FINAL REPORT

Patient:
Sex:
DOB:
Client:
Clinician:

Tumor Site:
Specimen ID:
Collected:
Received:
Reported:

DecisionDx-Melanoma Result

Class 1

Sub-class: 1A

Class 1A signature is associated with the lowest risk of recurrence within 5 years

In patients 55 years and older with T1-T2 tumors, a Class 1A result is associated with low probability of a positive sentinel lymph node

The **DecisionDx®-Melanoma** molecular test for cutaneous melanoma is a proprietary gene expression assay that uses RT-PCR to determine the expression of a panel of 31 genes (3 control) in primary tumor tissue. The DecisionDx-Melanoma classification is derived from the gene expression results and comparison to a training set of patients with known outcomes.[1]

The DecisionDx-Melanoma algorithm generates a value between 0 and 1 with a crossover point of 0.5. Sub-classification (A or B) is based on the proximity of this value to the crossover point.

STAGE I AND II CLINICAL INFORMATION[1-5]

Approximately 67% of patients in the clinical validation studies were clinically and/or pathologically node-negative (AJCC Stage I and II). The survival rates for Class 1 and Class 2 signatures are shown below. The test is an independent predictor of risk of recurrence compared to traditional clinicopathologic factors. The median time to first recurrence in Stage I and II, Class 2 patients (n=206) is 1.55 years.

Molecular Signature Result		Recurrence-free Survival at 5 years (Stage I and II)	Distant Metastasis-free Survival (Stage I and II)	Melanoma-Specific Survival (Stage I and II)
Class 1	1A	95%	97%	99%
	1B	88%	90%	97%
Class 2	2A	77%	84%	97%
	2B	51%	65%	89%

n=608 Stage I and II patients (median follow-up time = 7.5 years); Log-rank (Mantel-Cox) test; p<0.0001 for all endpoints

See page 2 of this report for data pertaining to SLNB guidance and Stage III disease

Digitally signed by Sherri Borman, PhD, HCLD, Lab Director
Date: 2019.12.09 09:20:47 GMT-7'00'

Castle Biosciences, Inc. | Sherri Borman, PhD, HCLD, Lab Director

CAP ACCREDITED
COLLEGE of AMERICAN PATHOLOGISTS

LIKELIHOOD OF POSITIVE SENTINEL LYMPH NODE BIOPSY (SLNB) in T1-T2 TUMORS[6]

In a prospective, multicenter cohort of 1,421 patients, 1,065 of which had T1-T2 melanoma by AJCC (Breslow thickness ≤2.0 mm), NPV for SLN positivity was 0.95 for patients 55-64 years old, and 0.98 for patients ≥65 years old. Data by age grouping is shown below.[6]

Probability of a Positive Sentinel Lymph Node (SLN +)				
	<55 years	55-64 years	≥65 years	All Ages
Class 1A	7.6%	4.9%	1.6%	4.6%
Class 1B/2A	19.6%	7.7%	6.9%	10.8%
Class 2B	24.0%	30.8%	11.9%	18.8%

Clinical Outcomes in T1 and T2 patients with Class 1A results	
T stage	5-year MSS
T1 n=258	>99% (100-100%)
T2 n=88	98.6% (95.9-100%)

Clinical outcomes for T1-T2, Class 1A patients are derived from an archival, multicenter cohort of 901 Stage I-III patients.[1-5]

STAGE III CLINICAL EXPERIENCE[1-5]

Sub-classification (A/B) is not applied in the Stage III population

Molecular Signature Result	Distant Metastasis-Free Survival at 5 Years (Stage III)	Melanoma-Specific Survival at 5 Years (Stage III)
Class 1	72%	93%
Class 2	46%	66%
n=293 Stage III patients (median follow-up time = 7.5 years); Log-rank (Mantel-Cox) test; p<0.0001 for DMFS; p=0.0001 for MSS		

Approximately 33% of patients in the clinical validation studies were AJCC Stage III. The distant metastasis-free survival and melanoma-specific survival rates for Class 1 and Class 2 signatures for Stage III patients are shown above. The median time to first recurrence in Stage III, Class 2 patients (n=198) is 0.77 years.

ABOUT THE TEST

The test was developed using a training set of 164 samples and validated for risk of recurrence in 901 Stage I-III patients with > 5 years follow-up (median overall follow-up time = 7.5 years). This cohort is a normal-risk population by stage with 5-year melanoma specific survival rates similar to AJCC v8. Further, this test was validated for risk of SLNB positivity in a separate 1,421 Stage I-III patient cohort. Likelihood of SLN positivity in this group was similar to that of the SLNB eligible population overall.

DecisionDx-Melanoma cutaneous melanoma assay uses RT-PCR to determine the expression of a panel of 31 genes (3 control) in the supplied tumor tissue. The twenty-eight discriminating genes are: BAP1 (two gene loci), MGP, SPP1, CXCL14, CLCA2, S100A8, BTG1, SAP130, ARG1, KRT6B, GJA1, ID2, EIF1B, S100A9, CRABP2, KRT14, ROBO1, RBM23, TACSTD2, DSC1, SPRR1B, TRIM29, AQP3, TYRP1, PPL, LTA4H and CST6. The three control genes are: FXR1, YKT6 and HNRNPL. The predicted classification is reported as Class 1 for low risk, and Class 2 for high risk of metastasis.

The DecisionDx-Melanoma 31 gene assay is an independent predictor of metastatic risk in multivariate analyses including Breslow's thickness, ulceration, mitotic rate, age, SLN status and AJCC stage. The performance characteristics reported in the multi-center clinical validation studies[1,2] and the multi-center performance studies[3-5] are consistent across four prospective clinical studies.[7-10] Data in this report has not been validated in patients with clinical features different from those described above.

REFERENCE LIST

[1]Gerami P, et al. Clin Cancer Res 2015; 21(1);175-183; [2]Gerami P, et al. J Am Acad Dermatol 2015; 72:780-785.e3; [3]Zager J, et al. BMC Cancer 2018; 18:130; [4]Gastman B, et al. J Am Acad Dermatol 2019; 80(1): 149-157.e4; [5]Prado G, et al. Fall Clinical Derm NP/PA meeting abstract; 2019; [6]Vetto J, et al. Future Oncol 2019; 15(11):1207-1217; [7]Hsueh E, et al. J Hematol Oncol 2017; 10(152); [8]Greenhaw B, et al. Dermatol Surg 2018; 44(12):1494-1500. [9]Keller J, et al. Cancer Med 2019; 8(5):2205-2212. [10]Podlipnik S, et al. J Eur Acad Dermatol Venereol 2019; 33:857-8612