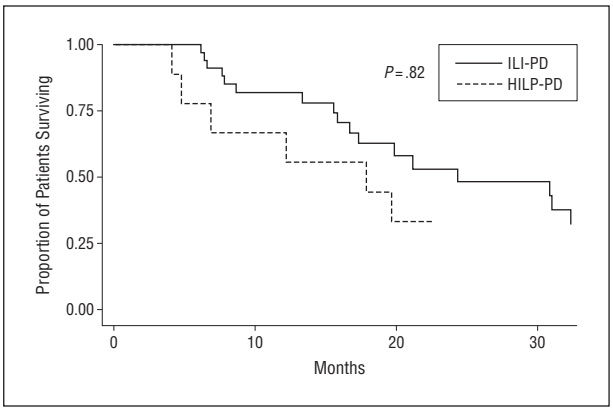

Figure 1. Overall survival rate of patients with progressive disease after undergoing either isolated limb infusion (ILI-PD) or hyperthermic isolated limb perfusion (HILP-PD), determined using the Kaplan-Meier method.

tio, 1.06 [95% CI, 0.90-0.98]; *P* = .002) was associated with progressive disease 3 months after ILI. Other clinical, pathological, and procedural variables did not correlate with progressive disease, including disease stage and disease burden, technical variables such as melphalan dosing and peak temperature attained, and the development of drug-induced toxicity. Similarly, there were no significant perioperative variables associated with progressive disease after HILP, including disease stage, disease burden, and procedural factors. Multivariate analyses were not performed owing to the low effective sample size. To evaluate survival specific to in-field progressive disease after ILI or HILP, Kaplan-Meier curves were created (**Figure 1**). The median survival for patients with in-field progressive disease was 20.3 months after ILI compared with 15.0 months after HILP (*P* = .82).

Melphalan pharmacokinetic data, including the circulating drug concentration within the extremity, were obtained for 17 patients who experienced a complete response and 19 patients with progressive disease. The average peak melphalan concentration after regional therapy was 23.63 μg/mL in patients who experienced a complete response and 26.24 μg/mL in patients with progressive disease. **Figure 2**A demonstrates the range of peak melphalan concentrations reached in patients who experienced a complete response and in patients with progressive disease, with the averages already mentioned represented by the horizontal bars. In addition, the average circulating melphalan concentration was determined at 5-minute intervals for 30 minutes in both patient populations (Figure 2B). The area under the curve was determined to be 416.90 and 465.50 μg · min/mL for patients who completely responded after regional therapy and for patients who progressed after regional therapy, respectively.

COMMENT

Advanced melanoma of the extremities develops in up to 10% of patients with disease limited to an extremity.[2,3] With a diagnosis of in-transit melanoma, patients may progress through an algorithm that includes treatment strategies such as surgical resection, intralesional therapy, regional chemotherapy, or systemic chemotherapy.[13] Regional therapy including ILI and HILP has become the mainstay of therapy for treating in-transit disease at our institution. Although complete response rates approach 55% after these procedures, a significant proportion of patients will develop a rapidly progressive disease.[5] Therefore, the objective of our study was to identify predictors of progressive disease, so as to identify patients likely to progress in such a way that alternative strategies can be prioritized. In our review of 215 patients who have undergone a first-time ILI or HILP, no patient-related clinical or pathological variables or technical factors proved to be a significant predictor of progressive disease. Although the rate of 5-year survival after resection of early-stage melanoma approaches 91%, systemic chemotherapy for advanced melanoma has historically been generally ineffective, with an overall response rate of approximately 20% and a complete response rate approaching only 5%.[14-16] Newer agents, such as the *BRAF*-specific inhibitor vemurafenib, benefit only 50% to 60% of patients with melanomas harboring the *BRAF* mutation and result in a 6-month survival rate of 84%, but median progression-free survival is only 5.3 months.[17] The use of ipilimumab improved the survival of patients with metastatic melanoma, but its side effect profile may not be tolerated by many patients.[18]

The current body of literature includes a number of studies reporting single-center experiences with ILI and HILP. The less invasive regional therapy technique, ILI, has been reported to elicit a complete response in approximately one-third of patients.[19,20] The more aggressive therapy, HILP, which requires the surgical isolation of the vessels to be cannulated with higher doses of chemotherapy circulated throughout the extremity, results in complete response rates between 40% and 80%.[8,21-26] The complete response rates in our cohort are similar to these other single-center studies, with complete responses in 30% of patients undergoing an ILI and in 44% of patients receiving an HILP. However, in our study, the rate of progressive disease, defined by the modified Response Evaluation Criteria in Solid Tumors[6] as an increase in size of a preexisting lesion by 20% or the appearance of a new lesion within 12 weeks of regional

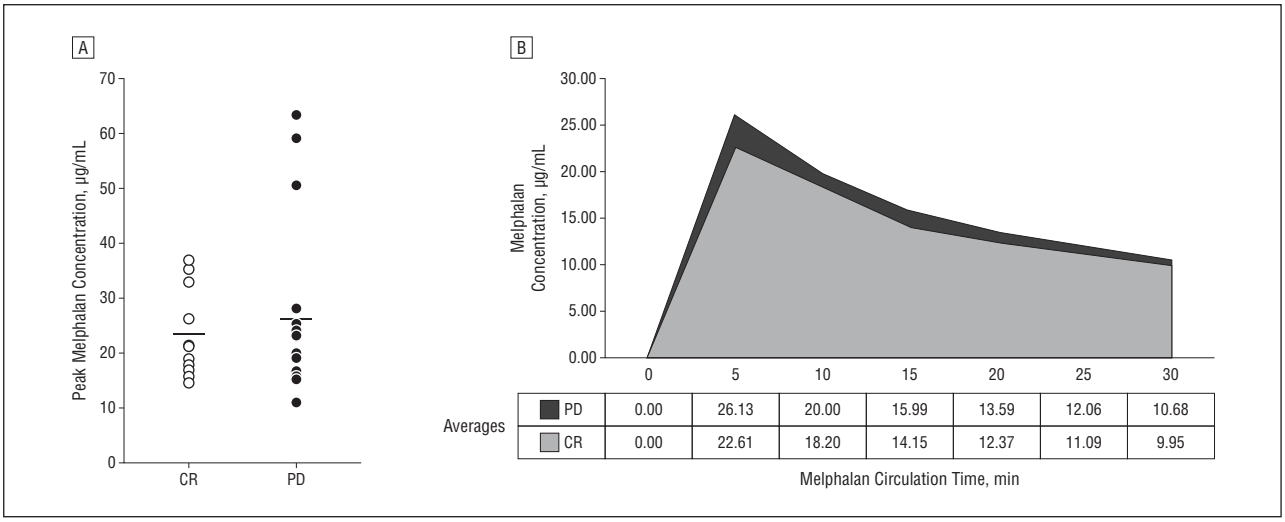

Figure 2. Pharmacokinetics of melphalan, which does not correlate with response to regional therapy. A, Dot plot comparing the peak melphalan concentration in patients exhibiting a complete response (CR) or progressive disease (PD) after regional therapy for in-transit melanoma. B, Circulating melphalan concentration within the extremity determined at 5-minute intervals for 30 minutes during regional therapy for in-transit melanoma. These curves represent the melphalan concentration at each time point for 17 patients with a CR and for 19 patients with PD, with the averages shown at the bottom.

therapy, was 32% for the ILI cohort and 11% for HILP cohort. Of note, correcting the melphalan dose for IBW in ILI is associated with similar complete response rates but lower partial response rates; correcting the dose for IBW also reduces toxicity.[19] Whether the same is true in HILP is currently unknown. In our study, more patients who had undergone an ILI did have their doses corrected for IBW compared with those who had undergone an HILP, which may explain the higher progressive disease rate in the ILI cohort. Furthermore, the peak concentration of circulating melphalan during regional therapy does not appear to correlate with progressive disease because the small number of patients in our study with available pharmacokinetic data had similar peak drug concentrations, regardless of their response.[10] The area under the curve, used as a surrogate for circulating melphalan concentration over a 30-minute period, was also similar between patients who experienced a complete response and patients with progressive disease, and therefore it did not predict response. Ultimately, the clinical meaningfulness of a patient who experienced a partial response is similar to that of a patient with stable disease or a patient with progressive disease in that the patient still requires additional interventions to achieve a NED (no evidence of disease) status.

The median survival for patients with progressive disease is 20.3 months after an ILI and 15.0 months after an HILP, whereas the median survival after a complete response may be as high as 100 months.[20,27-29] The ability to identify patients likely to experience progressive disease despite optimal regional therapy is necessary, so as to avoid delays in more appropriate therapy, such as systemic chemotherapy. Furthermore, the identification of other modifiable factors not investigated in our study, such as the role of papaverine hydrochloride to improve drug delivery to cutaneous vessels, or the identification of patients with favorable tumor biology, may yield the potential to further optimize treatment and individualize care, ultimately resulting in improved outcomes for this challenging patient population.[29,30]

Thus, regional therapy remains an important option for these patients. Interestingly, there is currently an ongoing trial from Memorial Sloan Kettering Cancer Center in New York, New York, investigating how augmentation of the immune response with ipilimumab in the context of regional therapy can improve outcomes and response rates (NCT01323517). If immunologic manipulation does indeed augment regional therapy, correlative scientific studies evaluating immunologic profiles of responders and nonresponders may prove to be more sensitive predictors of response than the patient- and treatment-related variables explored in our study. Rational combination therapies for individual patients, including immunologic therapy, targeted agents, and cytotoxic chemotherapy, may ultimately provide the most effective strategy.

The present study has several limitations. It focused only on in-field progressive disease for the purposes of statistical clarity; however, there does exist a population of patients with out-of-field progressive disease. Furthermore, by only comparing patients who experienced a complete response with those with progressive disease, in a retrospective fashion, our study may be underpowered; therefore, the variables tested could be potential predictors of progressive disease if analyzed within a larger series of patients. In addition, approximately 12% of patients were lost to follow-up and were therefore not evaluated or included in the analysis; data from these patients would have increased the power of our study.

Despite these limitations, we conclude from our study that patient-related clinical and pathological variables, as well as procedural factors, cannot reliably predict progressive disease after regional therapy for advanced melanoma of the extremities. Until more sensitive predictors of progressive disease are developed, we will continue to recommend regional therapy for patients with in-transit disease confined to a single extremity. Further research in the realm of molecular markers in the context of tumor signatures, as well as research to determine the

potential benefit of immunologic augmentation, may be the key to identifying patients most likely to benefit from regional therapy and therefore improve their survival and eliminate unnecessary morbidity.

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