Performance of a prognostic 31-gene expression profile test in Stage III cutaneous melanoma subjects

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Background

- The management of Stage III cutaneous melanoma (CM) patients has changed with the introduction of new, effective adjuvant therapies.[1,2]
- However, given the heterogeneity of distant metastasis risk and the toxicity associated with anti-CTLA-4 therapy, the identification of those patients who will benefit from therapy becomes paramount, particularly for Stage IIIA patients.
- A 31-gene expression profile (GEP) test that provides a prediction of low risk (Class 1) or high risk (Class 2) of melanoma metastasis has been validated as an independent prognosticator of distant metastasis-free (DMFS) and melanoma-specific survival (MSS) in stage I-II patients.[3,4]
- Identification of which stage III patients are at higher risk of progression with the GEP test might be useful for decision-making when considering the risks and benefits of adjuvant therapies.

Objective

- Here we examine the prognostic accuracy of the GEP test in a cohort of Stage III cutaneous melanoma from a multi-center validation study.

Methods

- A total of 207 subjects with Stage III cutaneous melanoma were identified from 16 U.S. centers. Primary tumor samples and associated clinical data were collected under an IRB-approved, multicenter protocol.
- RT-PCR was performed to determine expression levels of 28 prognostic genes and 3 control genes, and Radial Basis Machine modeling was used to predict binary metastatic risk class for each case (Class 1 vs. Class 2).
- Distant metastasis-free survival (DMFS; time to any metastatic event beyond the regional nodal basin), melanoma-specific survival (MSS; time from diagnosis to death documented as specifically resulting from melanoma), and overall survival (OS; time from diagnosis to death from any cause) were assessed.

References

1. Eggermont AM, Chiarion-Sileni V, Grob JJ, et al. N Engl J Med 2016;375:1845-55.
2. Eggermont AM, Suciu S, Testori A, et al. J Clin Oncol 2012;30:3810-8.
3. Gerami P, Cook RW, Wilkinson J, et al. Clin Cancer Res 2015;21:175-83.
4. Gerami P, Cook RW, Russell MC, et al. J Am Acad Dermatol 2015;72:780-5 e783.

Acknowledgements

We wish to thank Drs. Laura Ferris, Brian Gastman, Alex Miller, Sancy Leachman, John Vetto, Stephen Lyle, Daniel Rosen, Jane Messina, Vernon Sondak, David Lawson, Maria Russell, Keith Delman, Lee Cranmer, Rene Gonzalez, T. Christopher Windham, David Pariser, and Sarah Estrada for their contribution of samples to the study.

Disclosures

Funding for the study provided by Castle Biosciences (CBI). JSZ and PG are paid consultants for CBI. MDF and JDW have received honoraria from CBI for advisory board participation. BM and KRC are employees and stock option holders of CBI.

Results

Table 1. Patient demographics

	Overall n=207	Class 1 n=63	Class 2 n=144	p value*
Median age (range), years	58 (18-94)	55 (18-83)	60 (22-94)	0.037
Regional and/or distant metastasis	119	26	93	0.002
Distant metastasis	95	19	76	0.003
AJCC stage				
IIIA	76 (37%)	34 (54%)	42 (29%)	0.003
IIIB	80 (39%)	18 (29%)	62 (43%)	
IIIC	45 (22%)	8 (13%)	37 (26%)	
Unknown substage	6 (2%)	3 (5%)	3 (2%)	
Breslow thickness				<0.001
Median (range), mm	2.5 (0.4-29.0)	1.5 (0.4-10.0)	3.3 (0.6-29.0)	
≤1 mm	20 (10%)	12 (19%)	8 (6%)	
>1 mm	184 (89%)	51 (81%)	133 (92%)	
Unknown	3 (1%)	0 (0%)	3 (2%)	
Mitotic index				0.029
<1 mm²	22 (11%)	11 (17%)	11 (8%)	
≥1 mm²	130 (63%)	35 (56%)	95 (66%)	
Unknown	55 (26%)	17 (27%)	38 (26%)	
Ulceration				<0.001
Absent	79 (38%)	36 (57%)	43 (30%)	
Present	105 (51%)	15 (24%)	90 (63%)	
Unknown	23 (11%)	12 (19%)	11 (7%)	
Primary tumor location				0.167
Head & neck	42 (20%)	8 (13%)	34 (24%)	
Trunk	56 (27%)	17 (27%)	39 (27%)	
Extremity	109 (53%)	38 (60%)	71 (49%)	

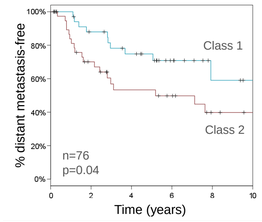
*p value indicates significance of differences between Class 1 and Class 2 groups

Table 2. Cox multivariate regression analysis in all Stage III cases

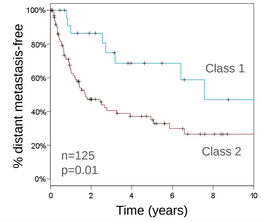
	DMFS		MSS		OS	
	HR (95% CI)	p value	HR (95% CI)	p value	HR (95% CI)	p value
Breslow thickness	1.3 (1.2-1.4)	0.0001	1.2 (1.0-1.4)	0.02	1.2 (1.1-1.3)	0.0006
Mitotic rate >1/mm²	0.7 (0.3-1.4)	0.3	0.4 (0.2-1.1)	0.08	0.5 (0.2-1.1)	0.08
Ulceration present	1.3 (0.8-2.2)	0.3	0.9 (0.4-2.0)	0.7	1.7 (0.9-3.1)	0.09
GEP Class 2	1.9 (1.0-3.5)	0.04	2.8 (0.9-8.2)	0.07	2.2 (1.0-4.8)	0.05

HR, hazard ratio; CI, confidence interval

Figure 1. Kaplan-Meier analysis of DMFS for GEP predicted outcomes in Stage IIIA (left) and Stage IIIB/C cases (right)

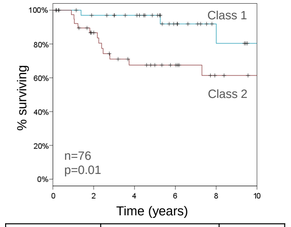


	5-yr DMFS (95% CI)	Events (%)
Class 1 (n=34)	75% (61-92%)	10 (30%)
Class 2 (n=42)	53% (39-74%)	19 (45%)

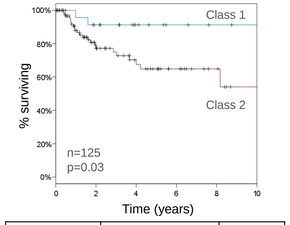


	5-yr DMFS (95% CI)	Events (%)
Class 1 (n=26)	69% (50-94%)	8 (31%)
Class 2 (n=99)	35% (26-48%)	55 (56%)

Figure 2. Kaplan-Meier analysis of MSS for GEP predicted outcomes in Stage IIIA (left) and Stage IIIB/C cases (right)

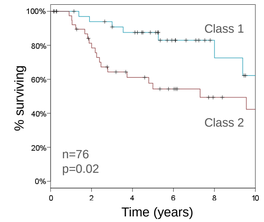


	5-yr MSS (95% CI)	Events (%)
Class 1 (n=34)	97% (91-100%)	3 (9%)
Class 2 (n=42)	68% (53-86%)	12 (29%)

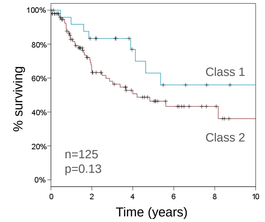


	5-yr MSS (95% CI)	Events (%)
Class 1 (n=26)	91% (80-100%)	2 (8%)
Class 2 (n=99)	65% (53-79%)	23 (23%)

Figure 3. Kaplan-Meier analysis of OS for GEP predicted outcomes in Stage IIIA (left) and Stage IIIB/C cases (right)



	5-yr OS (95% CI)	Events (%)
Class 1 (n=34)	88% (77-100%)	8 (24%)
Class 2 (n=42)	54% (40-74%)	20 (48%)



	5-yr OS (95% CI)	Events (%)
Class 1 (n=26)	63% (44-91%)	9 (35%)
Class 2 (n=99)	46% (35-61%)	40 (40%)

Results

Table 3. Distant metastasis events and rates in 125 patients who underwent completion lymph node dissection (CLND), according to GEP result (Class 1 or 2), non-sentinel lymph node (SLN) status, or combined.

	Distant metastasis events (rate)
GEP Class 1 (n=39)	11 (28%)
GEP Class 2 (n=86)	44 (51%)
Non-SLN neg (n=80)	30 (37%)
Non-SLN pos (n=45)	25 (55%)
Class 1/Non-SLN neg (n=31)	7 (22%)
Class 1/Non-SLN pos (n=8)	4 (50%)
Class 2/Non-SLN neg (n=49)	23 (47%)
Class 2/Non-SLN pos (n=37)	21 (58%)

Table 4. Accuracy metrics for identification of distant metastasis in a cohort of 125 patients who underwent CLND. Metrics are presented according to non-SLN status, GEP result, or combined. PPV=positive predictive value; NPV=negative predictive value.

	Non-SLN status	GEP Class	Combined
Sensitivity	45%	80%	87%
Specificity	71%	40%	34%
PPV	56%	51%	51%
NPV	63%	72%	77%

Conclusions

- The 31-gene GEP test is able to identify two significantly different groups of stage IIIA patients with low (Class 1) and high (Class 2) risk for distant metastasis and melanoma-specific death.
- GEP testing may be useful in identifying stage IIIA patients who would benefit from adjuvant therapies and/or enrollment in clinical trials.
- Results from a subset of patients undergoing CLND suggest that GEP testing could potentially guide use of the CLND procedure in SLN-positive patients.
- In patients who undergo CLND, sensitivity for distant metastatic disease improved to 87% when GEP testing was incorporated, compared to that of non-SLN status alone.