

Corporate Medical Policy

Molecular Testing for Cutaneous Melanoma AHS-M2029

File Name: molecular_testing_for_cutaneous_melanoma
Origination: 1/1/2019
Last Review: 7/2023

Description of Procedure or Service

Cutaneous melanoma is a common and serious form of skin cancer. Diagnosis of this type of cancer often involves visual examination of the skin by a dermatologist; however, due to the various presentations of nodules, it can be difficult to properly recognize a melanoma case. Although several visual systems have been developed to assist in diagnosis (such as the ABCDE system for signature features), a biopsy is often performed to diagnose a case (Swetter, 2021).

Genetic testing (particularly gene expression panels) has been explored by researchers to assist in diagnosing cases without a biopsy (Bollard et al., 2021). Another application of genetic testing is “targeted testing.” certain genetic mutations show better patient response with targeted treatment, and it is therefore clinically useful to identify these mutations to help guide selection of the most appropriate therapy.

Related Policies:

Testing for Colorectal Cancer Management AHS- M2026

Testing for Targeted Therapy of Non-Small-Cell Lung Cancer AHS- M2030

******Note: This Medical Policy is complex and technical. For questions concerning the technical language and/or specific clinical indications for its use, please consult your physician.***

Policy

BCBSNC will provide coverage for molecular testing for cutaneous melanoma when it is determined the medical criteria or reimbursement guidelines below are met.

Benefits Application

This medical policy relates only to the services or supplies described herein. Please refer to the Member's Benefit Booklet for availability of benefits. Member's benefits may vary according to benefit design; therefore member benefit language should be reviewed before applying the terms of this medical policy.

When Molecular Testing for Cutaneous Melanoma is covered

- 1) For individuals who have received genetic counseling and who have a diagnosis of cutaneous melanoma, genetic testing for cutaneous melanoma-associated gene mutations (single gene or multi-gene panel testing) is considered medically necessary when **one** of the following conditions is met:
 - a) When the individual has three or more invasive cutaneous melanomas
 - b) When the individual has invasive cutaneous melanoma and a personal history of pancreatic cancer and/or astrocytoma.

Molecular Testing for Cutaneous Melanoma AHS-M2029

- 2) For individuals who have received genetic counseling and who are in a family with a known deleterious familial mutation in a cutaneous melanoma-associated gene, the following genetic testing for inherited forms of melanoma is considered medically necessary:
 - a) Testing restricted to the known familial mutation
 - b) Comprehensive genetic testing, including multi-gene panel testing, when the specific familial mutation is unknown.
- 3) For individuals who have received genetic counseling, multi-gene panel testing for cutaneous melanoma-associated gene mutations is considered medically necessary only if a family mutation is unknown **and** the individual has a family history of **one** of the following:
 - a) Three or more invasive cutaneous melanomas.
 - b) Pancreatic, renal, and/or breast cancer.
 - c) Astrocytoma.
 - d) Uveal melanoma.
 - e) Mesothelioma.
- 4) For individuals who are being considered for molecular-targeted therapy and who have stage III or stage IV cutaneous melanoma, testing of tumor tissue for *BRAF*-, *KIT*, and *NRAS* gene mutations (single gene or multi-gene panel testing) is considered medically necessary.
- 5) For individuals with a suspicious primary melanocytic skin lesion, risk assessment of the lesion using the DermTech Pigmented Lesion Assay (PLA) (see Note 1) is considered medically necessary to help inform a biopsy decision when **ALL** of the following conditions are met:
 - a) When lesion size is 5-19 mm;
 - b) When the lesion meets 1 or more ABCDE criteria (Asymmetry, Border, Color, Diameter, Evolving);
 - c) When the lesion is located where the skin is intact (i.e., non-ulcerated or non-bleeding lesions) and free of psoriasis, eczema, or similar skin conditions;
 - d) When the lesion does not contain a scar;
 - e) When the lesion has not been previously biopsied;
 - f) When the lesion is not already clinically diagnosed as benign or melanoma;
 - g) When the lesion is NOT located on the palms of hands, soles of feet, nails, mucous membranes, or hair covered areas that cannot be trimmed.

When Molecular Testing for Cutaneous Melanoma is not covered

1. For all situations not described above, genetic testing for inherited forms of cutaneous melanoma is considered investigational.
2. For all other situations not described above, genetic expression profiling tests for cutaneous melanoma is considered investigational.
3. For all situations not described above (e.g., individuals not being considered for targeted therapy, other forms or stages of melanoma), testing for *BRAF*, *KIT*, *NRAS*, and other mutations is considered investigational.
4. Prognostic biomarker testing of a tumor biopsy to assess the risk of melanoma progression (e.g., AMBLor®) is considered investigational.

Molecular Testing for Cutaneous Melanoma AHS-M2029

***Note 1:** While this is not considered a surgical procedure, it is recommended that individuals who are receiving this test should first undergo an informed consent process to discuss the benefits and risks of pursuing this test versus receiving a biopsy to rule out melanoma.

***Note 2:** For 5 or more gene tests run on the same platform, please refer to AHS-R2162 Reimbursement Policy.

Policy Guidelines

Skin cancer is the most common form of cancer, arising from the metaplastic transformation from any of the cell types of the skin (Linares et al., 2015). Melanomas, which develop from the pigment-producing melanocytes, although much less prevalent than non-melanoma skin cancer, are increasing in incidence (Holmes, 2014; Lee & Lian, 2018). Early and accurate diagnosis is essential, as late-stage melanoma is among the most fatal forms of skin cancer (Cockerell et al., 2017). This, however, presents a significant challenge due to the difficulty of interpreting the histopathology of melanoma and the resulting interobserver and intra-observer variability (Elmore et al., 2017; Gerami et al., 2014).

Genetic Testing for Familial Cutaneous Melanoma

A family history of melanoma is reported by about 10% of melanoma patients (Soura et al., 2016b), and inherited germline mutations reportedly “increase melanoma risk from 4- to >1000-fold” (Ransohoff et al., 2016). Determining the genetic origin, however, is complicated, as a portion of familial melanoma can be attributed to shared sun exposure experiences in family members with susceptible skin types (Goldstein & Tucker, 2001).

The majority of familial cases lack identifiable germ-line mutations in either known susceptibility genes or in genes commonly mutated in sporadic melanoma (Hawkes et al., 2016). Uncommon, but high-risk, alleles have been found to contribute to the hereditary cancer phenotype that includes unilateral lineage, multi-generational, multiple primary lesions, and early onset of disease (Soura et al., 2016b). Additional research has identified a relationship between telomere length and familial melanoma; patients with familial melanoma had longer telomeres compared to patients with sporadic melanoma (Menin et al., 2016). As such, genes such as *TERT*—which encodes for the catalytic subunit of telomerase—and proteins such as POT1—a shelterin complex protein—have been implicated in the presence of multiple primary melanomas and early-onset melanoma in a subset of high-density families (Rossi et al., 2019).

Cyclin-dependent kinase inhibitor 2A (*CDKN2A*) and cyclin-dependent kinase 4 (*CDK4*) are the most commonly identified gene mutations in familial forms of melanoma, which can be defined as a family where either two first-degree relatives or three or more melanoma patients on the same side of the family are diagnosed with melanoma. Germline *CDKN2A* mutations have been identified in approximately 20-40% of familial melanoma cases where three or more family members are affected (Harland et al., 2016; Rossi et al., 2019). *CDKN2A* encodes several proteins involved in cell cycle regulation, including p16, which inhibits *CDK4* (Hussussian et al., 1994; Koh et al., 1995), and p14ARF, which inhibits *MDM2* from regulating p53 (Zhang et al., 1998). Germline *CDKN2A* mutations in melanoma families are usually missense or nonsense changes that impair the function of the p16 protein, allowing for unchecked cell cycle progression; however, rare mutations in the p14ARF protein have also been reported and result in proteasomal degradation of p53 with subsequent accumulation of DNA damage (Marzuka-Alcala et al., 2014). Overall survival is worse in those with a germline *CDKN2A* mutation than those with sporadic melanoma or familial melanoma with wild-type *CDKN2A* genes. Germline mutations also predisposed patients to an increased number of malignancies, such as pancreatic and lung cancer (Tsao & McCormick, 2022).

Mutations in *CDKN2A/p16* are associated with familial atypical multiple mole-melanoma (FAMMM syndrome), which is characterized by numerous nevi (some atypical), a family history of melanoma, and an increased risk of pancreatic cancer (Goldstein et al., 2007; Lynch & Krush, 1968). Carriers of a FAMMM mutation typically present with cancer at a younger age than non-carriers (Middlebrooks et

Molecular Testing for Cutaneous Melanoma AHS-M2029

al., 2019). Mutations in p14ARF are linked to Melanoma-Astrocytoma Syndrome (MAS), a variant of FAMMM characterized by both cutaneous melanomas and nervous system tumors (Randerson-Moor et al., 2001). Inheritance of *CDKN2A* mutations are autosomal dominant, but these mutations have variable penetrance based on sun exposure patterns and coinheritance of other melanoma-associated variants, conferring a 76% lifetime risk of developing melanoma in the US (Bishop et al., 2002; Cust et al., 2011). Mutations in *CDK4* are even less common, but were most often found affecting arginine 24, resulting in a CDK4 protein that is insensitive to inhibition by the p16 protein. No apparent differences exist in the phenotype (e.g., age at diagnosis, number of melanomas) of families carrying either *CDKN2A* or *CDK4* mutations. In aggregate, between 20-45% of familial melanomas are associated with germline mutations in *CDKN2A* or *CDK4* (Goldstein et al., 2007; Nelson & Tsao, 2009).

Other rare mutations have been associated with melanoma. Germline variations in the melanocortin-1 receptor (*MC1R*) gene alter the risk of melanoma in individuals with and without *CDKN2A* mutations (Marzuka-Alcala et al., 2014; Pasquali et al., 2015; Wendt et al., 2018). Germline variants in genes that encode for *BRCAl*-associated protein-1 (*BAP1*), telomerase reverse transcriptase (*TERT*), and microphthalmia-associated transcription factor (*MITF*) have also been added to the list of genes harboring familial melanoma-predisposing mutations (Simone et al., 2017; Soura et al., 2016a). These are more often associated with “mixed cancer syndrome,” where melanoma may appear in the context of a more general predisposition for malignancy. The *BAP1* tumor syndrome is associated with the appearance of cutaneous melanoma, uveal melanoma, and various internal malignancies (Wiesner et al., 2011). Mutations in the promoter region of *TERT*, the protein component of telomerase, and in various components of the shelterin complex have been associated with a higher incidence of melanoma and other internal malignancies (Burke et al., 2013; Horn et al., 2013). Mutations in *MITF* are associated with a higher nevus count, cutaneous malignant melanoma onset before 40 years of age, and non-blue eye color with no association to freckling, skin color, or hair color (Bertolotto et al., 2011; Yokoyama et al., 2011). Xeroderma pigmentosum (XP) is a rare disorder in which patients have a mutation in genes involved in nucleotide excision repair (NER). Patients with mutations in *XPC* and *XPD* have an increased risk of melanoma (Paszowska-Szczur et al., 2013). Lastly, Cowden syndrome, a type of *PTEN* hamartoma tumor syndrome characterized by the appearance of trichilemmomas, papillomatous papules, mucosal lesions (papules) and palmar-plantar keratosis within the first three decades of life, is associated with a higher risk of melanoma (Bubien et al., 2013; Soura et al., 2016a).

Several proprietary gene panels exist for assessment of familial cutaneous melanoma. For example, the DermTech Pigmented Lesion Assay (PLA) leverages their “Smart Sticker” technology to remove cellular material from the stratum corneum, the material of which is then analyzed for over-expression of *LINC00518* (*long intergenic non-coding RNA 518*) and *PRAME* (*preferentially expressed antigen in melanoma*), which may be incidental with genomic atypia associated with melanomas (Gerami et al., 2017). Moreover, Invitae offers a 12-gene panel (9 primary genes plus 3 genes with “preliminary evidence for melanoma”), Fulgent offers a 14-gene panel, and GeneDx offers a 9-gene panel (Fulgent, 2023; GeneDx, 2023; Invitae, 2023). These panels may include genes traditionally associated with familial melanoma itself (such as *CDKN2A*) as well as genes whose variants are not primarily associated with familial melanoma, but confer added risk regardless (Tsao & McCormick, 2022).

Clinical Utility and Validity of Genetic Testing for Familial Cutaneous Melanoma

The frequency of *CDKN2A* mutations in patients with a single primary melanoma or multiple primary melanoma were 1.2% and 2.9%, respectively (Berwick et al., 2006); however, depending on selection criteria, mutation frequency rates of *CDKN2A* can range from 5% to 72% (Delaunay et al., 2017) with a family history of melanoma considered the most important risk factor. The established rule of three is used when proposing genetic testing for primary melanomas; it is generally understood that when three or more melanomas or genetically related cancers are identified in the same patient, or in first- and second-degree relatives, the pretest probability is increased above 10% and the cost of genetic screening can be justified (Leachman et al., 2017).

In a study on *CDKN2A* genetic testing conducted in Sweden between 2015 and 2020, 913 members across 403 families were identified with cutaneous melanoma. Pissa et al. (2021) found that melanoma

Molecular Testing for Cutaneous Melanoma AHS-M2029

cases found in families testing positive for pathogenic variants of *CDK2NA* boasted significantly higher mortality rates, such that families with the variants had 37.6% melanoma cases that died from melanoma as compared to the 10.0% without ($p < 0.001$), independent of age, sex, and tumor stage. This significant melanoma-specific mortality associated with families with *CDK2NA* variants is motivation to identify and enroll carrier families in a preventive surveillance program (Pissa et al., 2021). However, a potential confounding variable involves the diagnoses of pancreatic cancers in the subjects in the study; as pathogenic strains of *CDK2NA* are also associated with cancers other than melanomas, the 129 of the 913 members present a limitation to the specificity of the study as there did not appear to be a clear manner of segregating its influence.

Stolarova et al. (2020) analyzed 264 Czech melanoma patients with early onset, double primary tumors or family history by next generation sequencing NGS analysis of 217 genes, and they identified that “mutations in high-to-moderate melanoma risk genes and in other cancer syndrome genes were significantly associated with melanoma risk,” with those genes including *CDKN2A*, *POT1*, and *ACD* for high-to-moderate melanoma risk, and *NBN*, *BRCA1/2*, *CHEK2*, *ATM*, *WRN*, and *RBI* for other cancer syndrome genes. An increased potential of carrying mutations was found in “patients with double primary melanoma, melanoma and other primary cancer, but not in patients with early age at onset” (Stolarova et al., 2020). *CDK2NA* was the most frequently mutated gene among those with high-to-moderate risk, and in other studies reviewed by Stolarova et al. (2020), there was an increased risk for pancreatic cancer among families with *CDK2NA* mutation, and a more established family history.

Leachman et al. (2017) published an updated algorithm for the identification, testing, and management of hereditary melanoma; the rule of three has been incorporated into this algorithm as an indication for genetic testing in multiple melanomas. The researchers state that “Any patient or family that meets the updated rule of threes should be considered a candidate for genetic testing. If melanoma is the only cancer in a pedigree, then to meet the threshold of genetic testing, a pedigree should have three primary melanomas in first- or second-degree relatives in areas with a high melanoma incidence or two primary melanomas in a low-incidence area. This melanoma panel should include *BAP1*, *CDK4*, and *CDKN2A*. Genes for which risk has not been established but for which studies suggest an elevated risk include *MITF* and *POT1* and we recommend including these in the melanoma panel” (Leachman et al., 2017).

Genetic testing for commonly known cutaneous melanoma mutations can be utilized to determine prognosis and overall survival. Aoude et al. (2020) found that “germline mutation status was the most significant biomarker for OS [overall survival]” and “survival outcomes for germline carriers are poor with the current standards of care.” When using *BRAF* status and tumor mutation burden (TMB) for prognosis of cutaneous melanoma patients, “*BRAF* V600 wild-type patients had significantly longer PFS [progression-free survival] than the V600 mutant group ($p = 0.0317$) ... For stage III/IV resected patients, TMB was also significantly associated with longer PFS ($p = 0.0034$)” (Aoude et al., 2020). The greater the number of recognizable mutations, the more targeted attacks against cancerous cells can be made and the better the prognosis.

Genetic Testing for Acquired Cutaneous Melanoma

Melanoma is a particularly lethal and aggressive cancer with the ability to metastasize to any organ (Leong et al., 2011). Cutaneous tumors as little as 1 mm in thickness are capable of lymph node metastasis (Stage III) which results in a significant decrease in the 5-year survival rate from 90% to 56% (Yee et al., 2005). If the cancer has spread beyond the lymph nodes (Stage IV), an even more dramatic decrease in 5-year survival to 15% will occur. (Grossmann, et al., 2012). Genetic testing for acquired melanoma can identify the activating genetic alterations.

Genetic testing is important to determine the most efficient treatment method for a melanoma patient. Despite the high frequency in nevi, the role of *BRAF* mutations in oncogenesis is well established (Davies & Gershenwald, 2011) and has been confirmed in clinical trials (Flaherty et al., 2010). *BRAF* is a member of the RAF family of protein serine/threonine kinases (*ARAF*, *BRAF*, *CRAF*) that is activated by Ras proteins during intracellular signaling cascades. Mutations in *BRAF* appear to be the most common genetic alteration in melanoma (Hocker & Tsao, 2007) and occur more frequently in

Molecular Testing for Cutaneous Melanoma AHS-M2029

melanoma than lung, colon, and ovarian carcinoma (Grossmann et al., 2012). The remarkable efficacy of *BRAF* inhibitors led to the accelerated approval of vemurafenib for unresectable and metastatic melanoma. Importantly, *BRAF* mutation testing is warranted for determining therapeutic eligibility as selective *BRAF* inhibitors pose significant risk of cutaneous squamous cell carcinoma and have the potential to increase disease progression in *BRAF* wild type (mutation negative) tumors (Grossmann et al., 2012).

BRAF, *KIT*, *PIK3CA* and *NRAS* mutations are commonly seen in melanoma cases (Alrabadi et al., 2019; Lokhandwala et al., 2019) with an estimated 50% of melanomas exhibiting the *BRAF* V600E mutation (Burjanivova et al., 2019).

Table 1. Frequency of melanoma subtypes with activating genetic alterations in *BRAF* and *KIT* (Grossmann et al., 2012).

Aberration	Cutaneous –CSD	Cutaneous + CSD	ALM	MM
BRAF mutation	53%	8–11%	10%	15%
KIT mutation; amplification	0	17%	11–38%; 19–27%	6– 19%; 20– 33%

A variety of methods are utilized for *BRAF* and *KIT* mutational analysis testing in melanoma, which has resulted in no standardized procedures for testing. Because numerous techniques are available and updated methods continue to be released, labs have been reluctant to switch *BRAF* platforms to accommodate one specific drug for one disease (Grossmann et al., 2012). Current *BRAF* genetic testing methods include *BRAF* V600E by real-time PCR, *BRAF* (V600E) mutation only by Sanger sequencing, *BRAF* full gene sequence analysis, and *BRAF* next generation sequencing (CMGL, 2014).

However, a recent study found good overall compliance of labs with the College of American Pathologists (CAP) (Cree et al., 2014) and National Comprehensive Cancer Network (NCCN) guidelines for molecular diagnosis of tumors (Volmar, et al., 2015) despite not using the specific FDA-approved test.

Table 2. Molecular Testing Adherence to NCCN Guidelines (taken from (Volmar et al., 2015))

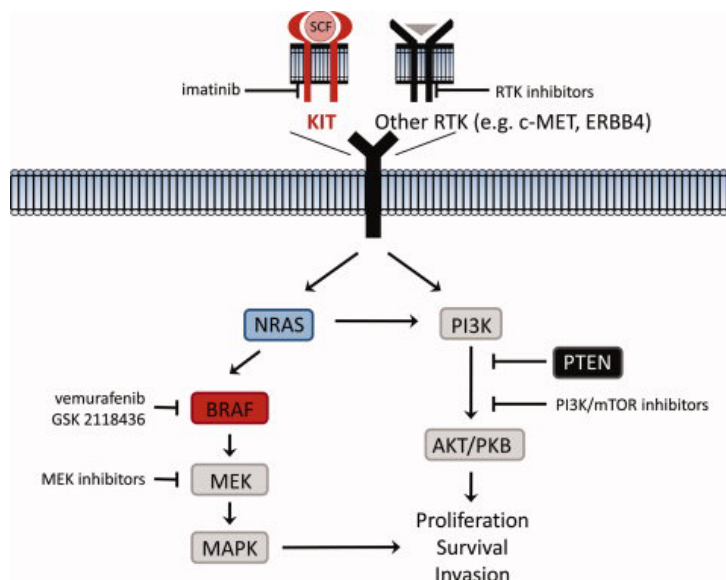
	All Institutions Percentiles					
	n	10 th	25 th	Median	75 th	90 th
Retrospective study (lung, colorectal, melanoma)						
Percentage of tests that strictly meet the guideline	26	32.6	64.7	70.9	82.7	89.7
Percentage of tests that at least loosely meet the guideline	26	57.4	90.7	95.1	98.9	100.0

Molecular Testing for Cutaneous Melanoma AHS-M2029

Prospective study (all case type)						
Percentage of tests that strictly meet the guideline	23	20.0	31.4	53.3	66.7	70.5
Percentage of tests that at least loosely meet the guideline	23	75.0	87.0	94.3	100.0	100.0

Historically, systemic therapy for metastatic melanoma provided very low response rates and little to no benefit in overall survival (Atkins, et al., 2000; Tsao, et al., 2004). However, this is beginning to change. For example, researchers have now identified that those with the *BRAF* V600 mutation are able to obtain both immune checkpoint inhibitor therapy and other targeted therapies for an improved overall treatment regimen (Ghate et al., 2019). Moreover, the immune-boosting anti-CTLA-4 antibody ipilimumab (Hodi et al., 2010) and testing and development of small molecule kinase (*KIT* and *BRAF*) inhibitors have yielded improvements in long-term survival for melanoma patients (Davies & Gershenwald, 2011; Ribas & Flaherty, 2011; Woodman & Davies, 2010).

Figure 1: Imatinib and RTK Inhibitor Pathways for Melanoma Treatment (image taken from (Grossmann et al., 2012)).

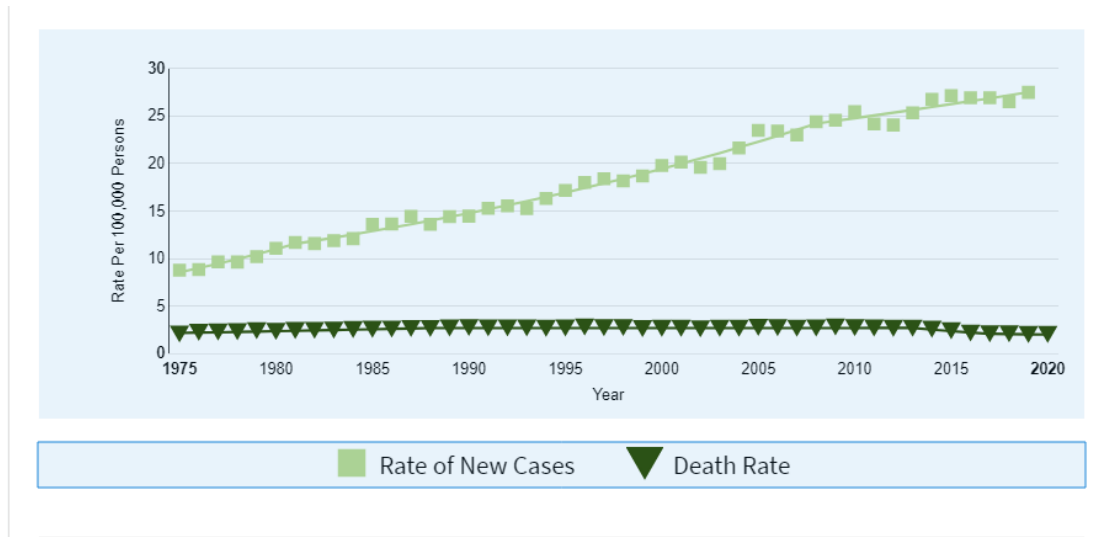


The lifetime risk for the general population of developing melanoma is 1 in 55 (Institute, 2023; NCI, 2023b) and that risk has increased approximately 2% annually since 1960 (Rashid & Zager, 2015). National Cancer Institute now reports that melanoma of the skin represents 5.2% of all new cancer cases in the United States; approximately 99,780 Americans in total will be diagnosed with melanoma in 2022 (NCI, 2023b).

In 2022, the number of new cases of melanoma of the skin per 100,000 people was 21.5. Further, the number of melanoma patient deaths in 2022 was 2.1 per 100,000 individuals, accounting for 1.3% of all cancer deaths in United States (NCI, 2023b).

Figure 2. New Cases and Deaths of Melanoma of the Skin per 100,000 Persons (taken from (NCI, 2023b))

Molecular Testing for Cutaneous Melanoma AHS-M2029



Clinical Utility and Validity of Genetic Testing for Acquired Cutaneous Melanoma

O'Brien et al. (2017) analyzed samples to determine if *BRAF* mutation identification by immunohistochemistry was a suitable alternative to PCR; 132 patients were included in this study, and the anti-*BRAF* V600E VE1 clone antibody was used for immunohistochemistry detection. A sensitivity of 86.1% and specificity of 96.9% was shown with the anti-*BRAF* V600E VE1 clone antibody; “The concordance rate between PCR and immunohistochemical *BRAF* status was 95.1% (116/122) (O'Brien et al., 2017).” As both methods were in high agreement, immunohistochemistry may be a viable alternative to PCR for *BRAF* mutation testing.

Corean et al. (2019) utilized several different techniques on metastatic melanoma samples, including bone marrow morphology, histology, immunophenotyping, molecular genetic testing and *BRAF* V600E immunohistochemistry. *BRAF* immunohistochemistry was detected in two patients, and molecular testing confirmed these results; researchers then stated that “*BRAF* V600E immunohistochemistry is useful as a surrogate marker of molecular results,” once again highlighting the fact that immunohistochemistry may be a viable alternative for *BRAF* mutation testing (Corean et al., 2019).

Available *BRAF* tests and analytical sensitivities:

- “The *BRAF* V600E by real-time PCR test uses a TaqMan® Mutation Detection Assay to detect the V600E mutation in exon 15 of *BRAF* in tumor (somatic) cells. The sensitivity of the TaqMan assay is ~0.1% mutant DNA in a wild-type background. Poor DNA quality, insufficient DNA quantity or the presence of PCR inhibitors can result in uninterpretable or (rarely) inaccurate results.
- The *BRAF* (V600E) mutation only by Sanger sequencing uses a DNA-based PCR-sequencing assay to detect the V600E in exon 15 of *BRAF*. The limit of detection for Sanger sequencing is >20% mutant DNA in a wild-type background.
- The *BRAF* full gene sequence analysis test uses a DNA-based PCR-sequencing assay to detect point mutations in the coding sequence and intron/exon boundaries of the *BRAF* gene. The sensitivity of DNA sequencing is over 99% for the detection of nucleotide base changes, small deletions and insertions in the regions analyzed. Rare variants at primer binding sites may lead to erroneous results. The limit of detection for Sanger sequencing is >20% mutant DNA in a wild-type background” (CMGL, 2014).

Molecular Testing for Cutaneous Melanoma AHS-M2029

- The *BRAF* NGS with TruSeq had sensitivity of over 99% (Froyen et al., 2016).

Colombino et al. (2020) compared *BRAF* mutation testing performed with conventional nucleotide sequencing approaches (Sanger sequencing and pyrosequencing) with real-time polymerase chain reaction (RT-PCR) or next-generation sequencing (NGS) assays to assess the levels of concordance between these various techniques. 319 tissue samples were analyzed and initially screened with conventional approaches. The initial screen found pathogenic *BRAF* mutations in 144 (45.1%) cases. RT-PCR (Idylla™ *BRAF* mutation assay) detected 11 (16.2%) and 3 (4.8%) additional *BRAF* mutations after Sanger sequencing and pyrosequencing, respectively. NGS detected one additional *BRAF*-mutated case (2.1%) among 48 wild-type cases previously tested with pyrosequencing and RT-PCR. According to the data, RT-PCR is more accurate than both Sanger sequencing and pyrosequencing in detecting *BRAF* mutations. Overall, RT-PCR had a good concordance with the other tests; “real-time PCR is a rapid method which achieves the same maximum level of sensitivity of NGS (up to 98%), without requiring particular skills” (Colombino et al., 2020). According to the author, “[RT-PCR and NGS] improved the diagnostic accuracy of *BRAF* testing via the detection of additional *BRAF* mutations in a subset of false-negative cases previously tested with Sanger sequencing or pyrosequencing. In attendance of further confirmations in larger prospectively designed studies, the use of two sensitive molecular methods may ensure the highest level of diagnostic accuracy” (Colombino et al., 2020).

BRAF analysis is an accepted medical practice for patients with unresectable, metastatic stage IV melanoma (NCCN, 2023). However, recent randomized controlled trials have indicated the benefits of expanding *BRAF* analysis. A study published in The Lancet Oncology by Amaria et al. (2018) compared standard of care in patients with high-risk, surgically resectable melanoma (stage III or IV) to similar patients receiving a regimen of a neoadjuvant plus adjuvant dabrafenib and trametinib. All patients had to be of confirmed *BRAF* V600E or *BRAF* V600K status to participate in either the control or experimental groups. In the follow-up (median of 18.6 months), 10/14 (or 71%) of patients in the experimental group remained event-free (i.e. alive without disease progression), whereas 0/7 (0%) of the control group receiving standard of care remained event-free. The authors conclude that the “Neoadjuvant plus adjuvant dabrafenib and trametinib significantly improved event-free survival versus standard of care in patients with high-risk, surgically resectable, clinical stage III-IV melanoma” (Amaria et al., 2018).

Another study published by Zippel et al. (2017) researched the use of perioperative *BRAF* inhibitors on patients with stage III melanoma. All patients had to be confirmed *BRAF* V600E to participate in the study. Of the thirteen patients, twelve “patients showed a marked clinical responsiveness to medical treatment, enabling a macroscopically successful resection in all cases”; moreover, “at a median follow up of 20 months, 10 patients remain free of disease” (Zippel et al., 2017). One patient died prior to surgery in this study. The authors conclude that the “Perioperative treatment with *BRAF* inhibiting agents in *BRAF*V600E mutated Stage III melanoma patients facilitates surgical resection and affords satisfactory disease free (sic) survival” (Zippel et al., 2017).

Gene Expression Profiling and Biomarkers for Diagnosis and Prognosis

Currently, patients presenting with a suspicious pigmented lesion undergo excisional biopsy which is then subjected to histopathologic examination by a pathologist (NCCN, 2023). The majority of melanocytic neoplasms can be accurately classified by this approach; however, in some cases confidently differentiating benign melanocytic nevi from malignant melanoma can be extremely difficult or impossible despite additions to histopathologic assessment, such as the evaluation of Breslow depth (Chiaravalloti et al., 2018; Lee & Lian, 2018). In these cases, even diagnoses from expert pathologists can be discordant (Elmore et al., 2017; Farmer, et al., 1996; Gerami et al., 2014) and subject to diagnostic drift (Bush, et al., 2015).

A number of diagnostic and prognostic genetic tests for melanoma have been developed as ancillary tests to assist in this differentiation and resultant risk stratification (Lee & Lian, 2018), including microRNAs as biomarkers to distinguish between melanomas and nevi (Torres et al., 2019). In particular, gene

Molecular Testing for Cutaneous Melanoma AHS-M2029

expression profiling is growing in popularity for the diagnosis and prognosis of cutaneous melanoma. Molecular tests based on gene expression profiling of cutaneous melanoma are commercially available. They include the MyPath Melanoma (Castle Biosciences, 2023b), 2-GEP Pigmented Lesion Assay, Decision-Dx Melanoma (Castle Biosciences, 2023b), and a clinicopathologic (CP)-GEP model by SkylineDx (SkylineDx, 2023).

Another risk stratification test is the melanoma prognostic test, AMBLor®, described by Avero Diagnostics as a test used to identify “early-stage melanomas at low risk of progression.” Avero Diagnostics licensed this biotechnology from AMLo Biosciences Ltd. The AMBLor® test looks for the presence of two prognostic biomarker proteins, AMBRA1 and loricrin, both of which are found in the skin overlying the tumor. An absence of these biomarkers in AJCC stages I and II indicates melanoma at a normal or high risk of progression, while normal expression of the biomarkers is linked to a low risk of cancer progression (AMLo Biosciences, 2023).

Clinical Utility and Validity of Gene Expression Profiling and Biomarkers for Diagnosis and Prognosis

MyPath Melanoma (Castle Biosciences)

A 23-gene expression profile and algorithm that assigns various weights and thresholds of expression for each gene was developed to differentiate benign melanocytic nevi from malignant melanoma; this gene expression profile and algorithm was determined to have a sensitivity of 89% and specificity of 93% (Clarke et al., 2015). Further, three experienced dermatopathologists validated this method against an independent histopathologic evaluation; a sensitivity of 91.5% and a specificity of 92.5% was determined (Clarke, et al., 2017). This gene expression profile was developed into a commercial test known as MyPath Melanoma which generates a single numerical score along with a classification of likely malignant, likely benign, or indeterminate, for a given cutaneous lesion. Across various validation studies, MyPath Melanoma has demonstrated a sensitivity in the range of 90-94% and a specificity in the range of 91-96% (Ko et al., 2017).

MyPath Melanoma correlates closely with long-term clinical outcomes by adding valuable adjunctive information to aid in the diagnosis of melanoma. An examination of the utility of this test (Cockerell et al., 2017) found that the results of this gene expression signature have a significant clinical impact with 71.4% (55/77) of cases changing from pretest recommendations to actual treatment. The majority of changes were consistent with the test result. There was an 80.5% (33/41) reduction in the number of biopsy site re-excisions performed for cases with a benign test result. However, when more challenging samples were included (Minca et al., 2016) with 39 histopathologically unequivocal lesions (15 malignant, 24 benign) and 78 challenging lesions interpreted by expert consensus (27 favor malignant, 30 favor benign, and 21 ambiguous), myPath Melanoma had a lower sensitivity and specificity than fluorescence in situ hybridization (FISH) (FISH: 69% sensitivity, 91% specificity; myPath; 55% sensitivity, 88% specificity).

In contrast, (Clarke et al., 2020) completed another study that evaluated the accuracy of myPath melanoma in diagnostically uncertain neoplasms. Out of 125 diagnostically “uncertain” cases, myPath melanoma demonstrated a sensitivity of 90.4%. In this study, the test displayed no significant difference in sensitivity between the diagnostically uncertain (equivocal) and unequivocal lesion groups. It is unclear why myPath melanoma would perform better in one population of diagnostically “uncertain” lesions over another, though the authors cite certain limitations of the study. These include reliance on a single diagnostic slide of each biopsy specimen, and lack of access to complete clinical and demographic information (Clarke et al., 2020). Given varying results, further study may be required to develop a consensus on the performance of myPath melanoma.

Pigmented Lesion Assay (PLA) (DermTech)

DermTech has developed a pre-diagnostic quantitative polymerase chain reaction (qPCR)-based pigmented lesion assay (PLA) that measures the expression of two genes in the stratum corneum to assist with diagnostic or prognostic information for potential melanoma cases (Varedi et al., 2019). DermTech’s PLA identifies malignant changes on a genomic level that cannot be detected with the human eye; this

Molecular Testing for Cutaneous Melanoma AHS-M2029

assay can be used to support clinicians in their decision to biopsy suspicious nevi. This test has the potential to increase the number of early melanomas biopsied and reduce the number of benign lesions biopsied, thereby improving patient outcomes (Ferris et al., 2017). A recent study has given this pigmented lesion assay a sensitivity of 91-96%, a specificity of 69-91%, and a negative predictive value of approximately 99% (Ferris, Rigel, et al., 2019).

To help support clinicians in their decision to biopsy, this noninvasive 2-gene expression assay of the *LINC00518* and *PRAME* genes has been developed for use on adhesive patch biopsies. Skin sampling via an adhesive patch allows for DNA, RNA, skin tissue and microbiome samples to be safely obtained and transported cost effectively by mail at room temperature; skin cells, T-cells, dendritic cells, melanocytes and other types of cells can be analyzed by this method (Yao et al., 2017). The use of an adhesive patch also allows 100% of the lesion's surface to be sampled, compared to less than 1-2% of surgical biopsies (DermTech, 2023b). Further, this technique is a much more cost-effective option. Hornberger and Siegel (2018) report that PLA testing could save approximately \$447 per lesion compared to traditional biopsies.

Gerami et al. (2017) tested the validity of the two-gene panel based on *LINC00518* and *PRAME* on differentiating melanoma from nonmelanoma in a multicenter study across 28 sites in the United States, Europe, and Australia. In a sample of 398 (87 melanomas and 311 nonmelanomas), it was found that this classification method was able to accurately identify melanomas from nonmelanomas with a sensitivity of 91% and a specificity of 69%. An application study of 381 patients found that the estimated real-world sensitivity of the DermTech was 95% and specificity was 91% (Ferris et al., 2018); overall, 93% of PLA results positive for both *LINC00518* and *PRAME* were diagnosed histopathologically as melanoma. Further, this study was also used to identify if the real-world clinical use of the DermTech PLA could change physician behavior and reduce the overall number of biopsies performed. The PLA identified 51 PLA(+) test results, and 100% of these pigmented skin lesions were biopsied (37% were melanomas). Furthermore, "Nearly all (99%) of 330 PLA(-) test results were clinically managed with surveillance. None of the three follow-up biopsies performed in the following 3-6 months, were diagnosed as melanoma histopathologically (Ferris et al., 2018)." The PLA test altered clinical management of pigmented lesions and shows high clinical performance. However, it is uncertain if the negative samples—as determined by the PLA—will remain so, as "we [the authors] cannot rule out that some PLA(-) lesions may not have been adequately reassessed in the follow-up period and we certainly recommend erring on the side of caution and surgically biopsying a lesion in question if additional risk factors, further clinical suspicion, or patient concern mandate such a step" (Ferris et al., 2018).

The PLA test has also been validated against driver mutations in melanoma, including *BRAF*, *NRAS*, and *TERT*. Ferris, Moy, et al. (2019) studied the mutation frequency of these genes using samples obtained from the PLA technique (e.g. adhesive patch) in both histopathologically confirmed melanomas (n = 30) and non-melanoma controls (n = 73). "The frequency of these hotspot mutations in samples of early melanoma was 77%, which is higher than the 14% found in nonmelanoma samples (P < 0.0001). *TERT* promoter mutations were the most prevalent mutation type in PLA-positive melanomas; 82% of PLA-negative lesions had no mutations, and 97% of histopathologically confirmed melanomas were PLA and/or mutation positive (Ferris, Moy, et al., 2019)." 86% of the non-melanomas within this validation cohort contained no mutations. The authors next analyzed 519 real-world PLA samples for the same mutations. Similar to the previous validation cohort, 88% of this larger cohort also contained no mutations., indicating that the PLA test can rule out lesions with few mutational risk factors for melanoma.

Skelsey et al. (2021) further confirmed the "real-world" negative predictive value (NPV) and positive predictive value (PPV) of the PLA, "by following a cohort of 1,233 PLA-negative pigmented lesions for evidence of malignancy for up to 36 months and by re-testing a separate prospective cohort of 302 PLA-negative lesions up to 2 years after initial testing." Out of this total PLA-negative cohort, 1,233 lesions had a confirmed follow-up evaluation; of these, 10 melanomas were subsequently detected by way of biopsy, resulting in an NPV of 99.2%. Out of 316 PLA-positive cases, 59 were diagnosed as melanoma by histopathology, resulting in a PPV of 18.7% (Skelsey et al., 2021). The PPV achieved through use of the PLA may represent a 5-fold improvement over the current standard according to the test's developers.

Molecular Testing for Cutaneous Melanoma AHS-M2029

In a continuing medical education series, Skudalski et al. (2022) confirm that the PLA is to be used as a risk stratification aid in determining whether to biopsy a skin lesion. The authors recommend that in the case of a positive PLA result, “biopsy *in toto* should be performed immediately.” They further comment that after a negative PLA result, clinicians have chosen to follow up within 12 months to further evaluate the skin lesion; however, a biopsy *in toto* should be considered if a patient remains concerned about a lesion, or if the lesion has evolved (Skudalski et al., 2022).

A recent report by Robinson and Jansen (2020) outlined a proof-of-concept pilot program of remote physician-guided self-sampling (i.e. telehealth administration of the adhesive patch) during the Illinois stay-at-home order of the COVID-19 pandemic. The authors also surveyed skin self-examination (SSE) anxiety as well. Two cohorts were used in this pilot, an experimental group (n=7), and a randomly selected physician-sampled control case group (n=10). The authors report that SSE-induced anxiety has increased during the COVID-19 pandemic. It should be noted that surveys were administered to much larger groups than those administering self-sampling tests. 258 surveys about SSE anxiety were conducted prior to the COVID-19 pandemic, and 211 surveys during the COVID-19 pandemic. The authors state, “Guided self-sampling led to molecular risk factor analyses in 7/7 (100%) of cases compared to 9/10 (90%) randomly selected physician-sampled control cases... Adhesive patch self-sampling under remote physician guidance is a viable specimen collection option” (Robinson & Jansen, 2020).

DecisionDx Melanoma (Castle Biosciences)

After a melanoma case has been identified, several management approaches may be considered. However, best melanoma management practices are constantly evolving. A common technique to assess the spread of a tumor, such as melanoma, is a sentinel lymph node biopsy (SLNB). This procedure is used to evaluate whether the cancer has spread beyond the original tumor site and into the lymphatic system. The lymphatic or lymph system transports fluid known as lymph throughout the body; this fluid contains white blood cells which help to fight infections. The lymphatic system also aids in ridding the body of other waste and toxins. The SLNB technique essentially helps the physician to stage the tumor. However, melanoma-related lymph node spread is very complex and is associated with many factors, including age, location, thickness, ulceration, gender and regression (Ribero et al., 2017).

The DecisionDx-Melanoma test is a gene expression profile (GEP) test that measures the expression of 31 different genes in a tumor tissue sample. This test was designed for cutaneous melanoma patients undergoing or considering SLNB and can help to identify the risk of cancer recurrence or metastasis in stage I-III melanoma (CastleBiosciences, 2023). DecisionDx-Melanoma may help physicians guide treatment options, including whether to perform a SLNB in eligible patients, and what type of follow up treatment is necessary (CastleBiosciences, 2023). After GEP analysis, the DecisionDx-Melanoma provides a score of either Class 1 (low risk) or Class 2 (high risk), as well as A or B scores with “A” reflecting a better outcome and “B” reflecting a worse outcome; this allows physicians to receive final scores of Class 1A, 1B, 2A, and 2B (Gastman, et al., 2019).

The DecisionDx-Melanoma is performed on tumor tissue biopsies that have been preserved as formalin-fixed, paraffin-embedded (FFPE) samples (CastleBiosciences, 2023). Researchers have highlighted that the identification of tissue-based prognostic markers in melanoma has been a challenging obstacle for researchers, as “One major limitation is that most primary melanomas are preserved as formalin-fixed, paraffin-embedded (FFPE) samples rather than fresh-frozen tissues because of the small size of the specimens (Weiss et al., 2015).” High-quality genetic material is more challenging to extract from FFPE samples, and messenger RNA is inconsistent in FFPE samples (Weiss et al., 2015).

The prognostic utility of this test has been measured by Keller et al. (2019); a total of 159 patients participated in this study and were followed up with, on average, 44.9 months after initial testing. Gene expression profiling results helped to categorize patients into two groups: low-risk patients were placed in Class 1, and high-risk patients in Class 2; 117 patients were placed in Class 1 and 42 patients in Class 2 (Keller et al., 2019). Results showed that this gene expression profiling test had great prognostic abilities. “Gender, age, Breslow thickness, ulceration, SNB positivity, and AJCC stage were significantly associated with GEP classification ($P < 0.05$ for all). Recurrence and distant metastasis rates were 5% and 1% for

Molecular Testing for Cutaneous Melanoma AHS-M2029

Class 1 patients compared with 55% and 36% for Class 2 patients. Sensitivities of Class 2 and SNB for recurrence were 79% and 34%, respectively” (Keller et al., 2019).

Berman et al. (2019) gathered an expert panel of nine dermatologists/dermatologic surgeons/dermatopathologists and completed 29 clinical scenarios in which gene expression tests could be used appropriately; several gene expression profiling (GEP) tests were used including a 2-GEP assay, 23-GEP assay and 31-GEP assay. “The 2-GEP assay for melanoma diagnosis received 1 B-strength and 6 C-strength recommendations. The 23-GEP diagnostic test received 1 A-strength, 3 B-strength, and 4 C-strength recommendations. The 31-GEP prognostic assay received 1 A-strength, 7 B-strength, and 6 C-strength recommendations”; these recommendations show that the 31-GEP assay received the highest recommendations by this expert panel (Berman et al., 2019).

Another study reported that the 31-GEP was validated in almost 1600 patients “as an independent predictor of risk of recurrence, distant metastasis and death in Stage I-III melanoma and can guide SLNB decisions in patient subgroups, as demonstrated in 1421 patients”; further, an appropriate 31-GEP testing population was identified and concluded that it is best used on patients with cutaneous melanoma tumors greater than or equal to 0.3 mm thick (Marks et al., 2019). However, this study reports several conflicts of interest that are important to note as multiple authors are employees at Castle Biosciences, Inc., and one author is a consultant and speaker for the company (Marks et al., 2019).

Greenhaw et al. (2020) completed a meta-analysis of the DecisionDx-Melanoma 31-GEP prognostic test in a total of 1,479 patients. This meta-analysis included participants from three different studies. The patient analysis showed that the five-year recurrence and distant metastasis-free survival rate for Class 1A patients was 91.4% and 94.1% respectively and were 43.6% and 55.5% for Class 2B patients. The 31-GEP was then used to estimate the likelihood of recurrence and distance metastasis. The GEP test exhibited a sensitivity of 76% for each endpoint, showing consistency and accuracy for the identification of at increased risk of metastasis (Greenhaw et al., 2020).

Moreover, Marchetti et al. (2020) completed an assessment of the prognostic accuracy of the 31-GEP in patients that had cancer labeled as AJCC stage I or II disease. Patients with localized melanoma were included from external validation studies, and after exclusion criteria, there were 7 studies analyzed. Five of these studies used the DecisionDx-Melanoma test. Authors looked at the results of the 7 included studies (1450 study participants) and concluded that the performance of both GEP tests (MelaGenix and DecisionDx-Melanoma) varied by AJCC stage. “For patients tested with DecisionDx-Melanoma, 623 had stage I disease (6 true-positive ([TP], 15 false-negative, 61 false-positive, and 541 true-negative [TN] results) and 212 had stage II disease 959 TP, 13 FN, 78 FP, and 62 TN results). Among patients with recurrence, Decision Dx-Melanoma correctly classified 29% with stage I disease and 82% with stage II disease. Among patients without recurrence, the test correctly classified 90% with stage I disease, and 44% with stage II disease.” The overall conclusion by authors was that the prognostic ability was “poor” in terms of accurately identifying recurrence in patients with stage I cancer (Marchetti et al., 2020).

Kangas-Dick et al. (2021) evaluated the DecisionDx-Melanoma assay in a retrospective chart review of patients at a large cancer institute. A total of 361 patients’ data were analyzed, and a follow-up period median of 15 months was established. First, sentinel node biopsy was performed for 75.9% of the patients, of which, 19.4% tested positive for recurrent cancer. Authors considered GEP class 2B status and noted that it was “significantly associated” with recurrence-free survival (RFS) and distant metastasis-free survival (MDFS) in a univariate analysis. The authors concluded that “genetic profiling of cutaneous melanoma can assist in predicting recurrence and help determine the need for close surveillance. However, traditional pathologic factors remain the strongest independent predictors of recurrence risk.” (Kangas-Dick et al., 2021).

CP-GEP (Merlin Assay) (SkylineDx)

An 8-gene expression profile test called CP [clinicopathologic]-GEP was developed to help identify patients who might safely avoid SLNB through reclassification of their risk of nodal metastasis to < 5%. In considering certain CP variables, the developers determined that Breslow thickness and patient age

Molecular Testing for Cutaneous Melanoma AHS-M2029

were sufficiently predictive of the risk of SLN metastasis. Double-loop cross-validation [DLCV] and least absolute shrinkage and selection operator (LASSO) further identified several genes that distinguished between patients with positive and negative SLNB results. Ultimately, 8 genes (*MLANA*, *GDF15*, *CXCL8*, *LOXL4*, *TGFBR1*, *ITGB3*, *PLAT*, and *SERPINE2*) along with the 2 CP variables (Breslow thickness and patient age) completed the CP-GEP model.

The CP-GEP model was tested in a cohort of 754 patients diagnosed with cutaneous melanoma, who underwent SLNB within 90 days of their diagnosis. The validity of the model changed based on different T categories of melanoma and demonstrated increasingly higher sensitivity with increasing T category. An inverse relationship was observed between specificity and T category. Theoretically, the model could have reduced the SNLB rate in T1b patients by 80%, though the rate reduction dropped as the T category increased across the patient population. The negative predictive value was 91% or higher across all T categories (Bellomo et al., 2020).

The CP-GEP model was independently validated by Yousaf et al. (2021) in a separate cohort of 208 adult patients with primary cutaneous melanoma and confirmed the trends originally published by Bellomo et al. (2020). As the T category of melanomas increased, the sensitivity of the model sequentially increased, while the specificity decreased. The highest SLNB reduction rates were observed in lower T category patients (T1), and an NPV of at least 93.3% was observed across all T categories in the study. Importantly, the model was further validated focusing only on patients 65 years of age or older, as the incidence of melanoma is greater in older patient populations. Similar results were obtained compared to the full patient cohort, demonstrating the applicability of the model across all age ranges included in the study (Yousaf et al., 2021).

11-GEP (Melagenix) (NeraCare)

Another GEP test was developed to better define the probability of patient survival at the time of first diagnosis. 11 prognostically-relevant genes previously identified through whole-transcriptome analysis (Brunner et al., 2013) of fresh-frozen melanomas were evaluated (*KRT9*, *DCD*, *PIP*, *SCGBID2*, *SCGB2A2*, *COL6A6*, *GBP4*, *KLHL41*, *ECRG2*, *HES6*, and *MUC7*), and 8 of these were found to be statistically-significantly associated with melanoma-specific survival (all but *ECRG2*, *HES6*, and *MUC7*). Based on the coded expression data of the 8 significantly associated genes, a binary GEP “score” of either 0 or 1 was generated, representing “low risk” or “high risk”, respectively.

To validate the GEP score, inter-assay variability was examined across 4 different laboratories. Concordance was high between replicates, with 93% of replicate determinations confirming the score. Next, the GEP score was challenged with a cohort of 211 melanomas whose prognostic assessment was erroneous by AJCC staging alone, where the test discriminated successfully between short-term and long-term survivors (Brunner et al., 2018).

An investigation by Amaral et al. (2020) supported the clinical validity of the Melagenix test (referred to in this study as the 11-gene expression profiling score [GEPS]). GEPS was calculated for 245 patients diagnosed with stage II cutaneous melanoma, and the scores were determined to be either “high” (median: 1.06) or “low” (median: -0.21). A significant difference in melanoma-specific survival, distant metastasis free survival, and relapse-free survival was confirmed between high-score and low-score patients. Similarly, Gambichler et al. (2021) used the 11-GEP test to determine whether a high or low score correlated with melanoma-specific survival. The authors documented decreasing probability of melanoma-specific survival with increasing 11-GEP scores. The authors further concluded that the 11-GEP is capable of identifying patients with 10-year survival probabilities above 90% (Gambichler et al., 2021).

AMBLor® test (Avero Diagnostics)

Cosgarea et al. (2022) investigated the potential contribution of melanoma paracrine transforming growth factor (TGF)- β signaling in the loss of a protein called AMBRA1 in the skin overlying a primary tumor. The study included 109 all-stage melanoma samples, in which AMBRA1 and (TGF)- β were analyzed.

Molecular Testing for Cutaneous Melanoma AHS-M2029

Another cohort had 30 samples analyzed for (TGF)- β and 42 samples analyzed for claudin-1. Results showed “increased tumoral TGF- β 2 was significantly associated with loss of peritumoral AMBRA1 ($P < 0.05$), ulceration ($P < 0.001$), AMLo high-risk status ($P < 0.05$) and metastasis ($P < 0.01$).” Although the American Joint Committee on Cancer (AJCC) staging system is an important tool, the authors noted, it leaves gaps in being unable to identify AJCC early-stage I/II tumors which are at risk of metastasis. The authors described an “urgent need” for credible prognostic information such as biomarkers in these early-stage tumors. “The combined loss of the autophagy regulatory protein autophagy-and-beclin-1 regulator 1 (AMBRA1) and epidermal differentiation marker loricrin (AMLo) in the tumour microenvironment as a prognostic biomarker for early-stage primary melanoma [is] associated with significantly increased risk of metastasis” (Cosgarea et al., 2022).

Labus et al. (2020) investigated the use of the combined immunohistochemical (IHC) expression of AMBRA1 and Loricrin in the top layer of skin as a prognostic biomarker. The study was a retrospective analysis of AMBLor performed in three different cohorts of people with AJCC stage II melanomas. A clinically validated IHC assay and semi quantitative scoring method was used to define risk groups. The results showed a loss of AMBLor in the epidermis over high risk AJCC state IIA or B tumours- that was “correlated with significant reduction in disease free survival (DFS) at 12 years to 55% compared to 89% for patients with AMBLor low risk tumours ($P = 0.015$; HR 4.83, 95% CI: 2.29-10.14).” In addition, multivariate analysis of 80 of these tumors showed reduced disease-free survival at 12 years to 68% in the AMBLor high-risk cohort compared to 83% in AMBLor low-risk groups. The assay sensitivity was 97% and had a negative predictive value of 90%. The authors concluded that “these data indicate AMBLor as a novel prognostic biomarker for patients with non-ulcerated AJCC stage II melanomas as well as companion/stratifying biomarker for adjuvant immunotherapy”(Labus et al., 2020).

Ellis et al. (2020) sought to determine the utility of AMBRA1 and loricrin (AMLo) expression as prognostic biomarkers for AJCC stage I cutaneous melanoma. This retrospective discovery study involved 76 AJCC stage I melanoma samples. Additionally, AMLo expression was correlated against clinical outcomes for up to 12 years in two independent and retrospective validation cohorts of 379 AJCC stage I melanomas. The outcome of decreased AMBRA1 expression was a 7-year disease-free survival rate (DFS) of 81.5% vs. 100% when AMBRA 1 expression was maintained ($P < 0.081$). A combined cohort analysis also revealed a DFS rate of 98.3% in the AMLo low-risk cohort vs. 85.4% in the AMLo high-risk group. The authors concluded that “loss of AMLo expression in the epidermis overlying primary AJCC stage I melanomas identifies high-risk tumour subsets independently of Breslow depth,” and advocated for these biomarkers to be integrated into risk stratification: “The integration of peritumoral epidermal AMBRA1/loricrin biomarker expression into melanoma care guidelines will facilitate more accurate, personalized risk stratification for patients with AJCC stage I melanomas” (Ellis et al., 2020).

Guidelines and Recommendations

The National Comprehensive Cancer Network

The NCCN provides common follow-up recommendations for all patients:

- “Clinical and family history can identify patients in whom multigene testing might indicate an increased genetic risk for cutaneous and uveal melanoma, astrocytoma, mesothelioma, and cancers of the breast, pancreas, and kidney. This information can guide recommendations for surveillance and early detection in appropriate patients and their relatives.
 - Consider genetic counseling referral for *p16/CDKN2A* mutation testing in the presence of 3 or more invasive cutaneous melanomas, or a mix of invasive melanoma, pancreatic cancer, and/or astrocytoma diagnoses in an individual or family.
 - Multigene panel testing that includes *CDKN2A* is recommended for patients with invasive cutaneous melanoma who have a first-degree relative diagnosed with pancreatic cancer.
 - Testing for other genes that can harbor melanoma-predisposing mutations [see Genetic Predisposition below] may be warranted” (NCCN, 2023).

Molecular Testing for Cutaneous Melanoma AHS-M2029

The NCCN includes the following as a risk factor for development of single or multiple primary melanomas:

- Genetic predisposition
 - Presence of germline mutations or polymorphisms predisposing to melanoma (eg, *CDKN2a*, *CDK4*, *MC1R*, *BAP1* [especially for uveal melanoma], *TERT*, *MITF*, *PTEN*, and potential other genes).
 - Family history of cutaneous melanoma (especially if multiple); pancreatic, renal, and/or breast cancer; astrocytoma; uveal melanoma; and/or mesothelioma” (NCCN, 2023).

Regarding indications for genetic testing using emerging molecular technologies for diagnosis and prognostication, the NCCN recommended the following:

- “The panel does not recommend *BRAF* or NGS [next generation sequencing] testing for resected stage I-II cutaneous melanoma unless it will inform clinical trial participation.
- *BRAF* mutation testing is recommended for patients with stage III at high risk for recurrence for whom *BRAF*-directed therapy may be an option.
- For initial presentation with stage IV disease or clinical recurrence, obtain tissue to ascertain alterations in *BRAF*, and in the appropriate clinical setting, *KIT* from either biopsy of the metastasis (preferred) or archival material if the patient is being considered for targeted therapy. Broader genomic profiling (e.g., larger NGS panels, *BRAF* non-V600 mutation) is recommended if feasible, especially if the test results might guide future treatment decisions or eligibility for participation in a clinical trial.
- If *BRAF* single-gene testing was the initial test performed, and is negative, clinicians should strongly consider larger NGS panels to identify other potential genetic targets (e.g., *KIT*, *BRAF* non-V600)” (NCCN, 2023).

The NCCN also recognizes *NRAS* as a relevant mutation for melanoma, noting that *NRAS* mutations are present in “approximately 15% of melanomas with chronic and intermittent sun exposure, acral surfaces, and mucosal surfaces.” Due to the low probability of overlapping targetable mutations (such as *BRAF* or *KIT*), the NCCN remarks that “the presence of an *NRAS* mutation may identify patients who will not benefit from additional molecular testing” (NCCN, 2022).

Regarding GEP tests (in a section devoted to molecular technologies for cutaneous melanoma prognostication) NCCN guidelines do not view risk stratification using commercially available GEP tests as “clinically actionable” or “reliable predictors of outcomes,” citing further research is needed.

- “Current GEP platforms do not provide clinically actionable prognostic information when combined or compared with known clinicopathologic factors (e.g., sex, age, primary tumor location, thickness, ulceration, mitotic rate, lymphovascular invasion, microsatellites, and/or SLNB status) or multivariable nomograms/risk calculators. Furthermore, the clinical utility of these tests to inform treatment recommendations and predict patient outcomes has not been established.
- “Various studies of prognostic GEP tests suggest their role as independent predictor of worse outcomes. However, GEP is not superior to Breslow thickness, ulceration, or SLN status and it remains unclear whether available GEP tests are reliably predictive of outcome across the risk spectrum of cutaneous melanoma. Validation studies on prospectively collected, independent cohorts (similar to those performed in breast cancer) are necessary to define the clinical utility of molecular prognostic GEP testing as an adjunct to AJCC staging or as part of the multidisciplinary decision-making process to guide surveillance imaging, SLBN, and adjuvant therapy.”

Providing further insight on GEP testing, while the NCCN does not recommend using GEP for prognostication, the NCCN does recognize this methodology as a potentially helpful tool to detect melanocytic neoplasms following histopathology. Additionally, apart from prognostication, the NCCN states that “Pre-diagnostic noninvasive patch testing may also be helpful to guide biopsy decisions” (NCCN, 2023).

Molecular Testing for Cutaneous Melanoma AHS-M2029

The American Academy of Dermatology (AAD)

The AAD recently published guidelines for the care and management of primary cutaneous melanoma. Regarding skin biopsies, the AAD states that while many different molecular and imaging techniques have been developed, “skin biopsy remains the first step to establish a definitive diagnosis of CM [cutaneous melanoma];” further, the guidelines also state that “Newer noninvasive techniques (eg, reflectance confocal microscopy [RCM], as well as electrical impedance spectroscopy, gene expression analysis, optical coherence tomography, and others [see the section Emerging Diagnostic Technologies]) can also be considered as these become more readily available” (Swetter et al., 2019).

The AAD notes that these guidelines highlight several gaps in research including “the clinical utility and prognostic significance of various biomarkers and molecular tests; optimal clinical situations in which to pursue multigene somatic and germline mutational analysis; and the value of ancillary molecular tests in comparison with well-established clinicopathologic predictors of outcome” (Swetter et al., 2019). It is then noted that “Efforts to standardize the histopathologic diagnosis and categorization of melanocytic neoplasms are under way to reduce the significant interobserver variability among pathologists. Ongoing advances in genomic medicine may make many of the aforementioned issues obsolete before the next AAD melanoma CPG is issued” (Swetter et al., 2019).

Regarding patients with a family history of invasive cutaneous melanoma (at least 3 affected members on 1 side of the family), “Cancer risk counseling by a qualified genetic counselor is recommended” (Swetter et al., 2019).

Finally, the AAD states “There is insufficient evidence to recommend routine molecular profiling assessment for baseline prognostication. Evidence is lacking that molecular classification should be used to alter patient management outside of current guidelines (eg. NCCN and AAD). The criteria for and the utility of prognostic molecular testing, including GEP, in aiding clinical decision making (eg. SLNB eligibility, surveillance intensity, and/or therapeutic choice) needs to be evaluated in the context of clinical study or trial” (Swetter et al., 2019).

European Society for Medical Oncology (ESMO)

The ESMO has stated that “testing for actionable mutations is mandatory in patients with resectable or unresectable stage III or stage IV (melanoma) [I, A], and is highly recommended in high-risk resected disease stage IIC but not for stage I or stage IIA-IIB. *BRAF* testing is mandatory [I, A]. If the tumor is *BRAF* wild type (WT) at the V600 locus (Class I *BRAF* mutant) sequencing the loci of the other known minor *BRAF* mutations (Class II and Class III *BRAF* mutant) to confirm WT status and testing for *NRAS* and *c-kit* mutations are recommended [II, C].” Additionally, “patients with metastatic melanoma should have metastasis (preferably) or the primary tumor screened for detection of *BRAF* V600 mutation treatment options for the first-and second-line settings include anti PD-1 antibodies (Pembrolizumab, Nivolumab), PD-1 and Ipilimumab for all patients, and BRAFi/MEKi combination for patients with *BRAF*-mutated melanoma [II, B]. (ESMO, 2019).

American Joint Committee on Cancer (AJCC)

The AJCC did not include any mention of genetic testing in the most recent 8th edition guidance on melanoma staging, though the authors do highlight that molecularly targeted antitumor therapies such as *BRAF* inhibitors are now in widespread clinical use (Gershenwald et al., 2017).

U.S. Preventive Services Task Force (USPSTF)

The USPSTF examined the utility of visual skin examination for the prevention of melanoma and found that “Only limited evidence was identified for skin cancer screening, particularly regarding potential

Molecular Testing for Cutaneous Melanoma AHS-M2029

benefit of skin cancer screening on melanoma mortality (Wernli et al., 2016).” The use of genetic tests in screening for melanoma is not mentioned.

American Society of Clinical Oncology (ASCO)

The ASCO expert panel recommends needle biopsy as the preferred diagnostic approach to clinically detected lymphadenopathy in any patient with known or suspected metastatic melanoma in regional nodes. Excisional biopsy, a more invasive approach, is not routinely required for diagnosis or characterization of melanoma. The Expert Panel recommends that *BRAF* mutation testing should be performed at time of diagnosis, but clinicians are not required to wait for the results of that testing if the decision has been made to initiate immunotherapy. If prior testing resulted in a false negative, the panel states that re-testing *RAF* status upon progression could be considered, but it would not be of value as the panel is not aware of data that supports re-testing. Regarding immunohistochemistry, the panel states “Immunohistochemistry for the *BRAF* V600E mutation has the most rapid turnaround time; however, less common mutations may be found with other methodologies such as *BRAF* gene sequencing.” Beyond *BRAF* testing though, the Expert Panel did not make formal recommendations about other genetic testing (e.g., *NRAS* and *KIT*), given insufficient data regarding the efficacy of inhibitors targeting these pathways (Seth et al., 2020).

The ASCO notes that some cancer treatments most commonly used in metastatic melanoma patients (e.g., *BRAF* inhibitors) may increase the risk of secondary skin cancers; hence, clinicians should be aware of this potential adverse effect when considering such therapeutic agents (Alberg et al., 2020).

European Dermatology Forum (EDF), the European Association of DermatoOncology (EADO), and the European Organization of Research and Treatment of Cancer (EORTC)

A panel of experts from EDF, EADO, and EORTC recommend that *BRAF* mutation testing be required “in patients with distant metastasis or non-resectable regional metastasis to identify those who are eligible to receive treatment with combined *BRAF* and MEK inhibitors, and in resected high-risk stage III melanoma patients in the adjuvant setting (Garbe et al., 2020).” Mutational analysis should be performed on metastatic tissue, either distance or regional, or on the primary tumor if metastatic tissue is not feasible. Though there may be a discrepancy rate in the *BRAF* status between the primary versus metastatic melanoma lesions, the authors note that there is “a high concordance rate in the *BRAF* status..between primary and metastatic melanoma lesions” (Garbe et al., 2022). Regarding next generation sequencing, the panel claims that it may help in identifying genetic alterations and, when limited to currently actionable genes like *BRAF*, *NRAS*, and *KIT* in a single experiment, may be cost and time effective in the clinical setting (Garbe et al., 2022).

The guideline also commented on *NRAS*, stating that *NRAS* mutations are present in 15-20% of melanoma cases and are almost always mutually exclusive with *BRAF* mutations. The guideline remarked that *NRAS* status may inform clinicians regarding the *BRAF* status, although *NRAS*-based treatments are still under investigation.

Regarding *c-KIT*, the guideline states that acral and mucosal melanomas should initially be tested for *BRAF* and *NRAS* mutations; if both genes were found to be wild-type, the sample should be tested for *c-KIT* (Garbe et al., 2020).

Tumor mutational burden (TMB) is highlighted in the guideline as a predictive tool for response to immune checkpoint inhibitors in melanoma. The panel recognized a study that demonstrated the positive association of a high TMB value with response and overall survival of melanoma patients treated with a combination of ipilimumab and nivolumab (Forschner et al., 2019).

Finally, the guideline comments on GEP tests like DecisionDx-Melanoma™ and their increased use by clinicians. While a growing pool of literature describes the potential application of GEP tests in the management of various cancers, the authors state “additional data are required in melanoma to address if they provide independent prognostic information in addition to known clinicopathologic factors before they can be integrated into clinical decision-making” (Garbe et al., 2022).

Molecular Testing for Cutaneous Melanoma AHS-M2029

Melanoma Prevention Working Group (MPWG)

This Working Group published a guideline regarding prognostic gene expression profile [GEP] testing for cutaneous melanoma. Although the Group is “optimistic” about the future use of prognostic GEP testing to “improve risk stratification and enhance clinical decision-making”, it states that “more evidence is needed to support GEP testing to inform recommendations regarding SLNB [sentinel lymph node biopsy], intensity of follow-up or imaging surveillance, and postoperative adjuvant therapy.” Overall, the Group remarks that “there are insufficient data to support routine use of currently available prognostic GEP tests to inform management of patients with CM [cutaneous melanoma]” (Grossman et al., 2020).

Skin Cancer Prevention Working Group (SCPWG)

An expert consensus on the appropriate use of prognostic gene expression profiling tests for the management of cutaneous melanoma was published in 2022. The SCPWG reviewed literature for DecisionDx-Melanoma, the 11-GEP (Melagenix) test by NeraCare, and the 8-GEP (CP-GEP; Merlin Assay) by SkylineDx. The eight-person consensus panel of dermatologists unanimously agreed that:

- “Gene expression profile (GEP) tests are validated, reproducible, and consistent across studies;
- Integrating GEPs into AJCC8 and NCCN models can improve prognostic accuracy;
- Prognostic GEP tests can inform clinical decision making regarding sentinel lymph node biopsies;
- Incorporating GEPs into real-world clinical management has positively impacted patient outcomes”

Additionally, 7 out of 8 panelists agreed that “the AJCC8 and NCCN prognostic model, which does not account for genomic expression, may not optimize melanoma prognostic assessment” (Farberg et al., 2022).

The National Society for Cutaneous Medicine (NSCM)

An expert panel gathered to develop consensus-based guidelines on the appropriate use criteria for the integration of diagnostic and prognostic GEP assays into the management of cutaneous malignant melanoma. These guidelines include recommendations for the 2-GEP test (DermTech PLA), the 23-GEP test (myPath®), and the 31-GEP test (DecisionDx-Melanoma). Regarding use of the 31-GEP test, a total of 14 recommendations were considered:

A-strength recommendation:

- “Use of the 31-GEP test to aid in the management of patients who are SLNBx negative”

B-strength recommendation:

- “Integration of 31-GEP results into the decision to adjust follow-up frequency
- Integration of 31-GEP results into the decision to order adjunctive imaging studies
- Integration of 31-GEP results into management of patients with T1a tumors with Breslow depth <0.8 mm and other adverse prognostic factors
- Integration of 31-GEP results into management of patients with T1 or T2 tumors who are sentinel lymph node biopsy (SLNBx) eligible
- Integration of 31-GEP results into management of patients with T1b tumors
- Integration of 31-GEP results into management of patients with T2 tumors
- Integration of 31-GEP results into management of patients with a low-risk category based on traditional AJCC factors”

C-strength recommendation:

Molecular Testing for Cutaneous Melanoma AHS-M2029

- “Integration of 31-GEP results into the assessment of prognosis and management options for patients with T1a tumors with a positive deep margin
- Integration of 31-GEP results into the assessment of prognosis and management options for patients with T1b tumors with a positive deep margin
- Integration of 31-GEP results to for risk-stratification of patients in clinical trials
- Use of 31-GEP results as a criterion for eligibility for a chemotherapy regimen
- T4 disease as a contraindication for use of the 31-GEP test
- Melanoma in situ as a contraindication for use of the 31-GEP test”

7 recommendations for use of the 2-GEP test were considered as follows:

B-strength recommendation:

- “Cases in which patients present with atypical lesions requiring additional assessment beyond inspection in order to aid in the biopsy decision”

C-strength recommendations:

- Cases in which patients refuse surgical biopsy
- Cases in which suspicious lesions present in cosmetically sensitive areas
- Cases in which patients have undergone numerous biopsies in the past and wish to avoid additional biopsy procedures
- Scenarios in which patients have a relative contraindication to surgical biopsy
- Patients with increased risk of infection (e.g. immunosuppressed patients)
- Patients with heightened risk of poor wound healing

Expert consensus was reached on 8 recommendations for use of the 23-GEP test:

A-strength recommendation:

- “Differentiation of a nevus from melanoma in an adult patient when the morphologic findings are ambiguous by light microscopic parameters”

B-strength recommendations:

- “Cases with pathology suggestive or suspicious for nevoid melanoma vs. benign melanocytic nevus
- Cases with pathology suggestive or suspicious for atypical Spitz tumor vs. Spitzoid melanoma
- Instances in which pathology is suggestive of or suspicious for severely atypical compound melanocytic proliferation vs melanoma on cosmetically sensitive areas”

C-strength recommendations:

- “Instances of pathology suggestive or suspicious for melanoma arising from within a severely dysplastic nevus
- Cases of pathology suggestive or suspicious for benign vs. malignant blue nevus
- To differentiate suspicious lesions in low-risk populations
- Upon request from the referring dermatologist following an ambiguous pathology report” (Berman et al., 2019)

In a separate editorial published by the NSCM discussing the management of pigmented lesions, the authors conclude that “[u]se of the 2-GEP [DermTech PLA] improves patient care and improves health outcomes by using genomic assessments to minimize avoidable surgical procedures on benign lesions, enriching the population of melanomas within biopsied lesions, and decreasing downstream costs of

Molecular Testing for Cutaneous Melanoma AHS-M2029

late-stage melanoma diagnoses. For all of these reasons, creating broad access and coverage to transformative diagnostic technologies such as the 2-GEP is in the best interest of our colleagues, the healthcare system, and most importantly, our patients” (Cockerell et al., 2022).

State and Federal Regulations, as applicable

Food and Drug Administration (FDA)

The FDA-approvals for all the BRAF-targeted therapies include the requirement that BRAF mutation testing be performed by an FDA-approved test.

On August 17, 2011 the U.S. Food and Drug Administration (FDA) announced the approval of Zelboraf (vemurafenib) for unresectable or metastatic melanoma with oncogenic BRAF mutation (V600E). The Cobas® 4800 BRAF V600 Mutation Test was approved as the companion diagnostic for vemurafenib (Bollag et al., 2012).

Dabrafenib was FDA-approved in May 2013 for the treatment of patients with unresectable or metastatic melanoma with *BRAF* V600E mutation, as detected by an FDA-approved test. Dabrafenib is specifically not indicated for the treatment of patients with wild-type *BRAF* melanoma (Tafinlar (dabrafenib), Jan 2014).

Trametinib was FDA-approved in May 2013 for the treatment of patients with unresectable or metastatic melanoma with BRAF V600E or V600K mutations, as detected by an FDA-approved test. Trametinib is specifically not indicated for the treatment of patients previously treated with BRAF inhibitor therapy (GlaxoSmithKline. Mekinist Aug 2014).

The companion diagnostic test coapproved for both dabrafenib and trametinib is the THxID™ BRAF Kit manufactured by bioMérieux. The kit is intended “as an aid in selecting melanoma patients whose tumors carry the BRAF V600E mutation for treatment with dabrafenib and as an aid in selecting melanoma patients whose tumors carry the BRAF V600E or V600K mutation for treatment with trametinib” (Genentech, Inc. Zelboraf® March, 2014).

The FDA approved the use of the Oncomine Dx target test NGS panel for somatic or germline variants, which includes the BRAF V600E mutation for consideration with dabrafenib therapy as one of the gene variants (Life Technologies Corporation, approved in June 2017).

The FDA approved the FoundationOne CDx NGS panel in November 2017, which does include both the V600E and V600K mutation for possible dabrafenib or vemurafenib therapy. (Foundation Medicine, Inc.)

The FDA approved the use of Therascreen BRAF V600E RGQ PCR Kit in April 2020, a real time PCR test that detects *BRAF V600E* mutations. The Therascreen BRAF V600E RGQ PCR Kit is for use on the Rotor-Gene Q MDx (US) instrument.

New therapies for melanoma continue to receive FDA approval, and the National Cancer Institute summarizes them here: <https://www.cancer.gov/about-cancer/treatment/drugs/melanoma> (NCI, 2023a).

Many labs have developed specific tests that they must validate and perform in house. These laboratory-developed tests (LDTs) are regulated by the Centers for Medicare and Medicaid (CMS) as high-complexity tests under the Clinical Laboratory Improvement Amendments of 1988 (CLIA '88). LDTs are not approved or cleared by the U. S. Food and Drug Administration; however, FDA clearance or approval is not currently required for clinical use.

Billing/Coding/Physician Documentation Information

This policy may apply to the following codes. Inclusion of a code in this section does not guarantee that it will be reimbursed. For further information on reimbursement guidelines, please see Administrative

Molecular Testing for Cutaneous Melanoma AHS-M2029

Policies on the Blue Cross Blue Shield of North Carolina web site at www.bcsnc.com. They are listed in the Category Search on the Medical Policy search page.

Applicable service codes: 81167, 81210, 81216, 81217, 81272, 81273, 81311, 81345, 81404, 81479, 81529, 0089U, 0090U, 0314U, 0387U

BCBSNC may request medical records for determination of medical necessity. When medical records are requested, letters of support and/or explanation are often useful, but are not sufficient documentation unless all specific information needed to make a medical necessity determination is included.

Scientific Background and Reference Sources

For Policy Titled: BRAF Genetic Testing in Patients with Melanoma

For Policy Re-Titled: Molecular Testing for Cutaneous Melanoma

Alberg, A. J., LoConte, N. K., Foxhall, L., Weinstock, M. A., Gomez, S. L., Francisco, M., Moushey, E. A., & Gershenwald, J. E. (2020). American Society of Clinical Oncology Policy Statement on Skin Cancer Prevention. *JCO Oncol Pract*, 16(8), 490-499. <https://doi.org/10.1200/JOP.19.00585>

Alrabadi, N., Gibson, N., Curless, K., Cheng, L., Kuhar, M., Chen, S., . . . Alomari, A. K. (2019). Detection of driver mutations in BRAF can aid in diagnosis and early treatment of dedifferentiated metastatic melanoma. *Mod Pathol*, 32(3), 330-337. <https://doi.org/10.1038/s41379-018-0161-0>

Amaria, R. N., Prieto, P. A., Tetzlaff, M. T., Reuben, A., Andrews, M. C., Ross, M. I., . . . Wargo, J. A. (2018). Neoadjuvant plus adjuvant dabrafenib and trametinib versus standard of care in patients with high-risk, surgically resectable melanoma: a single-centre, open-label, randomised, phase 2 trial. *Lancet Oncol*, 19(2), 181-193. [https://doi.org/10.1016/S1470-2045\(18\)30015-9](https://doi.org/10.1016/S1470-2045(18)30015-9)

AMLo Biosciences. (2023). *Avero Diagnostics launches melanoma progression test with AMLo*. Retrieved April 24, 2023 from <https://usa.amlo-biosciences.com/avero-diagnostics-launches-melanoma-progression-test-with-amlo/>

Aoude, L. G., Bonazzi, V. F., Brosda, S., Patel, K., Koufariotis, L. T., Oey, H., Nones, K., Wood, S., Pearson, J. V., Lonie, J. M., Arneil, M., Atkinson, V., Smithers, B. M., Waddell, N., Barbour, A. P., Aoude, L. G., Bonazzi, V. F., Brosda, S., Patel, K., . . . Barbour, A. P. (2020). Pathogenic germline variants are associated with poor survival in stage III/IV melanoma patients. *Sci Rep*, 10(1), 17687. <https://doi.org/10.1038/s41598-020-74956-3>

Atkins, M. B., Kunkel, L., Sznol, M., & Rosenberg, S. A. (2000). High-dose recombinant interleukin-2 therapy in patients with metastatic melanoma: long-term survival update. *Cancer J Sci Am*, 6 Suppl 1, S11-14. <http://dx.doi.org/>

Bellomo, D., Arias-Mejias, S. M., Ramana, C., Heim, J. B., Quattrocchi, E., Sominidi-Damodaran, S., Bridges, A. G., Lehman, J. S., Hieken, T. J., Jakub, J. W., Pittelkow, M. R., DiCaudo, D. J., Pockaj, B. A., Sluzevich, J. C., Cappel, M. A., Bagaria, S. P., Perniciaro, C., Tjien-Fooh, F. J., van Vliet, M. H., . . . Meves, A. (2020). Model Combining Tumor Molecular and Clinicopathologic Risk Factors Predicts Sentinel Lymph Node Metastasis in Primary Cutaneous Melanoma. *JCO Precis Oncol*, 4, 319-334. <https://doi.org/10.1200/po.19.00206>

Berman, B., Ceilley, R., COckerell, C., Ferria, L., High, W., Lebwohl, M., . . . Rigel, D. (2019). Appropriate Use Criteria for the Integration of Diagnostic and Prognostic Gene Expression Profile Assays into the Management of Cutaneous Malignant Melanoma: An Expert Panel Consensus-Based Modified Delphi Process Assessment. *SKIN The Journal of Cutaneous Medicine*, 3(5). <https://www.jofskin.org/index.php/skin/article/view/663>

Molecular Testing for Cutaneous Melanoma AHS-M2029

- Bertolotto, C., Lesueur, F., Giuliano, S., Strub, T., de Lichy, M., Bille, K., Dessen, P., d'Hayer, B., Mohamdi, H., Remenieras, A., Maubec, E., de la Fouchardiere, A., Molinie, V., Vabres, P., Dalle, S., Poulalhon, N., Martin-Denavit, T., Thomas, L., Andry-Benzaquen, P., . . . Bressac-de Paillerets, B. (2011). A SUMOylation-defective MITF germline mutation predisposes to melanoma and renal carcinoma. *Nature*, *480*(7375), 94-98. <https://doi.org/10.1038/nature10539>
- Berwick, M., Orlow, I., Hummer, A. J., Armstrong, B. K., Krickler, A., Marrett, L. D., Millikan, R. C., Gruber, S. B., Anton-Culver, H., Zanetti, R., Gallagher, R. P., Dwyer, T., Rebbeck, T. R., Kanetsky, P. A., Busam, K., From, L., Mujumdar, U., Wilcox, H., Begg, C. B., & Group, G. E. M. S. (2006). The prevalence of CDKN2A germ-line mutations and relative risk for cutaneous malignant melanoma: an international population-based study. *Cancer Epidemiol Biomarkers Prev*, *15*(8), 1520-1525. <https://doi.org/10.1158/1055-9965.EPI-06-0270>
- Bishop, D. T., Demenais, F., Goldstein, A. M., Bergman, W., Bishop, J. N., Bressac-de Paillerets, B., Chompret, A., Ghiorzo, P., Gruis, N., Hansson, J., Harland, M., Hayward, N., Holland, E. A., Mann, G. J., Mantelli, M., Nancarrow, D., Platz, A., Tucker, M. A., & Melanoma Genetics, C. (2002). Geographical variation in the penetrance of CDKN2A mutations for melanoma. *J Natl Cancer Inst*, *94*(12), 894-903. <https://www.ncbi.nlm.nih.gov/pubmed/12072543>
- Bollag, G., Tsai, J., Zhang, J., Zhang, C., Ibrahim, P., Nolop, K., & Hirth, P. (2012). Vemurafenib: the first drug approved for BRAF-mutant cancer. *Nat Rev Drug Discov*, *11*(11), 873-886. <https://doi.org/10.1038/nrd3847>
- Bollard, S. M., Casalou, C., & Potter, S. M. (2021). Gene expression profiling in melanoma: A view from the clinic. *Cancer Treat Res Commun*, *29*, 100447. <https://doi.org/10.1016/j.ctarc.2021.100447>
- Brunner, G., Heinecke, A., Falk, T. M., Ertas, B., Blodorn-Schlicht, N., Schulze, H. J., Suter, L., Atzpodien, J., & Berking, C. (2018). A Prognostic Gene Signature Expressed in Primary Cutaneous Melanoma: Synergism With Conventional Staging. *JNCI Cancer Spectr*, *2*(3), pky032. <https://doi.org/10.1093/jncics/pky032>
- Brunner, G., Reitz, M., Heinecke, A., Lippold, A., Berking, C., Suter, L., & Atzpodien, J. (2013). A nine-gene signature predicting clinical outcome in cutaneous melanoma. *J Cancer Res Clin Oncol*, *139*(2), 249-258. <https://doi.org/10.1007/s00432-012-1322-z>
- Bubien, V., Bonnet, F., Brouste, V., Hoppe, S., Barouk-Simonet, E., David, A., Edery, P., Bottani, A., Layet, V., Caron, O., Gilbert-Dussardier, B., Delnatte, C., Dugast, C., Fricker, J. P., Bonneau, D., Sevenet, N., Longy, M., Caux, F., & French Cowden Disease, N. (2013). High cumulative risks of cancer in patients with PTEN hamartoma tumour syndrome. *J Med Genet*, *50*(4), 255-263. <https://doi.org/10.1136/jmedgenet-2012-101339>
- Burjanivova, T., Malicherova, B., Grendar, M., Minarikova, E., Dusenka, R., Vanova, B., . . . Plank, L. (2019). Detection of BRAFV600E Mutation in Melanoma Patients by Digital PCR of Circulating DNA. *Genet Test Mol Biomarkers*, *23*(4), 241-245. <https://doi.org/10.1089/gtmb.2018.0193>
- Burke, L. S., Hyland, P. L., Pfeiffer, R. M., Prescott, J., Wheeler, W., Mirabello, L., Savage, S. A., Burdette, L., Yeager, M., Chanock, S., De Vivo, I., Tucker, M. A., Goldstein, A. M., & Yang, X. R. (2013). Telomere length and the risk of cutaneous malignant melanoma in melanoma-prone families with and without CDKN2A mutations. *PLoS One*, *8*(8), e71121. <https://doi.org/10.1371/journal.pone.0071121>
- Bush, J. W., Hunt, E. L., & Fraga, G. R. (2015). BAM! Utilizing the frequency of Benign, Atypical and Malignant diagnoses for quality improvement in the histopathologic diagnosis of melanocytic neoplasms. *J Cutan Pathol*, *42*(10), 712-716. <https://doi.org/10.1111/cup.12566>

Molecular Testing for Cutaneous Melanoma AHS-M2029

Castle Biosciences. (2023a). DecisionDx®-Melanoma Overview.

<https://castlebiosciences.com/products/decisiondx-melanoma/>

Castle Biosciences. (2023b). myPath® Melanoma. <https://castlebiosciences.com/products/mypath-melanoma-and-diffdx-melanoma/>

CastleBiosciences. (2023). *DecisionDx®-Melanoma Overview*.

<https://castlebiosciences.com/products/decisiondx-melanoma/>

Chiaravalloti, A. J., Jinna, S., Kerr, P. E., Whalen, J., & Grant-Kels, J. M. (2018). A deep look into thin melanomas: What's new for the clinician and the impact on the patient. *Int J Womens Dermatol*, 4(3), 119-121. <https://doi.org/10.1016/j.ijwd.2018.01.003>

Clarke, L. E., Flake, D. D., 2nd, Busam, K., Cockerell, C., Helm, K., McNiff, J., . . . Wenstrup, R. J. (2017). An independent validation of a gene expression signature to differentiate malignant melanoma from benign melanocytic nevi. *Cancer*, 123(4), 617-628. <https://doi.org/10.1002/cncr.30385>

Clarke, L. E., Mabey, B., Flake, D. D., Meek, S., Cassarino, D. S., Duncan, L. M., High, W. A., Napekoski, K. M., Prieto, V. G., Tetzlaff, M. T., Vitale, P., & Elder, D. E. (2020). Clinical validity of a gene expression signature in diagnostically uncertain neoplasms. *Per Med*, 17(5), 361-371. <https://doi.org/10.2217/pme-2020-0048>

Clarke, L.E., Warf, M. B., Flake, D. D., 2nd, Hartman, A. R., Tahan, S., Shea, C. R., Gerami, P., Messina, J., Florell, S. R., Wenstrup, R. J., Rushton, K., Roundy, K. M., Rock, C., Roa, B., Kolquist, K. A., Gutin, A., Billings, S., & Leachman, S. (2015). Clinical validation of a gene expression signature that differentiates benign nevi from malignant melanoma. *J Cutan Pathol*, 42(4), 244-252. <https://doi.org/10.1111/cup.12475>

CMGL. (2014). Molecular Genetic Testing For BRAF Mutations.

<file:///C:/Users/AHCS6886/OneDrive%20-%20AVALON%20HEALTH%20SERVICES,%20LLC/Downloads/BRAF%20Mutation%20Analysis%20Test%20Information.pdf>

Cockerell, C., Ceilley, R., Lebwohl, M., & Rigel, D. (2022). Using Genomics to Improve Pigmented Lesion Management & Health Outcomes. *SKIN The Journal of Cutaneous Medicine*, 6(2).

Cockerell, C., Tschen, J., Billings, S. D., Evans, B., Brown, K., Rock, C., & Clarke, L. E. (2017). The influence of a gene-expression signature on the treatment of diagnostically challenging melanocytic lesions. *Per Med*, 14(2), 123-130. <https://doi.org/10.2217/pme-2016-0097>

Colombino, M., Rozzo, C., Paliogiannis, P., Casula, M., Manca, A., Doneddu, V., Fedeli, M. A., Sini, M. C., Palomba, G., Pisano, M., Ascierio, P. A., Caracò, C., Lissia, A., Cossu, A., & Palmieri, G. (2020). Comparison of BRAF Mutation Screening Strategies in a Large Real-Life Series of Advanced Melanoma Patients. *J Clin Med*, 9(8). <https://doi.org/10.3390/jcm9082430>

Corean, J. L. E., George, T. I., Patel, J. L., & Li, K. D. (2019). Bone marrow findings in metastatic melanoma, including role of BRAF immunohistochemistry. *Int J Lab Hematol*, 41(4), 550-560. <http://doi.org/10.1111/ijlh.13051>

Cosgarea, I., McConnell, A. T., Ewen, T., Tang, D., Hill, D. S., Anagnostou, M., Elias, M., Ellis, R. A., Murray, A., Spender, L. C., Giglio, P., Gagliardi, M., Greenwood, A., Piacentini, M., Inman, G. J., Fimia, G. M., Corazzari, M., Armstrong, J. L., & Lovat, P. E. (2022). Melanoma secretion of transforming growth factor- β 2 leads to loss of epidermal AMBRA1 threatening epidermal integrity and facilitating tumour ulceration*. *British Journal of Dermatology*, 186(4), 694-704. <https://doi.org/https://doi.org/10.1111/bjd.20889>

Molecular Testing for Cutaneous Melanoma AHS-M2029

- Cree, I. A., Deans, Z., Ligtenberg, M. J. L., Normanno, N., Edsjö, A., Rouleau, E., Van Krieken, J. H. (2014). Guidance for laboratories performing molecular pathology for cancer patients. *Journal of Clinical Pathology*, 67(11), 923-931. <https://doi.org/10.1136/jclinpath-2014-202404>
- Cust, A. E., Harland, M., Makalic, E., Schmidt, D., Dowty, J. G., Aitken, J. F., Agha-Hamilton, C., Armstrong, B. K., Barrett, J. H., Chan, M., Chang, Y. M., Gascoyne, J., Giles, G. G., Holland, E. A., Kefford, R. F., Kukulizch, K., Lowery, J., Randerson-Moor, J. A., Schmid, H., . . . Jenkins, M. A. (2011). Melanoma risk for CDKN2A mutation carriers who are relatives of population-based case carriers in Australia and the UK. *J Med Genet*, 48(4), 266-272. <https://doi.org/10.1136/jmg.2010.086538>
- Davies, M. A., & Gershenwald, J. E. (2011). Targeted therapy for melanoma: a primer. *Surg Oncol Clin N Am*, 20(1), 165-180. <https://pubmed.ncbi.nlm.nih.gov/21111965/>
- Delaunay, J., Martin, L., Bressac-de Paillerets, B., Duru, G., Ingster, O., & Thomas, L. (2017). Improvement of Genetic Testing for Cutaneous Melanoma in Countries With Low to Moderate Incidence: The Rule of 2 vs the Rule of 3. *JAMA Dermatol*, 153(11), 1122-1129. <https://doi.org/10.1001/jamadermatol.2017.2926>
- DermTech. (2023a). The DermTech Melanoma Test. <https://dermtech.com/how-it-works/>
- DermTech. (2023b). PLA Pigmented Lesion Assay. <https://dermtech.com/physicians/>
- Elmore, J. G., Barnhill, R. L., Elder, D. E., Longton, G. M., Pepe, M. S., Reisch, L. M., Carney, P. A., Titus, L. J., Nelson, H. D., Onega, T., Tosteson, A. N. A., Weinstock, M. A., Knezevich, S. R., & Piepkorn, M. W. (2017). Pathologists' diagnosis of invasive melanoma and melanocytic proliferations: observer accuracy and reproducibility study. *BMJ*, 357, j2813. <https://doi.org/10.1136/bmj.j2813>
- ESMO. (2019). Cutaneous melanoma: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. *Annals of Oncology*. <https://www.esmo.org/Guidelines/Melanoma/Cutaneous-Melanoma>
- Farberg, A. S., Marson, J. W., Glazer, A., Litchman, G. H., Svoboda, R., Winkelmann, R. R., Brownstone, N., Rigel, D. S., & Skin Cancer Prevention Working, G. (2022). Expert Consensus on the Use of Prognostic Gene Expression Profiling Tests for the Management of Cutaneous Melanoma: Consensus from the Skin Cancer Prevention Working Group. *Dermatol Ther (Heidelb)*, 12(4), 807-823. <https://doi.org/10.1007/s13555-022-00709-x>
- Farmer, E. R., Gonin, R., & Hanna, M. P. (1996). Discordance in the histopathologic diagnosis of melanoma and melanocytic nevi between expert pathologists. *Hum Pathol*, 27(6), 528-531. <https://www.ncbi.nlm.nih.gov/pubmed/8666360>
- Ferris, L. K., Gerami, P., Skelsey, M. K., Peck, G., Hren, C., Gorman, C., Frumento, T., & Siegel, D. M. (2018). Real-world performance and utility of a noninvasive gene expression assay to evaluate melanoma risk in pigmented lesions. *Melanoma Res*, 28(5), 478-482. <https://doi.org/10.1097/CMR.0000000000000478>
- Ferris, L. K., Jansen, B., Ho, J., Busam, K. J., Gross, K., Hansen, D. D., Alsobrook, J. P., 2nd, Yao, Z., Peck, G. L., & Gerami, P. (2017). Utility of a Noninvasive 2-Gene Molecular Assay for Cutaneous Melanoma and Effect on the Decision to Biopsy. *JAMA Dermatol*, 153(7), 675-680. <https://doi.org/10.1001/jamadermatol.2017.0473>
- Ferris, L. K., Moy, R. L., Gerami, P., Sligh, J. E., Jansen, B., Yao, Z., & Cockerell, C. J. (2019). Noninvasive Analysis of High-Risk Driver Mutations and Gene Expression Profiles in Primary Cutaneous Melanoma. *J Invest Dermatol*, 139(5), 1127-1134. <https://doi.org/10.1016/j.jid.2018.10.041>

Molecular Testing for Cutaneous Melanoma AHS-M2029

Ferris, L. K., Rigel, D. S., Siegel, D. M., Skelsey, M. K., Peck, G. L., Hren, C., Gorman, C., Frumento, T., Jansen, B., Yao, Z., Rock, J., Knezevich, S. R., & Cockerell, C. J. (2019). Impact on clinical practice of a non-invasive gene expression melanoma rule-out test: 12-month follow-up of negative test results and utility data from a large US registry study. *Dermatol Online J*, 25(5).

<https://escholarship.org/uc/item/61w6h7mn>

Flaherty, K. T., Puzanov, I., Kim, K. B., Ribas, A., McArthur, G. A., Sosman, J. A., O'Dwyer, P. J., Lee, R. J., Grippo, J. F., Nolop, K., & Chapman, P. B. (2010). Inhibition of mutated, activated BRAF in metastatic melanoma. *N Engl J Med*, 363(9), 809-819. <https://doi.org/10.1056/NEJMoa1002011>

Forschner, A., Battke, F., Hadaschik, D., Schulze, M., Weissgraeber, S., Han, C. T., Kopp, M., Frick, M., Klumpp, B., Tietze, N., Amaral, T., Martus, P., Sinnberg, T., Eigentler, T., Keim, U., Garbe, C., Docker, D., & Biskup, S. (2019). Tumor mutation burden and circulating tumor DNA in combined CTLA-4 and PD-1 antibody therapy in metastatic melanoma - results of a prospective biomarker study. *J Immunother Cancer*, 7(1), 180. <https://doi.org/10.1186/s40425-019-0659-0>

Froyen, G., Broekmans, A., Hillen, F., Pat, K., Achten, R., Mebis, J., Rummens, J. L., Willemse, J., & . . . Maes, B. (2016). Validation and Application of a Custom-Designed Targeted Next-Generation Sequencing Panel for the Diagnostic Mutational Profiling of Solid Tumors. In *PLoS One* (Vol. 11). <https://doi.org/10.1371/journal.pone.0154038>

Fulgent. (2023). *Melanoma Comprehensive Panel*.

<https://www.fulgentgenetics.com/comprehensivecancer-melanoma>

Gambichler, T., Tsagoudis, K., Kiecker, F., Reinhold, U., Stockfleth, E., Hamscho, R., Egberts, F., Hauschild, A., Amaral, T., & Garbe, C. (2021). Prognostic significance of an 11-gene RNA assay in archival tissue of cutaneous melanoma stage I-III patients. *Eur J Cancer*, 143, 11-18.

<https://doi.org/10.1016/j.ejca.2020.10.016>

Garbe, C., Amaral, T., Peris, K., Hauschild, A., Arenberger, P., Basset-Seguín, N., Bastholt, L., Bataille, V., Del Marmol, V., Dreno, B., Fargnoli, M. C., Forsea, A. M., Grob, J. J., Holler, C., Kaufmann, R., Kelleners-Smeets, N., Lallas, A., Lebbe, C., Lytvynenko, B., . . . Treatment of, C. (2022). European consensus-based interdisciplinary guideline for melanoma. Part 1: Diagnostics: Update 2022. *Eur J Cancer*, 170, 236-255. <https://doi.org/10.1016/j.ejca.2022.03.008>

Garbe, C., Amaral, T., Peris, K., Hauschild, A., Arenberger, P., Bastholt, L., V., Del Marmol, V., Dréno, B., Fargnoli, M. C., Grob, J. J., Höller, C., Kaufmann, R., Lallas, A., Lebbé, C., Malvehy, J., Middleton, M., Moreno-Ramirez, D., Pellacani, G., Eggermont, A. M. M. (2020). European consensus-based interdisciplinary guideline for melanoma. Part 1: Diagnostics - Update 2019. *Eur J Cancer*, 126, 141-158. <https://doi.org/10.1016/j.ejca.2019.11.014>

Gastman, B. R., Zager, J. S., Messina, J. L., Cook, R. W., Covington, K. R., Middlebrook, B., . . . Vetto, J. T. (2019). Performance of a 31-gene expression profile test in cutaneous melanomas of the head and neck. *Head Neck*, 41(4), 871-879. <https://doi.org/10.1002/hed.25473>

GeneDx. (2023). *Melanoma Panel*. <https://www.genedx.com/tests/detail/melanoma-panel-83>

Gerami, P., Busam, K., Cochran, A., Cook, M. G., Duncan, L. M., Elder, D. E., Fullen, D. R., Guitart, J., LeBoit, P. E., Mihm, M. C., Jr., Prieto, V. G., Rabkin, M. S., Scolyer, R. A., Xu, X., Yun, S. J., Obregon, R., Yazdan, P., Cooper, C., Weitner, B. B., Barnhill, R. L. (2014). Histomorphologic assessment and interobserver diagnostic reproducibility of atypical spitzoid melanocytic neoplasms with long-term follow-up. *Am J Surg Pathol*, 38(7), 934-940. <https://doi.org/10.1097/PAS.0000000000000198>

Molecular Testing for Cutaneous Melanoma AHS-M2029

- Gerami, P., Yao, Z., Polsky, D., Jansen, B., Busam, K., Ho, J., Martini, M., & Ferris, L. K. (2017a). Development and validation of a noninvasive 2-gene molecular assay for cutaneous melanoma. *J Am Acad Dermatol*, 76(1), 114-120 e112. <https://doi.org/10.1016/j.jaad.2016.07.038>
- Gershenwald, J. E., Scolyer, R. A., Hess, K. R., Sondak, V. K., Long, G. V., Ross, M. I., Lazar, A. J., Faries, M. B., Kirkwood, J. M., McArthur, G. A., Haydu, L. E., Eggermont, A. M. M., Flaherty, K. T., Balch, C. M., Thompson, J. F., for members of the American Joint Committee on Cancer Melanoma Expert, P., the International Melanoma, D., & Discovery, P. (2017). Melanoma staging: Evidence-based changes in the American Joint Committee on Cancer eighth edition cancer staging manual. *CA Cancer J Clin*, 67(6), 472-492. <https://doi.org/10.3322/caac.21409>
- Ghate, S., Ionescu-Ittu, R., Burne, R., Ndife, B., Laliberte, F., Nakasato, A., & Duh, M. S. (2019). Patterns of treatment and BRAF testing with immune checkpoint inhibitors and targeted therapy in patients with metastatic melanoma presumed to be BRAF positive. *Melanoma Res*, 29(3), 301-310. <https://doi.org/10.1097/cmr.0000000000000504>
- Goldstein, A. M., Chan, M., Harland, M., Hayward, N. K., Demenais, F., Bishop, D. T., Azizi, E., Bergman, W., Bianchi-Scarra, G., Bruno, W., Calista, D., Albright, L. A., Chaudru, V., Chompret, A., Cuellar, F., Elder, D. E., Ghiorzo, P., Gillanders, E. M., Gruis, N. A., . . . Melanoma Genetics, C. (2007). Features associated with germline CDKN2A mutations: a GenoMEL study of melanoma-prone families from three continents. *J Med Genet*, 44(2), 99-106. <https://doi.org/10.1136/jmg.2006.043802>
- Goldstein, A. M., & Tucker, M. A. (2001). Genetic epidemiology of cutaneous melanoma: a global perspective. *Arch Dermatol*, 137(11), 1493-1496. <https://www.ncbi.nlm.nih.gov/pubmed/11708953>
- Greenhaw, B. N., Covington, K. R., Kurley, S. J., Yeniay, Y., Cao, N. A., Plasseraud, K. M., Cook, R. W., Hsueh, E. C., Gastman, B. R., & Wei, M. L. (2020). Molecular risk prediction in cutaneous melanoma: a meta-analysis of the 31-gene expression profile prognostic test in 1,479 patients. *J Am Acad Dermatol*. <https://doi.org/10.1016/j.jaad.2020.03.053>
- Grossman, D., Okwundu, N., Bartlett, E. K., Marchetti, M. A., Othus, M., Coit, D. G., Hartman, R. I., Leachman, S. A., Berry, E. G., Korde, L., Lee, S. J., Bar-Eli, M., Berwick, M., Bowles, T., Buchbinder, E. I., Burton, E. M., Chu, E. Y., Curiel-Lewandrowski, C., Curtis, J. A., Swetter, S. M. (2020). Prognostic Gene Expression Profiling in Cutaneous Melanoma: Identifying the Knowledge Gaps and Assessing the Clinical Benefit. *JAMA Dermatology*, 156(9), 1004-1011. <https://doi.org/10.1001/jamadermatol.2020.1729>
- Grossmann, A. H., Grossmann, K. F., & Wallander, M. L. (2012). Molecular testing in malignant melanoma. *Diagn Cytopathol*, 40(6), 503-510. <https://doi.org/10.1002/dc.22810>
- Harland, M., Petljak, M., Robles-Espinoza, C. D., Ding, Z., Gruis, N. A., van Doorn, R., Pooley, K. A., Dunning, A. M., Aoude, L. G., Wadt, K. A., Gerdes, A. M., Brown, K. M., Hayward, N. K., Newton-Bishop, J. A., Adams, D. J., & Bishop, D. T. (2016). Germline TERT promoter mutations are rare in familial melanoma. *Fam Cancer*, 15(1), 139-144. <https://doi.org/10.1007/s10689-015-9841-9>
- Hawkes, J. E., Truong, A., & Meyer, L. J. (2016). Genetic predisposition to melanoma. *Semin Oncol*, 43(5), 591-597. <https://doi.org/10.1053/j.seminoncol.2016.08.003>
- Hocker, T., & Tsao, H. (2007). Ultraviolet radiation and melanoma: a systematic review and analysis of reported sequence variants. *Hum Mutat*, 28(6), 578-588. <https://doi.org/10.1002/humu.20481>
- Hodi, F. S., O'Day, S. J., McDermott, D. F., Weber, R. W., Sosman, J. A., Haanen, J. B., Gonzalez, R., Robert, C., Schadendorf, D., Hassel, J. C., Akerley, W., van den Eertwegh, A. J., Lutzky, J., Lorigan, P., Vaubel, J. M., Linette, G. P., Hogg, D., Ottensmeier, C. H., Lebbe, C., Urba, W. J. (2010). Improved survival with ipilimumab in patients with metastatic melanoma. *N Engl J Med*, 363(8), 711-723. <https://doi.org/10.1056/NEJMoa1003466>

Molecular Testing for Cutaneous Melanoma AHS-M2029

Holmes, D. (2014). The cancer that rises with the sun. *Nature*, 515(7527), S110-111. <https://doi.org/10.1038/515S110a>

Horn, S., Figl, A., Rachakonda, P. S., Fischer, C., Sucker, A., Gast, A., Kadel, S., Moll, I., Nagore, E., Hemminki, K., Schadendorf, D., & Kumar, R. (2013). TERT promoter mutations in familial and sporadic melanoma. *Science*, 339(6122), 959-961. <https://doi.org/10.1126/science.1230062>

Hornberger, J., & Siegel, D. M. (2018). Economic Analysis of a Noninvasive Molecular Pathologic Assay for Pigmented Skin Lesions. *JAMA Dermatol*, 154(9), 1025-1031. <https://doi.org/10.1001/jamadermatol.2018.1764>

Hussussian, C. J., Struewing, J. P., Goldstein, A. M., Higgins, P. A., Ally, D. S., Sheahan, M. D., Clark, W. H., Jr., Tucker, M. A., & Dracopoli, N. C. (1994). Germline p16 mutations in familial melanoma. *Nat Genet*, 8(1), 15-21. <https://doi.org/10.1038/ng0994-15>

Institute, N.C. (2023). *Surveillance, epidemiology, and end results program*. Retrieved 3/10/23 from <https://seer.cancer.gov/statfacts/html/melan.html>

Invitae. (2023). *Invitae Melanoma Panel*. <https://www.invitae.com/en/physician/tests/01561/>

Kangas-Dick, A. W., Greenbaum, A., Gall, V., Groisberg, R., Mehnert, J., Chen, C., Moore, D. F., Berger, A. C., & Koshenkov, V. (2021). Evaluation of a Gene Expression Profiling Assay in Primary Cutaneous Melanoma. *Ann Surg Oncol*. <https://doi.org/10.1245/s10434-020-09563-7>

Keller, J., Schwartz, T. L., Lizalek, J. M., Chang, E. S., Patel, A. D., Hurley, M. Y., & Hsueh, E. C. (2019). Prospective validation of the prognostic 31-gene expression profiling test in primary cutaneous melanoma. *Cancer Med*, 8(5), 2205-2212. <https://doi.org/10.1002/cam4.2128>

Ko, J. S., Matharoo-Ball, B., Billings, S. D., Thomson, B. J., Tang, J. Y., Sarin, K. Y., Cai, E., Kim, J., Rock, C., Kimbrell, H. Z., Flake, D. D., 2nd, Warf, M. B., Nelson, J., Davis, T., Miller, C., Rushton, K., Hartman, A. R., Wenstrup, R. J., & . . . Clarke, L. E. (2017). Diagnostic Distinction of Malignant Melanoma and Benign Nevi by a Gene Expression Signature and Correlation to Clinical Outcomes. *Cancer Epidemiol Biomarkers Prev*, 26(7), 1107-1113. <https://doi.org/10.1158/1055-9965.EPI-16-0958>

Koh, J., Enders, G. H., Dynlacht, B. D., & Harlow, E. (1995). Tumour-derived p16 alleles encoding proteins defective in cell-cycle inhibition. *Nature*, 375(6531), 506-510. <https://doi.org/10.1038/375506a0>

Labus, M., Ellis, R., Ewen, T., Podlipnik, S., Greenwood, A., McConnell, A., Long, A., Ness, T., Jones, C., Stefanos, N., Sloan, P., Husain, A., Barrett, P., Puig, S., McArthur, G. A., & Lovat, P. (2020). AMBLor: A prognostic and stratifying biomarker for adjuvant immunotherapy of AJCC stage II melanomas. *Journal of Clinical Oncology*, 38(15_suppl), e22094-e22094. https://doi.org/10.1200/JCO.2020.38.15_suppl.e22094

Leachman, S. A., Lucero, O. M., Sampson, J. E., Cassidy, P., Bruno, W., Queirolo, P., & Ghiorzo, P. (2017). Identification, genetic testing, and management of hereditary melanoma. *Cancer Metastasis Rev*, 36(1), 77-90. <https://doi.org/10.1007/s10555-017-9661-5>

Lee, J. J., & Lian, C. G. (2018). Molecular Testing for Cutaneous Melanoma: An Update and Review. *Arch Pathol Lab Med*. <https://doi.org/10.5858/arpa.2018-0038-RA>

Leong, S. P., Gershenwald, J. E., Soong, S. J., Schadendorf, D., Tarhini, A. A., Agarwala, S., Hauschild, A., Soon, C. W., Daud, A., & Kashani-Sabet, M. (2011). Cutaneous melanoma: a model to study cancer metastasis. *J Surg Oncol*, 103(6), 538-549. <https://doi.org/10.1002/jso.21816>

Molecular Testing for Cutaneous Melanoma AHS-M2029

- Linares, M. A., Zakaria, A., & Nizran, P. (2015). Skin Cancer. *Prim Care*, 42(4), 645-659. <https://doi.org/10.1016/j.pop.2015.07.006>
- Lokhandwala, P. M., Tseng, L. H., Rodriguez, E., Zheng, G., Pallavajjala, A., Gocke, C. D., Eshleman, J. R., & Lin, M. T. (2019). Clinical mutational profiling and categorization of BRAF mutations in melanomas using next generation sequencing. *BMC Cancer*, 19(1), 665. <https://doi.org/10.1186/s12885-019-5864-1>
- Lynch, H. T., & Krush, A. J. (1968). Heredity and malignant melanoma: implications for early cancer detection. *Can Med Assoc J*, 99(1), 17-21. <https://www.ncbi.nlm.nih.gov/pubmed/5663000>
- Marchetti, M. A., Coit, D. G., Dusza, S. W., Yu, A., McLean, L., Hu, Y., Nanda, J. K., Matsoukas, K., Mancebo, S. E., & Bartlett, E. K. (2020). Performance of Gene Expression Profile Tests for Prognosis in Patients With Localized Cutaneous Melanoma: A Systematic Review and Meta-analysis. *JAMA Dermatol*, 156(9), 953-962. <https://doi.org/10.1001/jamadermatol.2020.1731>
- Marks, E., Caruso, H., Kurley, S., Ibad, S., Plasseraud, K., Monzon, F., & Cockerell, C. (2019). Establishing an evidence-based decision point for clinical use of the 31-gene expression profile test in cutaneous melanoma. *SKIN The Journal of Cutaneous Medicine*, 3(4). <https://www.jofskin.org/index.php/skin/article/view/628>
- Marzuka-Alcala, A., Gabree, M. J., & Tsao, H. (2014). Melanoma susceptibility genes and risk assessment. *Methods Mol Biol*, 1102, 381-393. https://doi.org/10.1007/978-1-62703-727-3_20
- Menin, C., Bojnik, E., Del Bianco, P., Elefanti, L., Giansin, K., Keppel, S., Stagni, C., Mocellin, S., Vecchiato, A., & De Rossi, A. (2016). Differences in telomere length between sporadic and familial cutaneous melanoma. *Br J Dermatol*, 175(5), 937-943. <https://doi.org/10.1111/bjd.14652>
- Middlebrooks, C. D., Stacey, M. L., Li, Q., Snyder, C., Shaw, T. G., Richardson-Nelson, T., Rendell, M., Ferguson, C., Silberstein, P., Casey, M. J., Bailey-Wilson, J. E., & Lynch, H. T. (2019). Analysis of the CDKN2A Gene in FAMMM Syndrome Families Reveals Early Age of Onset for Additional Syndromic Cancers. *Cancer Res*, 79(11), 2992-3000. <https://doi.org/10.1158/0008-5472.Can-18-1580>
- Minca, E. C., Al-Rohil, R. N., Wang, M., Harms, P. W., Ko, J. S., Collie, A. M., Kovalyshyn, I., Prieto, V. G., Tetzlaff, M. T., Billings, S. D., & Andea, A. A. (2016). Comparison between melanoma gene expression score and fluorescence in situ hybridization for the classification of melanocytic lesions. *Mod Pathol*, 29(8), 832-843. <https://doi.org/10.1038/modpathol.2016.84>
- NCCN. (2023). *NCCN Clinical Practice Guidelines in Oncology; Cutaneous Melanoma*. https://www.nccn.org/professionals/physician_gls/pdf/cutaneous_melanoma.pdf
- NCI. (2023a). *Drugs Approved for Melanoma*. <https://www.cancer.gov/about-cancer/treatment/drugs/melanoma>
- NCI. (2023b). Surveillance, epidemiology, and end results program. Retrieved 10/20/2021 from <https://seer.cancer.gov/statfacts/html/melan.html>
- Nelson, A. A., & Tsao, H. (2009). Melanoma and genetics. *Clin Dermatol*, 27(1), 46-52. <https://doi.org/10.1016/j.clindermatol.2008.09.005>
- O'Brien, O., Lyons, T., Murphy, S., Feeley, L., Power, D., & Heffron, C. (2017). BRAF V600 mutation detection in melanoma: a comparison of two laboratory testing methods. *J Clin Pathol*, 70(11), 935-940. <https://doi.org/10.1136/jclinpath-2017-204367>

Molecular Testing for Cutaneous Melanoma AHS-M2029

Pasquali, E., Garcia-Borron, J. C., Fargnoli, M. C., Gandini, S., Maisonneuve, P., Bagnardi, V., Specchia, C., Liu, F., Kayser, M., Nijsten, T., Nagore, E., Kumar, R., Hansson, J., Kanetsky, P. A., Ghiorzo, P., Debniak, T., Branicki, W., Gruis, N. A., Han, J., . . . Group, M. S. S. (2015). MC1R variants increased the risk of sporadic cutaneous melanoma in darker-pigmented Caucasians: a pooled-analysis from the M-SKIP project. *Int J Cancer*, *136*(3), 618-631. <https://doi.org/10.1002/ijc.29018>

Paszowska-Szczur, K., Scott, R. J., Serrano-Fernandez, P., Mirecka, A., Gapska, P., Gorski, B., Cybulski, C., Maleszka, R., Sulikowski, M., Nagay, L., Lubinski, J., & Debniak, T. (2013). Xeroderma pigmentosum genes and melanoma risk. *Int J Cancer*, *133*(5), 1094-1100. <https://doi.org/10.1002/ijc.28123>

Pissa, M., Helkkula, T., Appelqvist, F., Silander, G., Borg, Å., Pettersson, J., Lapins, J., Nielsen, K., Höiom, V., & Helgadottir, H. (2021). CDKN2A genetic testing in melanoma-prone families in Sweden in the years 2015-2020: implications for novel national recommendations. *Acta Oncol*, *60*(7), 888-896. <https://doi.org/10.1080/0284186x.2021.1914346>

Randerson-Moor, J. A., Harland, M., Williams, S., Cuthbert-Heavens, D., Sheridan, E., Aveyard, J., Sibley, K., Whitaker, L., Knowles, M., Bishop, J. N., & Bishop, D. T. (2001). A germline deletion of p14(ARF) but not CDKN2A in a melanoma-neural system tumour syndrome family. *Hum Mol Genet*, *10*(1), 55-62. <https://www.ncbi.nlm.nih.gov/pubmed/11136714>

Ransohoff, K. J., Jaju, P. D., Tang, J. Y., Carbone, M., Leachman, S., & Sarin, K. Y. (2016). Familial skin cancer syndromes: Increased melanoma risk. *J Am Acad Dermatol*, *74*(3), 423-434; quiz 435-426. <https://doi.org/10.1016/j.jaad.2015.09.070>

Rashid, O. M., & Zager, J. S. (2015). Genetic Testing in the Multidisciplinary Management of Melanoma. *Surg Oncol Clin N Am*, *24*(4), 779-793. <https://doi.org/10.1016/j.soc.2015.06.003>

Ribas, A., & Flaherty, K. T. (2011). BRAF targeted therapy changes the treatment paradigm in melanoma. *Nat Rev Clin Oncol*, *8*(7), 426-433. <https://doi.org/10.1038/nrclinonc.2011.69>

Ribero, S., Sportoletti Baduel, E., Osella-Abate, S., Dika, E., Quaglino, P., Picciotto, F., Macripo, G., & Bataille, V. (2017). Sentinel lymph node biopsy in cutaneous melanoma. *G Ital Dermatol Venereol*, *152*(4), 355-359. <https://doi.org/10.23736/s0392-0488.16.05358-x>

Robinson, J. K., & Jansen, B. (2020). Caring for Melanoma Survivors with Self-Detected Concerning Moles During COVID-19 Restricted Physician Access: a Cohort Study. *SKIN The Journal of Cutaneous Medicine*, *4*(3), 248-251.

Rossi, M., Pellegrini, C., Cardelli, L., Ciciarelli, V., Di Nardo, L., & Fargnoli, M. C. (2019). Familial Melanoma: Diagnostic and Management Implications. *Dermatol Pract Concept*, *9*(1), 10-16. <https://doi.org/10.5826/dpc.0901a03>

Seth, R., Messersmith, H., Kaur, V., Kirkwood, J. M., Kudchadkar, R., McQuade, J. L., Provenzano, A., Swami, U., Weber, J., Alluri, K. C., Agarwala, S., Ascierto, P. A., Atkins, M. B., Davis, N., Ernstoff, M. S., Faries, M. B., Gold, J. S., Guild, S., Gyorki, D. E., Funchain, P. (2020). Systemic Therapy for Melanoma: ASCO Guideline. *Journal of Clinical Oncology*, *0*(0), JCO.20.00198. <https://doi.org/10.1200/jco.20.00198>

Simone, Valiante, M., & Silipo, V. (2017). Familial melanoma and multiple primary melanoma. *G Ital Dermatol Venereol*, *152*(3), 262-265. <https://doi.org/10.23736/s0392-0488.17.05554-7>

Skelsey, M. K., Brouha, B., Rock, J., Howell, M. D., Jansen, B., Clarke, L. E., & Peck, G. (2021). Non-Invasive Detection of Genomic Atypia Increases Real-World NPV and PPV of the Melanoma Diagnostic Pathway and Reduces Biopsy Burden. *SKIN*, *5*(5), 512-523.

Molecular Testing for Cutaneous Melanoma AHS-M2029

Skudalski, L., Waldman, R., Kerr, P. E., & Grant-Kels, J. M. (2022). Melanoma: How and when to consider clinical diagnostic technologies. *J Am Acad Dermatol*, 86(3), 503-512. <https://doi.org/10.1016/j.jaad.2021.06.901>

SkylineDx. (2020). Melanoma. <https://www.skylinedx.com/what/our-pipeline>

Soura, E., Eliades, P. J., Shannon, K., Stratigos, A. J., & Tsao, H. (2016a). Hereditary melanoma: Update on syndromes and management: Emerging melanoma cancer complexes and genetic counseling. *J Am Acad Dermatol*, 74(3), 411-420; quiz 421-412. <https://doi.org/10.1016/j.jaad.2015.08.037>

Soura, E., Eliades, P. J., Shannon, K., Stratigos, A. J., & Tsao, H. (2016b). Hereditary melanoma: Update on syndromes and management: Genetics of familial atypical multiple mole melanoma syndrome. *J Am Acad Dermatol*, 74(3), 395-407; quiz 408-310. <https://doi.org/10.1016/j.jaad.2015.08.038>

Stolarova, L., Jelinkova, S., Storchova, R., Machackova, E., Zemankova, P., Vocka, M., Kodet, O., Kral, J., Cerna, M., Volkova, Z., Janatova, M., Soukupova, J., Stranecky, V., Dunder, P., Foretova, L., Macurek, L., Kleiblova, P., & Kleibl, Z. (2020). Identification of Germline Mutations in Melanoma Patients with Early Onset, Double Primary Tumors, or Family Cancer History by NGS Analysis of 217 Genes. *Biomedicines*, 8(10). <https://doi.org/10.3390/biomedicines8100404>

Swetter, S. M. (2021, 2/22/2021). Melanoma: Clinical features and diagnosis. Retrieved 10/19/21 from <https://www.uptodate.com/contents/melanoma-clinical-features-and-diagnosis>

Swetter, S. M., Tsao, H., Bichakjian, C. K., Curiel-Lewandrowski, C., Elder, D. E., Gershenwald, J. E., Guild, V., Grant-Kels, J. M., Halpern, A. C., Johnson, T. M., Sober, A. J., Thompson, J. A., Wisco, O. J., Wyatt, S., Hu, S., & Lamina, T. (2019). Guidelines of care for the management of primary cutaneous melanoma. *J Am Acad Dermatol*, 80(1), 208-250. <https://doi.org/10.1016/j.jaad.2018.08.055>

Torres, R., Lang, U. E., Hejna, M., Shelton, S. J., Joseph, N. M., Shain, A. H., Yeh, I., Wei, M. L., Oldham, M. C., Bastian, B. C., & Judson-Torres, R. L. (2019). MicroRNA ratios distinguish melanomas from nevi. *J Invest Dermatol*. <https://doi.org/10.1016/j.jid.2019.06.126>

Tsao, H., Atkins, M. B., & Sober, A. J. (2004). Management of cutaneous melanoma. *N Engl J Med*, 351(10), 998-1012. <https://doi.org/10.1056/NEJMra041245>

Tsao, H., & McCormick, S. R. (2022, May 22). *Inherited susceptibility to melanoma*. UpToDate. Retrieved October 30 from <https://www.uptodate.com/contents/inherited-susceptibility-to-melanoma>

Varedi, A., Gardner, L. J., Kim, C. C., Chu, E. Y., Ming, M. E., Leachman, S. A., Curiel-Lewandrowski, C., Swetter, S. M., & Grossman, D. (2019). Use of new molecular tests for melanoma by pigmented-lesion experts. *J Am Acad Dermatol*. <https://doi.org/10.1016/j.jaad.2019.08.022>

Volmar, K. E., Idowu, M. O., Souers, R. J., & Nakhleh, R. E. (2015). Molecular Testing in Anatomic Pathology and Adherence to Guidelines: A College of American Pathologists Q-Probes Study of 2230 Testing Events Reported by 26 Institutions. *Arch Pathol Lab Med*, 139(9), 1115-1124. <https://doi.org/10.5858/arpa.2014-0513-CP>

Weiss, S. A., Hanniford, D., Hernando, E., & Osman, I. (2015). Revisiting determinants of prognosis in cutaneous melanoma. *Cancer*, 121(23), 4108-4123. <https://doi.org/10.1002/cncr.29634>

Wendt, J., Mueller, C., Rauscher, S., Fae, I., Fischer, G., & Okamoto, I. (2018). Contributions by MC1R Variants to Melanoma Risk in Males and Females. *JAMA Dermatol*, 154(7), 789-795. <https://doi.org/10.1001/jamadermatol.2018.1252>

Molecular Testing for Cutaneous Melanoma AHS-M2029

Wernli, K. J., Henrikson, N. B., Morrison, C. C., Nguyen, M., Pocobelli, G., & Blasi, P. R. (2016). Screening for Skin Cancer in Adults: Updated Evidence Report and Systematic Review for the US Preventive Services Task Force. *JAMA*, *316*(4), 436-447. <https://doi.org/10.1001/jama.2016.5415>

Wiesner, T., Obenauf, A. C., Murali, R., Fried, I., Griewank, K. G., Ulz, P., Windpassinger, C., Wackernagel, W., Loy, S., Wolf, I., Viale, A., Lash, A. E., Pirun, M., Succi, N. D., Rutten, A., Palmedo, G., Abramson, D., Offit, K., Ott, A., . . . Speicher, M. R. (2011). Germline mutations in BAP1 predispose to melanocytic tumors. *Nat Genet*, *43*(10), 1018-1021. <https://doi.org/10.1038/ng.910>

Woodman, S. E., & Davies, M. A. (2010). Targeting KIT in melanoma: a paradigm of molecular medicine and targeted therapeutics. *Biochem Pharmacol*, *80*(5), 568-574. <https://doi.org/10.1016/j.bcp.2010.04.032>

Yao, Z., Moy, R., Allen, T., & Jansen, B. (2017). An Adhesive Patch-Based Skin Biopsy Device for Molecular Diagnostics and Skin Microbiome Studies. *J Drugs Dermatol*, *16*(10), 979-986. <https://www.ncbi.nlm.nih.gov/pubmed/29036251>

Yee, V. S., Thompson, J. F., McKinnon, J. G., Scolyer, R. A., Li, L. X., McCarthy, W. H., O'Brien, C. J., Quinn, M. J., Saw, R. P., Shannon, K. F., Stretch, J. R., & Uren, R. F. (2005). Outcome in 846 cutaneous melanoma patients from a single center after a negative sentinel node biopsy. *Ann Surg Oncol*, *12*(6), 429-439. <https://doi.org/10.1245/aso.2005.03.074>

Yokoyama, S., Woods, S. L., Boyle, G. M., Aoude, L. G., MacGregor, S., Zismann, V., Gartside, M., Cust, A. E., Haq, R., Harland, M., Taylor, J. C., Duffy, D. L., Holohan, K., Dutton-Regester, K., Palmer, J. M., Bonazzi, V., Stark, M. S., Symmons, J., Law, M. H., . . . Brown, K. M. (2011). A novel recurrent mutation in MITF predisposes to familial and sporadic melanoma. *Nature*, *480*(7375), 99-103. <https://doi.org/10.1038/nature10630>

Yousaf, A., Tjien-Fooh, F. J., Rentroia-Pacheco, B., Quattrocchi, E., Kobic, A., Tempel, D., Kolodney, M., & Meves, A. (2021). Validation of CP-GEP (Merlin Assay) for predicting sentinel lymph node metastasis in primary cutaneous melanoma patients: A U.S. cohort study. *Int J Dermatol*, *60*(7), 851-856. <https://doi.org/10.1111/ijd.15594>

Zhang, Y., Xiong, Y., & Yarbrough, W. G. (1998). ARF promotes MDM2 degradation and stabilizes p53: ARF-INK4a locus deletion impairs both the Rb and p53 tumor suppression pathways. *Cell*, *92*(6), 725-734. <https://www.ncbi.nlm.nih.gov/pubmed/9529249>

Zippel, D., Markel, G., Shapira-Frommer, R., Ben-Betzalel, G., Goitein, D., Ben-Ami, E., Nissan, A., Schachter, J., & Schneebaum, S. (2017). Perioperative BRAF inhibitors in locally advanced stage III melanoma. *J Surg Oncol*, *116*(7), 856-861. <https://doi.org/10.1002/jso.24744>

For Policy Titled: Genetic Testing and Genetic Expression Profiling in Patients with Cutaneous Melanoma

Specialty Matched Consultant Advisory Panel review 3/2020

Medical Director review 3/2020

Medical Director review 1/2021

Specialty Matched Consultant Advisory Panel review 3/2021

Medical Director review 3/2021

Medical Director review 1/2022

Molecular Testing for Cutaneous Melanoma AHS-M2029

Medical Director review 11/2022

For Policy Re-Titled: Molecular Testing for Cutaneous Melanoma

Medical Director review 7/2023

Policy Implementation/Update Information

For Policy Titled: BRAF Genetic Testing in Patients with Melanoma

- 1/1/2019 New policy developed. BCBSNC will provide coverage for BRAF genetic testing in patients with melanoma when it is determined to be medically necessary and criteria are met. Medical Director review 1/1/2019. Policy noticed 1/1/2019 for effective date 4/1/2019. (lpr)
- 11/12/19 Wording in the Policy, When Covered, and/or Not Covered section(s) changed from Medical Necessity to Reimbursement language, where needed. (hb)

For Policy Titled: Genetic Testing and Genetic Expression Profiling in Patients with Cutaneous Melanoma

- 2/11/20 Reviewed by Avalon Q4 2019 CAB. Under “When Not Covered” section added statement: Genetic expression profiling testing for cutaneous melanoma is considered investigational. Added Note: For testing of 5 or more genes for an affected individual with cutaneous melanoma, please refer to AHS-M2109 Molecular Panel Testing of Cancers for Diagnosis, Prognosis, and Identification of Targeted Therapy. Under Billing/Coding section: Deleted CPT codes 81445, 81450, 81455 and added PLA codes: 0089U and 0090U. **Policy Title changed from: BRAF Genetic Testing in Patients with Melanoma to: Genetic Testing and Genetic Expression Profiling in Patients with Cutaneous Melanoma.** Medical Director review 1/2020. (lpr)
- 3/31/20 Specialty Matched Consultant Advisory Panel review 3/18/2020. No change to policy statement. (lpr)
- 2/9/21 Reviewed by Avalon Q4 2020 CAB. Under “When Covered” section: added NRAS and consolidated statements 1. and 2. into one statement addressing both stage III and IV melanoma. References and Description section updated. Extensive revisions/updates to Policy Guidelines section. Added related policies section. Added CPT codes 81311, 81529 to Billing/Coding section. Medical Director review 1/2021. (lpr)
- 4/6/21 Specialty Matched Consultant Advisory Panel review 3/17/2021. No change to policy statement. (lpr)
- 2/8/22 Reviewed by Avalon Q4 2021 CAB. Policy Guidelines and references updated. Medical Director review 1/2022. No change to policy statement. (lpr)
- 5/17/22 Reviewed by Avalon Q1 2022 CAB. Medical Director review 4/2022. Removed reference to AHS-M2109. Edited Note to read “For 5 or more gene tests being run on the same platform, such as multi-gene panel next generation sequencing, please refer to AHS-R2162 Reimbursement Policy.” Updated policy guidelines and references.(lpr)
- 12/13/22 Reviewed by Avalon Q3 2022 CAB. Medical Director review 11/2022. Updated the following sections: description, policy guidelines, guidelines/recommendations, and references. When covered section updated for clarity. No change to policy statement. (lpr)

Molecular Testing for Cutaneous Melanoma AHS-M2029

For Policy Re-Titled: Molecular Testing for Cutaneous Melanoma

8/15/23 Reviewed by Avalon Q2 2023 CAB. Medical Director review 7/2023. Extensive revisions with information and coverage from M2037 Genetic Testing for Familial Cutaneous Malignant Melanoma transferred into this policy. M2029 and M2037 combined into one single policy. Updated description, coverage criteria, policy guidelines, and references. Added CPT codes: 81167, 81216, 81217, 81345, 81404, 81479, 0387U, 0314U to Billing/Coding section. **Policy title changed from: Genetic Testing and Genetic Expression Profiling in Patients with Cutaneous Melanoma to Molecular Testing for Cutaneous Melanoma.** (lpr)

Medical policy is not an authorization, certification, explanation of benefits or a contract. Benefits and eligibility are determined before medical guidelines and payment guidelines are applied. Benefits are determined by the group contract and subscriber certificate that is in effect at the time services are rendered. This document is solely provided for informational purposes only and is based on research of current medical literature and review of common medical practices in the treatment and diagnosis of disease. Medical practices and knowledge are constantly changing and BCBSNC reserves the right to review and revise its medical policies periodically.