DermTech
CAP ACCREDITED

Nevome™ Pigmented Lesion Assay
MOLECULAR PATHOLOGY TEST REPORT

1.866.450.4223 | 1.858.450.4222 | dermtech.com
11099 N. Torrey Pines Rd., Suite 100 | La Jolla, CA 92037

PATIENT NAME

Patient Name:		Sample ID:	
MRN#:		Body Site:	
DOB:		Date Collected:	
Age:		Date Received:	
Sex:		Date Reported:	
Referring Physician:		Fax Number:	
Address:		City/State/Zip:	

TEST RESULTS

Gene Expression Status: POSITIVE

Key Driver Mutation Status: POSITIVE

Risk Status: SEVERE (RED+)

***LINC00518*:**	**DETECTED**
PRAME:	**DETECTED**
BRAF:	**DETECTED**
NRAS:	**DETECTED**
TERT Promoter:	**DETECTED**

Expression of LINC00518 and /or PRAME is found in lesions with a histopathologic diagnosis of melanoma. If one or both of the genes are detected the gene expression test is positive.

High Risk Driver mutations in BRAF, NRAS, and TERT promoter region are found in early cutaneous and metastatic melanoma. The presence of mutations in BRAF and NRAS genes or in TERT gene promoter is associated with in situ and invasive melanoma.

MINIMAL RISK (GREEN): No molecular risk factors identified. Consider surveillance per standard of care.

GUARDED RISK (ORANGE): Single gene expression molecular risk factor. Lower probability of melanoma. Recommend surgical biopsy.

ELEVATED RISK (ORANGE+): Single gene expression plus one mutation risk factor. Lower probability of melanoma. Recommend surgical biopsy.

HIGH RISK (RED): Double gene expression or double mutation risk factors. High probability of melanoma. Recommend surgical biopsy. Consider prognostic implications.

SEVERE RISK (RED+): Double gene expression plus one or more mutation risk factors. Highest probability of melanoma. Recommend surgical biopsy. Consider prognostic implications.

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TEST RESULTS

MUTATIONS DETECTED (CIRCLED)

BRAF: V600E; V600K; K601E; G469A

NRAS: G12C; G13D; Q61K; Q61R

TERT Promoter: -124G>A (C>T); -138G>A (C>T); -139G>A (C>T); -146G>A (C>T)

Prognostic Implications: The presence of two or more mutations involving BRAF/NRAS/TERT in melanoma is associated with increased tumor thickness, a higher mitotic rate, more advanced stage, and lower progression-free and melanoma specific survival. PRAME has independent prognostic implications on recurrence and progression free survival in a variety of cancers including uveal melanoma. The significance of PRAME expression in conjunction with mutations in BRAF/NRAS/TERT has not been elucidated. Lesions with multiple molecular risk factors should have careful histopathologic evaluation and clinical follow-up.

BRAF: BRAF is a member of the RAF kinase family of growth signal transduction protein kinases. This protein plays a role in regulating the MAP kinase/ERKs signaling pathway, which affects cell division and differentiation. Constitutively active BRAF mutants commonly contribute to oncogenesis by excessively signaling cells to grow. Somatic mutations in BRAF occur in 37-50% of malignant melanomas and are most common in melanomas found in skin without chronic sun damage. The most prevalent BRAF mutations in melanoma lead to an amino acid substitution of valine at the 600th amino acid in BRAF (i.e. BRAF V600). While BRAF V600E can be found in nevi, it is not a suitable discriminator between nevi and melanoma by itself. Other mutations leading to V600K, V600M, and K601E are almost exclusively found in melanoma, in less than 15% of cases. The result of these mutations is increased BRAF kinase activity. However, if BRAF V600E is combined with other mutations like in NRAS or the TERT promoter, consider prognostic implications.

NRAS: Mutations in RAS genes can lead to the production of permanently activated RAS proteins. As a result, this can cause unintended and overactive signaling inside the cell, even in the absence of incoming signals. Because these signals result in cell growth and division, overactive RAS signaling can ultimately lead to cancer. Mutations which changed amino acid residues 12, 13 or 61 activate the potential of NRAS to transform cultured cells and are implicated in a variety of human tumors, including melanoma. Somatic mutations in NRAS are found in 13-25% of malignant melanomas and may be more common in melanomas arising in chronic sun damaged skin. The most prevalent NRAS mutations occur at positions 12, 13 or 61 and result in activation of NRAS signaling pathways.

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TERT: TERT promoter mutations are common genetic mutations in cutaneous melanoma (found in about 43% of melanomas). The mutations are most often C>T(G>A) transitions, predominantly located either −124 base pairs (bp) or −146 bp upstream of the TERT translational start site (ATG). The mutations are associated with functional increases in TERT protein, telomerase activity, and telomere length, and are associated with cell immortalization and proliferation leading to oncogenesis. TERT promoter mutations in melanoma are associated with histopathologic features of aggressiveness and poor survival.

PRAME: PRAME (Preferentially Expressed Antigen in Melanoma) encodes protein that is preferentially expressed in human melanomas. The encoded protein acts as a repressor of retinoic acid receptor, and likely confers a growth advantage to cancer cells via this function. PRAME is a key cancer target in 2 molecular uveal and cutaneous melanoma tests designed to support pathologists.

LINC: LINC (Long Intergenic Non-Protein Coding RNA 518) is a key member of a rapidly growing family of regulatory RNA molecules that are not translated into proteins. LINC is overexpressed in cutaneous melanoma.

ASSAY DESCRIPTION AND INTENDED USE

The DermTech pigmented lesion assay is intended for use under the direction of a physician to provide information on gene expression risk factors for melanoma in pigmented skin lesions. The assay detects gene expression for LINC00518 (Long Intergenic Non-protein Coding RNA518), and/or PRAME (Preferentially Expressed Antigen in Melanoma). The test is intended for use on pigmented skin lesions suspicious for melanoma, including those that meet one or more ABCDE criteria, and for which a clinician would like additional information prior to surgical biopsy. If the pigmented lesion assay is positive for gene expression, a surgical biopsy for histologic assessment should follow. The assay is not intended for use on non-melanocytic lesions. The test has been validated in samples collected using the Adhesive Skin Sample Collection Kit, and used according to the Instructions For Use (IFU). Lesions should be at least 5mm in diameter. For lesions larger than 16mm, multiple kits should be used. For valid lesion assessment, a careful demarcation of the desired tissue to be analyzed should be provided on the sample collection adhesive patches. The test has not been validated for samples collected from mucosal surfaces, the palms of hands, the soles of feet, sites that have been previously biopsied, areas where hair cannot sufficiently be trimmed, bleeding or ulcerated lesions, pediatric patients, and patients with Fitzpatrick skin type IV or higher. Samples with blood or excessive amounts of non-vellus hair (hair other than 'peach fuzz') on the adhesive patches will not be processed. Clinically obvious or frank melanoma should be surgically biopsied, per standard guidelines. Non-melanoma skin cancers may be negative based on LINC00518 and/or PRAME gene expression. As with all tests, results should be interpreted by the physician in conjunction with clinical findings, histopathologic findings as available, and patient risk assessment.

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REFERENCE MATERIAL AND ASSAY PERFORMANCE

Expression of LINC00518 and/or PRAME genes has been studied in a validation set of 398 samples. Of the studied melanomas, 39% were read histopathologically as melanoma in situ or lentigo maligna, and the median thickness of invasive melanomas was 0.45 mm. Eighty-one percent of nevi were read as atypical nevi. In this study, 91% of melanomas demonstrated detectable levels of LINC00518 and/or PRAME expression versus only 31% of non-melanoma samples, giving a sensitivity of 91% and a specificity of 69%. In a recent review of adult PLA positive results, the frequency of histopathologically confirmed melanomas in double positive samples, in which both LINC and PRAME are detected, was higher than in in single gene positive samples (LINC only or PRAME only detected) (92% vs. 12%). These proportions are similar to the ones found in Gerami et al. (Journal of the American Academy of Dermatology, 2017). In an additional PLA validation study evaluating hot spot mutations in BRAF, NRAS, and the TERT promoter region in 103 histopathologically confirmed samples relative to PLA results, statistically significant differences were observed between PLA positive and histopathology positive (melanoma) lesions compared to PLA negative histopathology negative (non-melanoma) lesions (75% vs. 15%, $p<<0.0001$) (Data on File, DermTech). There were no statistical differences in mutation frequency between the validation cohort and a 523 real-world test result cohort. 89% of PLA negative results in the real-world test cohort were mutation negative. Acquired mutations in these genes are well characterized during early stage and metastatic melanoma (Shain et al., The New England Journal of Medicine, 2015).

At a calculated 7% melanoma prevalence, the PLA negative predictive value is greater than 99% indicating that, a patient with a negative test has a 1% probability of being positive for melanoma (Gerami et al., Journal of the American Academy of Dermatology, 2017). In a review of 280 PLA-negative test results, less than 2% were surgically biopsied at 3-6 months of clinical follow up and none of the biopsied lesions were found to be melanomas consistent with the high negative predictive value and low false negative rate. In a utility study by Ferris et al. (JAMA Dermatology, 2017), pigmented lesion experts biopsy about half as often and miss fewer melanomas when adding the pigmented lesion assay to their decision process, allowing more melanomas to be found with fewer surgical procedures. Additional studies using the PLA in clinical settings demonstrate that the PLA identifies early stage melanoma (melanoma in situ, Stage 1) and that the biopsy ratio declines from >7.5 with visual assessment, to less than 1.7 with the PLA (Data on File, DermTech).

This test was developed and its performance characteristics determined by DermTech. This laboratory is certified under the Clinical Laboratory Improvement Amendments (CLIA) as qualified to perform high complexity clinical laboratory testing. The DermTech CLIA laboratory is also accredited by the College of American Pathologists (CAP). This test has not been cleared or approved by the US Food and Drug Administration; FDA does not require this test to go through premarket FDA review. This test is used for clinical purposes and is not considered investigational or for research.

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LITERATURE

1. Haqq C et al., The gene expression signatures of melanoma progression. PNAS, 2005, 102 (17):6092-6097
2. Gerami P et al., Development and validation of a non-invasive 2-gene molecular assay for cutaneous melanoma. Journal of the American Academy of Dermatology, 2017; 76(1):114-120.e2
3. Ferris, L et al., Validation of non-invasive gene expression (PLA) against high risk driver mutations (BRAF, NRAS, and TERT) in cutaneous melanoma. Accepted late breaking presentation AAD, 2018, full manuscript submitted.
4. Ferris L et al., Utility of a noninvasive 2-gene molecular assay for cutaneous melanoma and effect on the decision to biopsy. JAMA Dermatology, 2017; 153(7):675-680
5. Ferris, L et al., Real-world performance and utility of a non-invasive gene expression assay to evaluate melanoma risk in pigmented lesions. Submitted, Melanoma Research, December 2017, under review.
6. Yao Z et al., Analytical characteristics of a noninvasive gene expression assay for pigmented skin lesions. Assay and Drug Development Technologies, 2016: 14(6); 355-363
7. Yao Z et al., An adhesive patch-based skin biopsy device for molecular diagnostics and skin microbiome studies. Journal of Drugs in Dermatology, 2017; 16(10): 611-618
8. Hornberger J and Siegel D. Clinical and economic implications of a noninvasive molecular pathology assay for early detection of melanoma. Abstract published Journal of Skin, Vol 1, Nov 2017, Full manuscript submitted JAMA Dermatology, December 2017.
9. Wachsman W et al., Noninvasive genomic detection of melanoma. British Journal of Dermatology, 2011; 164:797-806
10. Shain H et al. The genetic evolution of melanoma from precursor lesions. New England Journal of Medicine, 2015; 373:1926-1936.
11. Shain H and Bastian B. From melanocytes to melanomas. Nat Rev Cancer, 2016 Jun;16(6):345-58
12. The Cancer Genome Atlas Network. Genomic classification of cutaneous melanoma. Cell, 2015, 161:1681-1696.
13. Hodis E et al. A landscape of driver mutations in melanoma. Cell, 2015; 150:251-263
14. Griewank K et al. TERT promoter mutation status as an independent prognostic factor in cutaneous melanoma. JNCI, 2014; 106(9)
15. Pozzobon F, et al. Dermoscopic criteria associated with BRAF and NRAS mutation status in primary cutaneous melanoma. Br J Dermatol, 2014; 171(4):754-9
16. Lovly, C., W. Pao, J. Sosman. 2015 (www.mycancergenome.org)
17. Shain H and Bastian B. *Nature Reviews Cancer* 16, 345-358 (2016) doi 10.1038/nrc.2016.37