A comprehensive diagnostic offering workflow increases the rate of actionable results of the 23- and 35-gene expression profile tests for use as ancillary diagnostic tools for difficult-to-diagnose melanocytic lesions

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Background

 › Diagnostic discrepancy in suspected cutaneous melanocytic lesions is well documented and particularly prevalent among difficult-to-diagnose cases, for which histopathology may be insufficient for a definitive diagnosis.1

 › The 23-gene expression profile (GEP, myPath Melanoma) and 35-GEP (DiffInk-Melanoma) tests are clinically available objective ancillary tools that facilitate diagnosis of melanocytic lesions with ambiguous histopathology. The tests use proprietary algorithms to produce results of likely benign, intermediate, or malignant.2,3

 › The 23-GEP has shown accuracy metrics of over 90% sensitivity in multiple clinical studies that included patient outcomes,4,5 However, the 23-GEP historically has resulted in ~23% of cases receiving either a technical failure or an intermediate result, which can be perceived as nonactionable.6,7,8,9 The 35-GEP test can address this shortcoming and showed both an increased sensitivity in the first validation cohort and a decreased nonactionable rate of 8.5%.6,9

 › Clinical utility has been demonstrated with benign and malignant GEP test results2,10,11 therefore, those test results are defined as actionable.

Objective

 › Today, both the 23- and 35-GEP are offered from a single laboratory as part of a comprehensive diagnostic offering (CDO) workflow. Unless preferred otherwise by the ordering clinician, clinical samples are processed first through the 23-GEP test, and if a technical failure or intermediate result is received, processed to the 35-GEP (Figure 1).

 › Here, we report test result metrics from archival research cases and from this clinical workflow.

Figure 1. Clinical workflow of the comprehensive diagnostic offering

Results

 › The Research Cohort was comprised of 738 FFPE archival biopsy samples from adults ≥18 years of age with cutaneous melanocytic lesions with a consensus diagnosis reviewed by at least three independent dermatopathologists who were blinded to the original diagnosis. All samples were run on the 23-GEP, and any intermediate or technical fail samples were subsequently run on the 35-GEP per the current clinical CDO workflow (Figure 1).

 › Accuracy metrics demonstrate high performance of the CDO workflow (Table 1).

Table 1. Accuracy metrics in research cases from the comprehensive diagnostic offering

<table>
<thead>
<tr>
<th>Research Cohort, n=738</th>
<th>CDO</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sensitivity</td>
<td>94.7%</td>
<td>92.4-96.8</td>
</tr>
<tr>
<td>Specificity</td>
<td>89.5%</td>
<td>86.3-92.7</td>
</tr>
<tr>
<td>PPV</td>
<td>90.1%</td>
<td>87.7-93.4</td>
</tr>
<tr>
<td>NPV</td>
<td>94.1%</td>
<td>91.4-96.4</td>
</tr>
<tr>
<td>Intermediate</td>
<td>0.8% n=6</td>
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</tbody>
</table>

NPV, negative predictive value; PPV, positive predictive value; CI, confidence interval.

 › Clinical test results were analyzed over a 3-month period.

 › The 23-GEP test gave an actionable result of benign or malignant in 77.8% of cases, which is comparable to past reporting in ambiguous cases for this test6,12 (Table 2).

 › Nonactionable classifications of the 23-GEP test were 22.2% (12.9% intermediate and 9.4% technical failure). These cases then underwent testing with the 35-GEP test, and an additional 20.9% of originally submitted cases received an actionable result. Only 1.1% of cases received a final intermediate test result (i.e., from both tests); the technical failure rate for the CDO was 0.2% (Table 2).

 › This clinical workflow increased the rate of an actionable result from 77.8% to 98.7% when compared with 23-GEP testing alone (Table 2).

 › The clinical workflow results were 59.5% benign, 39.1% malignant, 1.1% intermediate, and 0.2% technical failure.

Methods

 › Melanocytic lesions and associated de-identified clinical data from patients ≥18 years of age were included in this study. Samples were acquired under an IRB-approved protocol or were previously submitted for clinical testing for the 31-GEP Research samples were independently reviewed by at least 2 dermatopathologists for diagnostic adjudication, were blinded to the original diagnosis, and included in the study if they received at least 2 of 3 diagnostic concordance (Table 1). The study also included clinical cases submitted to Castle Biosciences for CDO testing with results reported since implementation of the CDO workflow between June 3 and August 31, 2021 (Table 2).

 › All cases not receiving a benign or malignant result from the 23-GEP were run on the 35-GEP, except for pediatric cases (<18 years), which were only run on the 23-GEP and excluded from this analysis. Technical fail included samples with insufficient quantity of RNA and/or control or discriminant gene amplification failure based on the requirements for each test.

Table 2. Clinical test results of the comprehensive diagnostic offering

<table>
<thead>
<tr>
<th>Clinical CDO Testing</th>
<th>Actionable (%)</th>
<th>Nonactionable (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>23-GEP alone</td>
<td>77.8%</td>
<td>22.2%</td>
</tr>
<tr>
<td>Subsequent 35-GEP</td>
<td>20.8%</td>
<td>1.3%</td>
</tr>
<tr>
<td>Overall</td>
<td>98.7%</td>
<td>1.3%</td>
</tr>
</tbody>
</table>

Cases with Intermediate or Technical Fail results from the 23-GEP undergo testing with the 35-GEP, gene expression profile.

References


Conclusions

 › Combining the 23-GEP and 35-GEP tests into one workflow leverages the strengths of both assays.

 › The CDO workflow demonstrated a high rate of accuracy in research cases, with 94.7% sensitivity and 89.5% specificity.

 › The CDO workflow for ambiguous melanocytic lesions has substantially improved reporting of clinically actionable results from a historic rate of ~77% for the 23-GEP alone to over 98%.

 › Eligible cases with a malignant result from the CDO can also be subsequently run on the 31-GEP prognostic test (Decision Dx-Melanoma), without requiring extra tissue, to predict the likelihood of recurrence and sentinel lymph node biopsy positivity.

Acknowledgments & Disclosures

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 › All authors are employees and shareholders of Castle Biosciences, Inc.

 › Notes