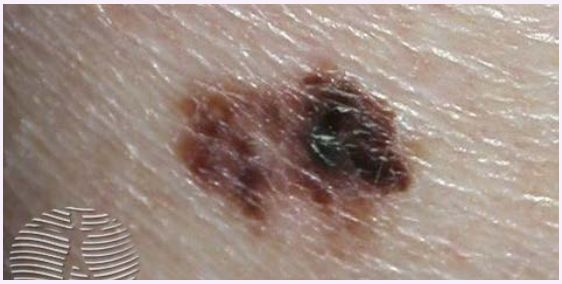
AUSTRALIAN Clinicallabs

MELASEQ™
melanoma genomic score

Melaseq™ Solid Tissue
microRNA Biomarker Profile of Cutaneous Melanoma for Enhanced Diagnostic Clarity

Australia leads the world in melanoma incidence with approximately 17,000 new cases of melanoma diagnosed each year, and approximately 1,400 deaths annually.[1]



Successful treatment of cutaneous Melanoma depends on the early and accurate diagnosis of clinically suspicious melanocytic skin lesions. Advances in screening and diagnosis have increased the incidence of early-stage melanoma diagnoses worldwide, but have not reduced overall mortality rates.[2] New treatments, such as targeted therapies and immunotherapy, have shown promise in reducing mortality. However, these are only available for late-stage melanoma patients, are costly and their benefit is still being evaluated.[3,4]

Currently, melanocytic lesions suspected of being malignant are diagnosed using visual methods, including dermoscopy, biopsy and histopathologic examination. Studies show however, that major diagnostic discordances in melanoma histopathology may occur in over 25% of cases, resulting in a change in patient management in 18% of cases.[5]
There is consensus among experts that the introduction of standardised pathology terminology and new molecular tests are needed to improve diagnostic accuracy, patient outcomes and resource use efficiency.[6-11]

Why Choose Melaseq?

The Melaseq test uses genomic profiling of microRNAs in the biopsy tissue to improve the diagnostic clarity for complex melanocytic lesions. This can be used to make informed decisions for further clinical management.

Clinical adaptation of the Melaseq test has the potential to add novel, robust and personalised genomic information to the diagnostic picture of patients with clinically suspicious melanocytic lesions.

Combining diagnostic modalities is becoming the standard of care for many cancer types, including breast and prostate cancer. These cancers have seen promising reductions in mortality over recent years, in part due to increased diagnostic precision and molecular disease subtyping.
The Melaseq score is intended as a complement to conventional histopathology, not a replacement for current diagnostic practices. Genomic testing, microscopic and immunohistochemical assessments all add unique and sometimes overlapping data points that contribute to a complete diagnostic picture. Incorporating genomic profiling into diagnostic workups may be useful for understanding future clinical events, including treatment responses that are inconsistent with the original diagnosis and stage of disease.

Early Diagnosis

Melanocytic lesions are not always overtly malignant on histopathology, particularly when small or undifferentiated in appearance. Some lesions may have 'atypical' features which defy definitive diagnosis, which lead to patient anxiety and unclear guidelines for follow up. Use of the Melaseq Genomic Test in such lesions may clarify appropriate use of clinical management guidelines, potentially preventing disease progression from underdiagnosis or lack of a definitive diagnosis.

More Accurate

The Melaseq Genomic Test has the potential to improve outcomes by enhancing diagnostic accuracy and precision, thereby reducing patient anxiety and medicolegal risks involved in melanoma diagnosis.

"This is the first known study to demonstrate that a circulating series of microRNAs also exhibit disease specific levels of expression in solid tissue"

A Pathway to Precision

MicroRNAs are post-transcriptional regulators of gene expression with tissue and disease specific patterns of expression.[12] Their role in melanoma oncogenesis and progression is well documented.

A signature of 38 circulating microRNAs (known as MEL38) was identified by genomic profiling of individuals with Stage 1 to 4 melanoma. Biological functions include regulators of angiogenesis and inflammation [n=2], invasion and metastasis [n=14], immune response and treatment resistance [n=11], and tumour suppression and oncogenic activation [n=8].

The protein coding mRNAs identified to be regulated by MEL38 microRNAs, overlap significantly with the MAPK signalling pathway.[13] Modulation of this pathway is responsible for BRAF and NRAS changes found in melanoma cells.[13,14]

The MEL38 genomic signature was originally discovered in patient plasma, and is now additionally validated as a robust biomarker of disease status in solid tissue. This is the first known study to demonstrate that a circulating series of microRNAs also exhibit disease-specific levels of expression in solid tissue, and allows the test to be performed on the excisional biopsy material submitted for histopathological diagnosis.

FIG 1: CLINICAL CONSIDERATIONS: Reducing diagnostic ambiguity
This clinical diagnosis pathway for excised lesions ensures a comprehensive diagnosis, improving ambiguity for appropriate treatment.

The Melaseq Score

A gene weighting algorithm is applied to the quantified microRNA counts to generate a classification score MEL38. The scores range from 0 to 10 and are positively associated with increasing levels of melanoma malignancy, from benign naevi to malignant melanoma with metastatic capability. In multiple validation cohorts, with various specimen types, the score has shown robust and statistically significant associations with melanoma status, treatment response and prognosis.[13,15,16] When analysed in multivariate models, including age, gender, histological subtype, Breslow depth, tumour cell percentage, clinical stage of MPATH-Dx (v1.0) class (see Table 1), the Melaseq score remains statistically significant.[5]

How is the Melaseq Score Used?

The Melaseq Score can be evaluated as a continuous variable, or binary decision point for clinically higher versus lower risk melanocytic lesions. Scores of 2.9 or greater indicate a high likelihood of malignancy and more intensive suggested clinical management actions. A Melaseq Score of ≥2.9 is able to identify higher risk melanomas with high sensitivity (89%) and specificity (94%). As a continuous variable, the Melaseq score exhibits a strong positive correlation with the progressive stages of melanoma.[5]

The largest difference in Melaseq Score occurs between MPATH-Dx (v1.0) class IV (AJCC stage 1A) and V (stage 1B) disease. Notable differences in suggested clinical actions and patient outcome, particularly survival rates, are seen between these two classes. The clarity of diagnosis is markedly improved with the Melaseq test. Using the Melaseq score to identify MPATH-Dx (v1.0) Class V/higher risk lesions would result in an underdiagnosis rate of 8.4% compared with the reported rate of 27% for conventional pathology alone.[17] Technical reproducibility studies show stability of the score on repeated analysis.[5]

Table 1: Summary of MPATH -Dx reporting schema for classification of melanocytic skin lesions into 5 diagnostic categories			
MPATH-Dx class	**Perceived risk for progression**	**Suggested intervention***	**Examples**
0	Incomplete study due to sampling or technical limitations	Repeat biopsy or short term follow-up	NA
I	Very low risk	No further treatment	Common melanocytic nevus; blue nevus; mildly dysplastic naevus
II	Low risk	Narrow but complete excision (< 5mm)	Moderately dysplastic nevus; -Spitz nevus
III	Higher risk. Greater need for intervention	Complete excision with at least 5mm but < 1cm margins	Severely dysplastic nevus; melanoma in situ; atypical Spitz tumor
IV	Substantial risk for local or regional progression	Wide local excision with ≥ 1cm margins	Thin, invasive melanomas (eg, pT1at)
V	Greatest risk for regional and/or distant metastases	Wide local excision with ≥ 1cm margins. Consideration of staging sentinel lymph node biopsy, adjuvant therapy	Thicker, invasive melanomas (eg, pT1b, stage 2 or greater**)
*Assuming representative sampling of lesion. **According to American Joint Committee on Cancer seventh edition cancer staging manual			

Interpreting the Melaseq Report

The Melaseq Report generates a ***Melanoma Genomic Score Test Result*** as a numeric value, and an ***Interpretation*** which classifies the lesion into Class A - low probability of invasive cutaneous melanoma, and Class B - high probability of invasive cutaneous melanoma. A table integrating MPATH-Dx (v1.0) diagnostic classes, AJCC T stage and Melaseq score is provided to give guidance for perceived risk for progression, 5yr and 10yr melanoma specific survival and suggested clinical action (see Table 2 below).

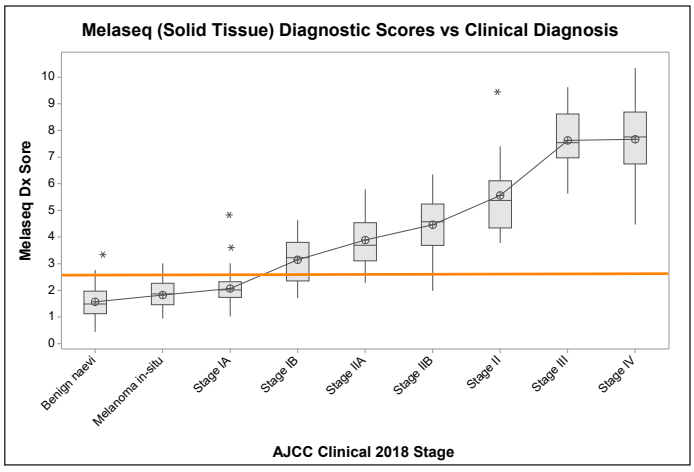


Table 2: MPATH-Dx classes, mean MEL38 scores and associated clinicopathological variables, including suggested clinical actions.					
MPATH-Dx class	**I**	**II**	**III**	**IV**	**V**
Average MEL38 score	1.5	1.7	1.7	2.1	5.2
Typical Melaseq probability category	Low Probability (≤2.4)				High Probability (≥2.9)
T stage	N/A	N/A	0	T1a	≥T1b
Perceived risk for progression	Very-low risk	Low risk	Higher risk	Substantial risk for local/regional progression	Greatest risk for regional and/or distant metastases
5/10-year melanoma-specific survival*	100% / 100%	100% / 100%	100% / 100%	98% / 96%	≤93% / ≤89%
Suggested clinical action**:	Follow-up as necessary	Narrow but complete excision (<5mm)	Complete excision with at least 5mm but <10mm margins.	Wide local excision with ≥10mm margins	Wide local excision with ≥10mm margins. Consideration of sentinel lymph node biopsy, adjuvant therapy.
*Melanoma specific survival figures based on AJCC melanoma staging and outcome data corresponding to the T-stage associated with each MPath-Dx class. ** Suggested clinical actions assuming representative sampling of lesion.					

The impact of misdiagnosis can be significant. Under-treatment can result in tumor progression and increase melanoma mortality.

Uncertainty complicates patient management decisions. Combining both the histopathology report and Melaseq Sold Tissue result improves diagnostic clarity for complex melanocytic lesions and enables, confidence in the right treatment plan.

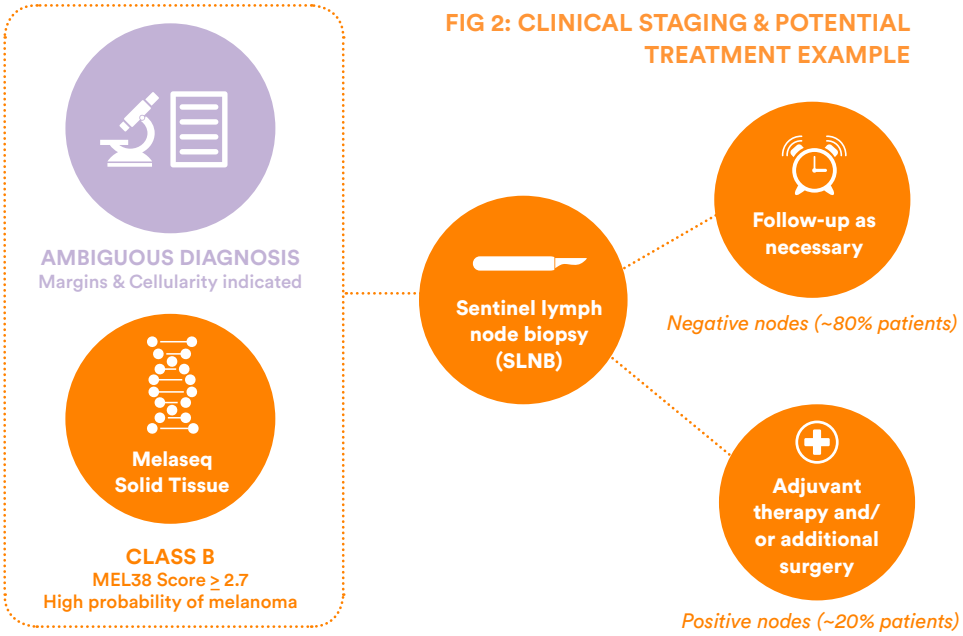
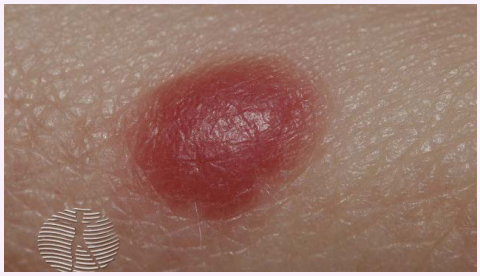


FIG 2: CLINICAL STAGING & POTENTIAL TREATMENT EXAMPLE

Spitz Naevi - A Specific Subtype



Misdiagnosis of Spitz naevus as melanoma remains an ongoing challenge. Expression profile of this subtype has similarities to both benign and malignant disease, which is also reflected by their Melaseq scores. Very few genomic studies of Spitz naevi have been performed.[18] Melaseq analysis of Spitz naevi reveals an intra-subtype profile which differs from traditional melanocytic lesions and there may be subsets of Spitz naevi, with corresponding higher Melaseq scores, that benefit from additional treatment and monitoring.[5]

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Dr Peter Kaub is a Consultant Genetic Pathologist (FRCPA), with extensive experience in the introduction, regulatory approval and clinical service provision of new genetic-based tests. After many years experience in basic research as a Molecular Biologist, he commenced medical training at the University of Sydney in 2004. A deep knowledge of the technologies that underpin genetic testing informs his approach to how they are best applied to real world clinical use.

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The Melaseq Solid Tissue Test is currently undergoing a pilot launch in Australia. If you are a doctor and interested in taking part, please email us your contact details and expression of interest at : melaseq@clinicallabs.com.au

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