GENE EXPRESSION TESTS

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Table of Contents

<table>
<thead>
<tr>
<th>BENEFIT CONSIDERATIONS</th>
<th>1</th>
</tr>
</thead>
<tbody>
<tr>
<td>COVERAGE RATIONALE</td>
<td>2</td>
</tr>
<tr>
<td>APPLICABLE CODES</td>
<td>2</td>
</tr>
<tr>
<td>DESCRIPTION OF SERVICES</td>
<td>2</td>
</tr>
<tr>
<td>CLINICAL EVIDENCE</td>
<td>4</td>
</tr>
<tr>
<td>U.S. FOOD AND DRUG ADMINISTRATION</td>
<td>13</td>
</tr>
<tr>
<td>CENTERS FOR MEDICARE AND MEDICAID SERVICES (CMS)</td>
<td>13</td>
</tr>
<tr>
<td>REFERENCES</td>
<td>14</td>
</tr>
<tr>
<td>POLICY HISTORY/REVISION INFORMATION</td>
<td>19</td>
</tr>
</tbody>
</table>

Related Policies:
- Cardiovascular Disease Risk Tests
- Chemosensitivity and Chemoresistance Assays in Cancer

INSTRUCTIONS FOR USE

This Medical Policy provides assistance in interpreting UnitedHealthcare benefit plans. When deciding coverage, the enrollee specific document must be referenced. The terms of an enrollee’s document (e.g., Certificate of Coverage (COC) or Summary Plan Description (SPD) and Medicaid State Contracts) may differ greatly from the standard benefit plans upon which this Medical Policy is based. In the event of a conflict, the enrollee’s specific benefit document supersedes this Medical Policy. All reviewers must first identify enrollee eligibility, any federal or state regulatory requirements and the enrollee specific plan benefit coverage prior to use of this Medical Policy. Other Policies and Coverage Determination Guidelines may apply. UnitedHealthcare reserves the right, in its sole discretion, to modify its Policies and Guidelines as necessary. This Medical Policy is provided for informational purposes. It does not constitute medical advice.

UnitedHealthcare may also use tools developed by third parties, such as the MCG™ Care Guidelines, to assist us in administering health benefits. The MCG™ Care Guidelines are intended to be used in connection with the independent professional medical judgment of a qualified health care provider and do not constitute the practice of medicine or medical advice.

BENEFIT CONSIDERATIONS

Essential Health Benefits for Individual and Small Group:
For plan years beginning on or after January 1, 2014, the Affordable Care Act of 2010 (ACA) requires fully insured non-grandfathered individual and small group plans (inside and outside of Exchanges) to provide coverage for ten categories of Essential Health Benefits (“EHBs”). Large group plans (both self-funded and fully insured), and small group ASO plans, are not subject to the requirement to offer coverage for EHBs. However, if such plans choose to provide coverage for benefits which are deemed EHBs (such as maternity benefits), the ACA requires all dollar limits on those benefits to be removed on all Grandfathered and Non-Grandfathered plans. The determination of which benefits constitute EHBs is made on a state by state basis. As such, when using this guideline, it is important to refer to the enrollee’s specific plan document to determine benefit coverage.
COVERAGE RATIONALE

Oncology
Multi-panel gene expression tests (e.g., Afirma®) are proven and medically necessary for assessing thyroid nodules that are not clearly benign or malignant based on fine-needle aspiration biopsy results alone.

Gene expression tests are unproven and not medically necessary for the following:
- Predicting the likelihood of colon cancer recurrence (e.g., Oncotype DX® Colon Cancer Assay)
- Guiding therapy in patients with myeloma (e.g., MyPRS™)
- Identifying tissue of origin in difficult to diagnose cancers (e.g., ResponseDX Tissue of Origin or CancerTYPE ID®)
- Predicting metastatic risk of uveal melanoma (e.g., DecisionDx-UM)

There is insufficient evidence in the clinical literature demonstrating that these tests have a role in clinical decision-making or have a beneficial effect on health outcomes. Further studies are needed to determine the analytic validity, clinical validity and/or clinical utility of these tests.

Non-Oncology
Gene expression tests are unproven and not medically necessary for the following:
- Assessing cardiovascular risk (e.g., Corus® CAD)

There is insufficient evidence in the clinical literature demonstrating that this test has a role in clinical decision-making or has a beneficial effect on health outcomes. Further studies are needed to determine the analytic validity, clinical validity and clinical utility of this test.

APPLICABLE CODES

The Current Procedural Terminology (CPT®) codes and Healthcare Common Procedure Coding System (HCPCS) codes listed in this policy are for reference purposes only. Listing of a service code in this policy does not imply that the service described by this code is a covered or non-covered health service. Coverage is determined by the enrollee specific benefit document and applicable laws that may require coverage for a specific service. The inclusion of a code does not imply any right to reimbursement or guarantee claims payment. Other policies and coverage determination guidelines may apply. This list of codes may not be all inclusive.

<table>
<thead>
<tr>
<th>CPT® Code</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>81504</td>
<td>Oncology (tissue of origin), microarray gene expression profiling of &gt; 2000 genes, utilizing formalin-fixed paraffin-embedded tissue, algorithm reported as tissue similarity scores</td>
</tr>
<tr>
<td>84999</td>
<td>Unlisted chemistry procedure</td>
</tr>
<tr>
<td>88299</td>
<td>Unlisted cytogenetic study</td>
</tr>
</tbody>
</table>

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DESCRIPTION OF SERVICES

Gene expression is the process by which the coded information of a gene is translated into the structures present and operating in the cell (either proteins or ribonucleic acids (RNA)). Gene expression profiling (GEP) studies the patterns of many genes in a tissue sample at the same time to assess which ones are turned on (producing RNA and proteins) or off (not producing RNA or proteins). By simultaneously measuring the levels of RNA of thousands of genes, GEP creates a snapshot of the rate at which those genes are expressed in a tissue sample.

Gene expression tests are not the same as genetic tests. Genetic tests measure an individual DNA signature to identify genetic changes or mutations, which stay constant throughout an
individual’s lifetime. Genetic tests can help estimate an individual’s risk of developing disease in the future. In contrast, gene expression tests measure the activity of RNA in a given tissue or bodily fluid at a given point in time to provide information about an individual’s current disease state or the likelihood of future disease. RNA levels are dynamic and change as a result of disease processes or environmental signals. Because gene expression changes under pathological conditions, dynamic changes in these processes can be studied over time. Certain patterns of gene activity may be used to diagnose a disease or to predict how an individual responds to treatment (Arnett et al., 2007; CardioDX website; Centers for Disease Control and Prevention; National Cancer Institute; National Human Genome Research Institute).

The Centers for Disease Control and Prevention (CDC) created the ACCE model process for evaluating genetic or genomic-based tests. The 4 main components of the ACCE process include analytic validity, clinical validity, clinical utility and ELSI. Analytic validity refers to how accurately and reliably the test measures the genotype of interest. Clinical validity refers to how consistently and accurately the test detects or predicts the intermediate or final outcomes of interest. Is what’s measured associated with the outcome of interest? Clinical utility refers to how likely the test is to significantly improve patient outcomes. What is the clinical value? ELSI refers to the ethical, legal and social implications that may arise in the context of using the test (CDC, 2010).

Thyroid Cancer
Thyroid cancer is most commonly found on routine physical examination as a palpable thyroid nodule. A fine-needle aspiration (FNA) biopsy is usually performed to rule out malignancy. In some cases, the nodules are not clearly benign or malignant based on FNA results alone. Those patients with cytologically indeterminate nodules are often referred for diagnostic surgery, though most of these nodules turn out to be benign. The Afirma gene expression classifier measures the expression of 142 genes to classify nodules as benign or suspicious for malignancy. Test results may help patients avoid unnecessary surgeries (Veracyte® website; ECRI, 2012).

Colon Cancer
There is disagreement over when adjuvant chemotherapy should be used for stage II colon cancers. Gene expression profiling has been proposed as a method for predicting which of these patients are likely to have a recurrence. The Oncotype DX Colon Cancer Assay is a 12-gene test that provides an individualized score reflective of the risk of colon cancer recurrence for individual patients with stage II colon cancer. Based on the biology of a patient’s specific colon cancer tumor, the test combines a multigene panel, which includes 7 colon cancer-related genes and 5 reference genes, with a proprietary algorithm for determining risk of recurrence (Genomic® Health website). ColoPrint® is a microarray-based gene expression profile for predicting the risk of distant recurrence of stage II and III colon cancer patients (Agendia® website).

Multiple Myeloma
Using microarray technology, My Prognostic Risk Signature (MyPRS™) and MyPRS Plus™ are proposed as tools for guiding treatment in patients with multiple myeloma. MyPRS analyzes all of the nearly 25,000 genes in a patient’s genome to determine the gene expression profile (GEP) that is associated with his/her condition. In the case of myeloma, the GEP is made up of the 70 most relevant genes (GEP70) which aid in the prediction of the patient’s outcome (Signal Genetics™ website).

Cancer of Unknown Primary
Cancers are treated according to their primary site. Accurately classifying the site of a tumor’s origin helps physicians choose the best course of treatment for the patient. Cancers of unknown primary, also referred to as occult primaries, are tumors that have metastasized from an unknown primary site. Gene expression profiling has been proposed as a tool for guiding diagnosis. The ResponseDX Tissue of Origin test compares the molecular profile in a patient’s tumor tissue sample with the profiles of 15 known tumor types (Response Genetics website). CancerTYPE ID uses real-time reverse transcription polymerase chain reaction (RT-PCR) to measure the gene expression of 87 genes associated with tumors and 5 reference genes (bioTheranostics website).
Uveal Melanoma
Uveal (ocular) melanoma is an aggressive cancer that often forms undetectable micrometastases before diagnosis of the primary tumor. The main goals of treatment are to reduce the risk of metastasis, prevent local growth and destruction of ocular tissues and preserve as much vision as possible. The DecisionDx-UM test is a multi-gene expression test that identifies patients who have a near term (5-year) low risk (Class 1 molecular signature) versus high risk (Class 2 molecular signature) of developing metastatic disease. Proponents of the test state that test results can be used to change the frequency and intensity of surveillance or offer prophylactic therapy for high risk patients (Castle Biosciences website).

Cardiovascular Risk Assessment
Gene expression profiling, using Corus CAD, has been proposed as a noninvasive diagnostic tool for evaluating patients with symptoms of coronary artery disease (CAD). Corus CAD is a blood test that integrates expression levels of 23 genes and other patient characteristics to predict the likelihood of obstructive CAD. According to the manufacturer, the test yields an objective result of cardiac risk in the form of a numeric score (0-40) that quantifies the likelihood that a patient with stable chest pain has obstructive CAD. The test is intended for nondiabetic patients with chest pain or other symptoms of obstructive CAD and no history of heart disease (CardioD X® website).

CLINICAL EVIDENCE

Thyroid Cancer
Alexander et al. (2014) analyzed all patients who had received Afirma GEC testing at five academic medical centers between 2010 and 2013. Three hundred thirty-nine patients underwent Afirma gene expression classifier (GEC) testing of cytologically indeterminate nodules (165 atypical (or follicular lesion) of undetermined significance; 161 follicular neoplasm; 13 suspicious for malignancy). Of these 339 nodules, 174 (51%) were GEC benign, and 148 were GEC suspicious (44%). GEC results significantly altered care recommendations, as 4 of 175 GEC benign were recommended for surgery in comparison to 141 of 149 GEC suspicious. Of 121 cytologically indeterminate, GEC suspicious nodules surgically removed, 53 (44%) were malignant. Seventy-one of 174 GEC benign nodules had documented clinical follow-up for an average of 8.5 months, in which 1 of 71 nodules proved cancerous. The authors concluded that this clinical experience data confirms originally published Afirma test performance and demonstrates its impact on clinical care recommendations.

In a prospective, multicenter clinical validation study involving 49 sites, 3789 patients and 4812 fine-needle aspirates from thyroid nodules, Alexander et al. (2012) assessed the performance of a gene-expression classifier (Afirma) on 265 cytologically indeterminate nodules. Of the 265 indeterminate nodules, 85 were malignant. The gene-expression classifier correctly identified 78 of the 85 nodules as suspicious (92% sensitivity), with a specificity of 52%. The negative predictive values for "atypia (or follicular lesion) of undetermined clinical significance," "follicular neoplasm or lesion suspicious for follicular neoplasm" or "suspicious cytologic findings" were 95%, 94% and 85%, respectively. Analysis of 7 aspirates with false negative results revealed that 6 had a paucity of thyroid follicular cells, suggesting insufficient sampling of the nodule. The authors concluded that these results suggest consideration of a more conservative approach for most patients with thyroid nodules that are cytologically indeterminate on fine-needle aspiration and benign according to gene-expression classifier results.

In a cross-sectional cohort study, Duick et al. (2012) demonstrated that obtaining a gene expression classifier (GEC) test (Afirma) in patients with cytologically indeterminate nodules was associated with a reduction in the rate of diagnostic thyroidectomies. The authors reported that approximately one surgery was avoided for every two GEC tests run on thyroid fine-needle aspirations (FNA) with indeterminate cytology. Data was contributed retrospectively by 51
endocrinologists at 21 practice sites. Compared to a 74% previous historical rate of surgery for cytologically indeterminate nodules, the operative rate fell to 7.6% during the period that GEC tests were obtained. The rate of surgery on cytologically indeterminate nodules that were benign by the GEC reading did not differ from the historically reported rate of operation on cytologically benign nodules. The four primary reasons reported by the physicians for operating on nodules with a benign GEC reading were, in descending order, large nodule size (46.4%), symptomatic nodules (25.0%), rapidly growing nodules (10.7%) or a second suspicious or malignant nodule in the same patient (10.7%). According to the authors, these reasons are concordant with those typically given for operation on cytologically benign nodules.

Walsh et al. (2012) verified the analytical performance of the Afirma gene expression classifier (GEC) in the classification of cytologically indeterminate thyroid nodule fine-needle aspirates (FNAs). Studies were designed to characterize the stability of RNA during collection and shipment, analytical sensitivity, analytical specificity and assay performance. The authors reported that analytical sensitivity, analytical specificity, robustness and quality control of the GEC were successfully verified, indicating its suitability for clinical use.

Chudova et al. (2010) based the Afirma gene expression classifier test on an empirical assessment of more than 247,000 mRNA transcripts associated with pathologically proven benign or malignant thyroid lesions.

National Comprehensive Cancer Network (NCCN) clinical practice guidelines state that molecular diagnostics to detect individual mutations, or pattern recognition approaches using molecular classifiers, may be useful in the evaluation of fine-needle aspiration (FNA) samples that are indeterminate. The diagnosis of follicular carcinoma or Hurthle cell carcinoma requires evidence of either vascular or capsular invasion, which cannot be determined by FNA. Molecular diagnostics may be useful to allow reclassification of follicular lesions (follicular neoplasm or follicular lesions of undetermined significance) as more likely to be benign or more likely to be malignant (NCCN, 2013).

Professional Societies
American Association of Clinical Endocrinologists/Associazione Medici Endocrinologi/European Thyroid Association
A joint guideline on the diagnosis and management of thyroid nodules addresses molecular testing, but it does not specifically discuss the use of a panel of markers. The guideline states that molecular and immunohistochemical markers may improve the accuracy of cytologic diagnosis, but they do not have consistent predictive value for malignancy and their use is still expensive and restricted to specialized centers. On the basis of current limited evidence, routine use of molecular and immunohistochemical markers in clinical practice is not recommended and should be reserved for selected cases (Gharib et al., 2010). Grade D (action not based on any evidence or not recommended); Best evidence level 3 (on a scale of 1 to 4).

American Thyroid Association (ATA)
An ATA guideline on the management of patients with thyroid nodules addresses molecular testing, but it does not specifically discuss the use of a panel of markers. The guideline states that the use of molecular markers may be considered for patients with indeterminate cytology on fine-needle aspiration to help guide management (ATA, 2009). Recommendation rating: C (based on expert opinion).

Colon Cancer
An Agency for Healthcare Research and Quality (AHRQ) technical brief states that, although information is emerging about the use of gene expression profiling (GEP) assays to inform the decision about use of adjuvant chemotherapy in patients with stage II colon cancer, studies to date have not provided the type of information needed to address major uncertainties. Published studies have not provided information related to clinical utility. Limited information was found for analytic validity. The report concluded that the current evidence does not provide the type of
Lu et al. (2009) performed a systematic review and meta-analysis of gene expression profiles (GEPs) to assess their utility for risk stratification and prediction of poor outcomes in stage II colorectal cancer (CRC). Eight cohorts involving 271 patients contributed to the analysis. The average accuracy, sensitivity and specificity were 81.9%, 76.2% and 84.5%, respectively, with a prognostic likelihood ratio (LR) of 4.7 and a prognostic odds ratio (OR) of 15.1. No evidence for significant interstudy heterogeneity was noted in either analysis. Subgroup analysis found no difference in results for the prediction of cancer recurrence or death. The authors concluded that GEP assays as predictors of poor outcomes in stage II CRC have promising potential, but to maximize their utility and availability, further studies are needed to identify and validate specific gene signatures.

**Oncotype DX**

Gray et al. (2011) developed a quantitative gene expression assay to assess recurrence risk and benefits from chemotherapy in patients with stage II colon cancer. These assays were validated using RNA extracted from fixed paraffin-embedded primary colon tumor blocks from 1,436 patients with stage II colon cancer in the QUASAR (Quick and Simple and Reliable) study. A recurrence score (RS) and a treatment score (TS) were calculated from gene expression levels of 13 cancer-related genes (n = 7 recurrence genes and n = 6 treatment benefit genes) and from five reference genes with prespecified algorithms. Recurrence risks at 3 years were 12%, 18% and 22% for predefined low, intermediate and high recurrence risk groups, respectively. T stage and mismatch repair (MMR) status were the strongest histopathologic prognostic factors. The TS was not predictive of chemotherapy benefit.

A validation study by Clark-Langone et al. (2010) describes the analytical performance of the Oncotype DX Colon Cancer Assay. The study illustrates the algorithm used to calculate the recurrence score and reports the assay’s performance regarding analytical sensitivity, analytical precision and reproducibility when used to test colon cancer specimens.

**ColoPrint**

Maak et al. (2013) conducted a validation study of the ColoPrint test for patients with stage II colon cancer. The assay was performed on 135 patients who underwent resection for stage II colon cancer. ColoPrint identified most stage II patients (73.3%) as at low risk. The 5-year distant-metastasis free survival was 94.9% for low-risk patients and 80.6% for high-risk patients.

Salazar et al. (2011) developed a gene expression classifier to predict disease relapse in patients with early-stage colorectal cancer (CRC). The authors used a cross-validation procedure to score all genes for their association with 5-year distant metastasis-free survival in patients with CRC. Frozen tumor tissue from 188 patients with stage I to IV CRC undergoing surgery were analyzed. The majority of patients (83.6%) did not receive adjuvant chemotherapy. An optimal set of 18 genes was identified and used to construct a prognostic classifier (ColoPrint). The signature was validated on an independent set of 206 samples from patients with stage I, II and III CRC. The signature classified 60% of patients as low risk and 40% as high risk. Five-year relapse-free survival rates were 87.6% and 67.2% for low- and high-risk patients, respectively, with a hazard ratio (HR) of 2.5. In multivariate analysis, the signature remained one of the most significant prognostic factors, with an HR of 2.69. In patients with stage II CRC, the signature had an HR of 3.34 and was superior to American Society of Clinical Oncology criteria in assessing the risk of cancer recurrence. The authors concluded that ColoPrint significantly improves the prognostic accuracy of pathologic factors in patients with stage II and III CRC and facilitates the identification of patients with stage II disease who may be safely managed without chemotherapy.

The Prospective Study for the Assessment of Recurrence Risk in Stage II Colon Cancer Patients Using ColoPrint (PARSC) is a validation study currently underway (NCT00903565). Additional information is available at:

National Comprehensive Cancer Network (NCCN) clinical practice guidelines state that there is insufficient data to recommend the use of multigene assay panels to determine adjuvant therapy in colon cancer patients (NCCN, 2014a).

**Multiple Myeloma**

A Mayo Clinic consensus statement on the management of newly diagnosed patients with multiple myeloma states that due to current lack of influence on therapy, gene expression profiling (GEP) is neither routinely performed nor recommended in a nonresearch setting. However, as commercial tests are being developed, GEP will likely play a greater role in the management of multiple myeloma in the future (Mikhael et al., 2013).

The International Myeloma Workshop Consensus Panel 2 published recommendations for risk stratification in multiple myeloma. The document states that a more robust and comprehensive analysis is needed to analyze the significance of risk stratification using comparative genomic hybridization/single nucleotide polymorphism array. In the future, a specific polymorphism may help identify patients with differential response profile and/or higher risk of toxicity. However, there is lack of data to propose any specific single nucleotide polymorphisms that can be used for such decisions (Munshi et al., 2011).

Shaughnessy et al. (2007) performed gene expression profiling on tumor cells from 532 newly diagnosed myeloma patients treated on 2 separate protocols. The goal was to identify a signature associated with shorter survival. Seventy genes linked to shorter durations of complete remission, event-free survival and overall survival were identified. A subset of patients with a high-risk score had a 3-year continuous complete remission rate of only 20%, as opposed to a 5-year continuous complete remission rate of 60% in the absence of a high-risk score. Further analysis identified a 17-gene subset that performed as well as the 70-gene model.

To better define the molecular basis of multiple myeloma, Zhan et al. (2006) performed gene expression profiling on plasma cells from 414 newly diagnosed patients who went on to receive high-dose therapy and tandem stem cell transplants. The group identified and validated seven disease subtypes based on common gene expression signatures. Select subgroups were associated with superior event-free and overall survival. It was noted that the development of therapies that target the molecular pathways unique to high-risk disease should be encouraged.

National Comprehensive Cancer Network (NCCN) clinical practice guidelines state that gene expression profiling has the potential to further refine risk-stratification, help therapeutic decisions and inform novel drug design and development. At the present time, standardized testing for gene expression profiling is not available and there is inadequate data to determine how this prognostic information should be used to direct patient management (NCCN, 2014b).

**Cancer of Unknown Primary**

An AHRQ technology assessment (2013) on testing cancers of unknown primary concluded the following:

- The clinical accuracy of commercially available molecular pathology tests is similar.
- The evidence that these tests contribute to identifying cancers of unknown primary is moderate.
- There is insufficient evidence to assess the effect of these tests on treatment decisions and outcomes.
- Most studies evaluated were funded wholly or partially by the manufacturers of the tests.

*Pathwork/ResponseDX Tissue of Origin*
As of April 2, 2013, Pathwork Diagnostics is no longer in business. Response Genetics has acquired all assets and intellectual property related to the Pathwork Tissue of Origin Test and is marketing the test as ResponseDX Tissue of Origin™ Test.

In a prospective, multicenter study, Handorf et al. (2013) compared the diagnostic accuracy of gene expression profiling (GEP) and immunohistochemistry (IHC) in identifying the primary site of metastatic tumors. Four pathologists rendered diagnoses by selecting from 84 stains in 2 rounds. Overall, GEP accurately identified 89% of specimens, compared with 83% accuracy using IHC. In a subset of 33 poorly differentiated and undifferentiated carcinomas, GEP accuracy exceeded that of IHC (91% to 71%). Further studies are needed to demonstrate that identifying the tissue of origin of unknown primary tumors leads to improvements in health outcomes.

Pillai et al. (2011) performed a validation study on the Pathwork Tissue of Origin Test, a gene expression-based diagnostic test that aids in determining the tissue of origin using formalin-fixed, paraffin-embedded (FFPE) specimens. Microarray data files were generated for 462 metastatic, poorly differentiated, or undifferentiated FFPE tumor specimens, all of which had a reference diagnosis. The microarray data files were analyzed using a 2000-gene classification model. The algorithm used for the test quantifies the similarity between RNA expression patterns of the study specimens and the 15 tissues on the test panel. Among the 462 specimens analyzed, overall agreement with the reference diagnosis was 89%. Further studies are needed to determine how test results change patient management and impact clinical outcomes.

Monzon et al. (2010b) evaluated the ability of a microarray-based gene expression test to identify the tissue of origin (TOO) in tumor specimens from 21 patients with a diagnosis of carcinoma of unknown primary (CUP). The Pathwork TOO Test was used to measure gene expression patterns for 1550 genes. These were compared for similarity to patterns from 15 known tissue types. The TOO Test yielded a clear single positive call for the primary site in 16 of 21 (76%) specimens and was indeterminate in 5 (24%). The positive results were consistent with clinicopathologic suggestions in 10 of the 16 cases (62%). In the remaining six cases the positive results were considered plausible based on clinical information. Positive calls included colorectal (5), breast (4), ovarian (3), lung (2) and pancreas (2). The Pathwork TOO Test reduced diagnostic uncertainty in all CUP cases and could be a valuable addition or alternative to current diagnostic methods for classifying uncertain primary cancers. Further studies are needed to determine how test results change patient management and impact clinical outcomes.

Monzon et al. (2009) conducted a large, blinded, multicenter validation study for the Pathwork Tissue of Origin (TOO) test, which consists of a test panel and a proprietary algorithm. Four separate laboratories processed 547 frozen specimens representing 15 tissues of origin using microarrays. Half of the specimens were metastatic tumors, with the remainder being poorly differentiated and undifferentiated primary cancers. The study found an overall sensitivity of 87.8% and an overall specificity of 99.4%. The test performed best using the undifferentiated and indeterminate tissue samples (n=289), yielding 90.7% agreement with the original diagnosis. Whereas the metastatic tissue samples (n=258) resulted in 84% agreement. The four facilities reported slightly different overall agreement percentages, but none of the differences were statistically significant. Results suggest that the test is sufficiently sensitive and informative for routine diagnostic use in patients presenting with uncertain primary cancers. Further studies are needed to determine how test results change patient management and impact clinical outcomes.

National Institute for Health and Clinical Excellence (NICE) guidelines state that gene expression-based profiling should not be used to identify primary tumors or guide treatment decisions in patients with carcinoma of unknown primary (CUP). There is limited evidence that gene expression-based profiling improves the management or changes the outcomes for patients with CUP. Prospective randomized trials should be undertaken in patients with confirmed CUP to evaluate whether chemotherapy guided by gene-expression-based profiling is superior to treatment guided by conventional clinical and pathological factors. The guideline noted that this is a rapidly changing field (NICE, 2010).
National Comprehensive Cancer Network (NCCN) clinical practice guidelines state that several gene expression profiling tests are being evaluated in prospective clinical studies in an attempt to determine if the information they provide translates into clinically meaningful benefit for patients. The clinical utility of these tests remains to be determined (category 2B). Consequently, the panel does not recommend molecular profiling for the identification of tissue origin as standard management in the diagnostic workup of patients with cancer of unknown primary (NCCN, 2014c).

**CancerTYPE ID**

In a prospective study, Hainsworth et al. (2013) used tumor profiling results to direct site-specific therapy for patients with carcinoma of unknown primary (CUP). Tumor biopsy specimens from previously untreated patients with CUP were tested with a 92-gene reverse transcriptase polymerase chain reaction cancer classification assay (CancerTYPE ID). When a tissue of origin was predicted, patients who were treatment candidates received standard site-specific first-line therapy. Of 289 patients enrolled, 252 had successful assays performed, and 247 (98%) had a tissue of origin predicted. Sites most commonly predicted were biliary tract (18%), urothelium (11%), colorectal (10%) and non-small-cell lung (7%). Two hundred twenty-three patients were treatment candidates, and 194 patients received assay-directed site-specific treatment. In these 194 patients, the median survival time was 12.5 months. When the assay predicted tumor types that were clinically more responsive, the median survival was significantly improved when compared with predictions of more resistant tumors (13.4 v 7.6 months, respectively). The authors concluded that molecular tumor profiling predicted a tissue of origin in most patients with CUP but noted that larger numbers of patients are required to make definitive statements regarding therapeutic implications of individual primary site predictions.

Kerr et al. (2012) conducted a multisite validation study to determine performance characteristics of a 92-gene molecular cancer classifier (CancerTYPE ID). Case selection incorporated specimens from more than 50 subtypes, including a range of tumor grades, metastatic and primary tumors and limited tissue samples. The assay showed overall sensitivities of 87% for tumor type and 82% for subtype. Analyses of metastatic tumors, high-grade tumors or cases with limited tissue showed no decrease in comparative performance. High specificity (96%-100%) was showed for ruling in a primary tumor in organs commonly harboring metastases. The assay incorrectly excluded the adjudicated diagnosis in 5% of cases. The authors concluded that results of this validation study support the clinical utility of the 92-gene assay in tumors of uncertain origin as a molecular adjunct to clinicopathologic evaluation for primary site diagnosis, discrimination between primary and metastatic tumor in common metastatic sites and for tumor subclassification. Prospective studies will help further define how molecular data can be successfully integrated into the clinical decision making process and allow for increased diagnostic certainty.

Erlander et al. (2011) reported the expansion of a second-generation gene expression profiling test (CancerTYPE ID) and demonstrated the ability of the 92-gene assay to classify 30 cancer types and 54 histological subtypes. For main cancer type, the sensitivity was 87% with a specificity of 100%, resulting in a positive predictive value (PPV) of 87% and a negative predictive value (NPV) of 100%. The accuracy for cancer subtype was a sensitivity of 85% and a specificity of 100%, resulting in a PPV of 85% and NPV of 100%. The authors also evaluated an additional 300 consecutive cases submitted for clinical testing to characterize clinical utility in a real-world setting: the 92-gene assay confirmed 78% of samples having a single suspected primary tumor and provided a single molecular prediction in 74% of cases with two or more differential diagnoses. To firmly establish the clinical validity of the 92-gene assay, a multi-institutional study is ongoing to determine the analytical performance within many diverse cancer types. In addition, prospective studies are being conducted to assess whether the use of the predictions from the 92-gene assay to select treatment positively affects patient outcome.

**Uveal Melanoma**
In a prospective multi-center validation study, Onken et al., (2012) evaluated the prognostic performance of a 15 gene expression profiling (GEP) assay that assigned primary posterior uveal melanomas to prognostic subgroups: class 1 (low metastatic risk) and class 2 (high metastatic risk). A total of 459 patients were enrolled. Analysis was performed to compare the prognostic accuracy of GEP with Tumor-Node-Metastasis (TNM) classification and chromosome 3 status. Patients were managed for their primary tumor and monitored for metastasis. The GEP assay successfully classified 446 of 459 cases (97.2%). The authors concluded that the GEP assay had a high technical success rate and was the most accurate prognostic marker among all of the factors analyzed. The GEP provided a highly significant improvement in prognostic accuracy over clinical TNM classification and chromosome 3 status. Further studies are needed to determine the clinical utility of these tests and the role they have in clinical decision-making.

To make the test more clinically practical, it was migrated from a microarray platform to a polymerase chain reaction (PCR)-based 15-gene assay. Onken et al., (2010) analyzed the technical performance of the assay in a prospective study of 609 tumor samples, including 421 samples sent from distant locations. Preliminary outcome data from the prospective study affirmed the prognostic accuracy of the assay.

Worley et al. (2007) compared the gene expression profile (molecular signature) to the chromosome 3 marker (monosomy 3) for predicting metastasis in 67 primary uveal melanomas. The gene expression-based molecular classifier assigned 27 tumors to class 1 (low risk) and 25 tumors to class 2 (high risk). Advanced patient age and scleral invasion were the only clinicopathologic variables significantly associated with metastasis. A less significant association with metastasis was observed for monosomy 3 detected by array comparative genomic hybridization (aCGH) and fluorescence in situ hybridization (FISH). The sensitivity and specificity for the molecular classifier (84.6% and 92.9%, respectively) were superior to monosomy 3 detected by aCGH (58.3% and 85.7%, respectively) and FISH (50.0% and 72.7%, respectively). Positive and negative predictive values (91.7% and 86.7%, respectively) and positive and negative likelihood ratios (11.9 and 0.2, respectively) for the molecular classifier were also superior to those for monosomy 3. The authors concluded that molecular classification based on gene expression profiling of the primary tumor was superior to monosomy 3 and clinicopathologic prognostic factors for predicting metastasis in uveal melanoma.

In 2004, Onken et al., reported that primary uveal melanomas cluster into two distinct molecular classes based on gene expression profile: class 1 (low-grade) and class 2 (high-grade). The authors found that this molecular classification strongly predicted metastatic death and outperformed other clinical and pathological prognostic indicators.

National Comprehensive Cancer Network (NCCN) clinical practice guidelines do not address uveal melanoma.

**Cardiovascular Risk Assessment**

In the IMPACT-CARD study, McPherson et al. (2013) assessed the impact of gene expression testing (Corus CAD) on clinical decision-making in patients with symptoms of suspected coronary artery disease (CAD) presenting to the cardiology setting. The study included a prospective cohort of 83 patients eligible for analysis, including 57 (69%) women. These patients were referred to six cardiologists for evaluation of suspected CAD and were matched to 83 patients in a historical cohort. The cardiologist’s diagnostic strategy was evaluated before and after gene expression score (GES) testing. The primary objective of the study was to measure whether the use of the GES changed the cardiologist’s evaluation and management of the patient. After GES, changes in diagnostic testing occurred in 58% of patients (n = 48). Of note, 91% (29/32) of patients with decreased testing had low GES (≤ 15), whereas 100% (16/16) of patients with increased testing had elevated GES. The historical cohort had higher diagnostic test use compared with the post-GES prospective cohort. The authors concluded that the GES showed clinical utility in the evaluation of patients with suspected obstructive CAD presenting to the cardiologist’s office. A potential for bias exists due to manufacturer sponsorship of the study.
Additional limitations include short term follow-up, small sample size and inclusion of individuals at low risk for CAD. Clinical trial # NCT01251302.

In a companion study (IMPACT-PCP), Herman et al. (2014) assessed the impact of gene expression testing (Corus CAD) on clinical decision-making in patients with symptoms of suspected coronary artery disease (CAD) presenting to a primary care setting. Providers initially determined patients’ pretest probability for CAD based on risk factors, assessment of clinical symptoms and results of any prior testing. All patients underwent gene expression score (GES) testing, with clinicians documenting their planned diagnostic strategy both before and after GES. The primary objective was to assess whether the use of GES altered patient management. The study enrolled 261 consecutive stable, nonacute, nondiabetic patients presenting with typical and atypical symptoms of CAD. Of the 251 eligible study patients, 140 were women (56%). After 30 days, a change in the diagnostic plan before and after GES testing was noted in 145 patients (58%). More patients had decreased (n=93, 37%) versus increased (n=52, 21%) intensity of testing. In particular, among the 127 low score Corus CAD patients (51% of study patients), 60% (76/127) had decreased testing, and only 2% (3/127) had increased testing. The authors concluded that the incorporation of GES into the diagnostic workup showed clinical utility above and beyond conventional clinical factors by optimizing the patient’s diagnostic evaluation. A potential for bias exists due to manufacturer sponsorship of the study. Additional limitations include short term follow-up, modest sample size and inclusion of individuals at low risk for CAD. Clinical trial # NCT01594411.

The prospective, multicenter COMPASS validation study (Thomas et al., 2013) evaluated the performance of the Corus CAD test in symptomatic patients referred for myocardial perfusion imaging (MPI). Blood samples for gene expression scoring (GES) were obtained prior to MPI. Based on MPI results, 431 patients were referred for either invasive coronary angiography or computed tomographic angiography. Patients were followed for 6 months with clinical end points defined as major adverse cardiac events. Sensitivity, specificity and negative predictive value were reported at 89%, 52% and 96%, respectively. The GES outperformed clinical factors and showed significant correlation with maximum percent stenosis (≥50%). Six-month follow-up on 97% of patients showed that 27 of 28 patients with adverse cardiovascular events or revascularization had GES >15. The authors concluded that GES has high sensitivity and negative predictive value for obstructive coronary artery disease. In this population clinically referred for MPI, the GES outperformed clinical factors and MPI. A potential for bias exists due to manufacturer sponsorship of the study. Additional limitations include short term follow-up and inclusion of individuals at low risk for CAD. Clinical trial #NCT01117506.

The PREDICT (Personalized Risk Evaluation and Diagnosis in the Coronary Tree) trial was a prospective, multicenter validation study of a peripheral blood-based gene expression test for determining the likelihood of obstructive coronary artery disease (CAD). Patients with chronic inflammatory disorders, elevated levels of leukocytes or cardiac protein markers or diabetes were excluded. Blood samples were obtained from 526 patients with chest pain or another indication for coronary angiography. Obstructive CAD was defined as 50% or greater stenosis in 1 or more major coronary arteries by quantitative coronary angiography. The sensitivity and specificity for the gene expression test were 85% and 43% respectively. The investigators reported a statistically significant but modest improvement in classification of patient CAD status compared with clinical factors or noninvasive imaging (myocardial perfusion imaging). Further studies are needed to define the performance characteristics and clinical utility of these tests in the general population (Rosenberg et al., 2010). A potential for bias exists due to manufacturer sponsorship of the study. Clinical trial #NCT00500617.

In a follow-up to the PREDICT study, Rosenberg et al. (2012) evaluated the relationship between gene expression testing and both major adverse cardiovascular events (MACE) and revascularization. A cohort of the original trial (n=1,116) underwent angiography and gene expression scoring (GES), and was followed for 1 year. A total of 267 (23.9%) patients had clinical endpoints within 30 days of testing. An additional 25 (2.2%) patients had clinical endpoints
within a year. Overall, the rate of MACE was 1.5% for 12 months. Using a GES cutoff of ≤ 15 (i.e.,
low likelihood of CAD), the sensitivity, specificity, PPV and NPV for MACE or revascularization
within 12 months of testing were 86%, 41%, 33% and 90%, respectively. The authors concluded
that a low GES appeared to identify individuals at low risk for both obstructive coronary artery
disease and subsequent procedures or events. The authors noted several limitations to the study
including limited follow-up and exclusion of patients with high-risk unstable angina and low-risk
asymptomatic patients. Further studies with larger patient populations and long-term outcomes
are needed.

In an additional analysis of the PREDICT study, Lansky et al. (2012) reported that Corus CAD
performed similarly in women and men.

Using a series of microarray and real-time polymerase chain reaction (RT-PCR) data sets,
comprising more than 1000 patients, Elashoff et al. (2011) developed a blood-based gene
expression algorithm for assessing obstructive coronary artery disease (CAD) in non-diabetic
patients. The algorithm consists of the expression levels of 23 genes, sex and age.

Wingrove et al. (2008) performed a microarray analysis on 41 patients with angiographically
significant coronary artery disease (CAD) and 14 controls without coronary stenosis to identify
genes expressed in peripheral blood that may be sensitive to the presence of CAD. A multistep
approach was used, starting with gene discovery from microarrays, followed by real-time
polymerase chain reaction (RT-PCR) replication. The authors observed that gene expression
scores based on 14 genes, independently associated with the presence or absence of CAD, were
proportional to the extent of disease burden. This study is limited by its size and retrospective
nature. Larger, prospective studies are needed to confirm these initial results.

The Evaluation of Genomic Applications in Practice and Prevention (EGAPP) Working Group
found insufficient evidence to recommend genomic profiling for assessing cardiovascular risk.
The group reported that clinical evidence was inadequate to determine analytic validity, clinical
validity or clinical utility. Clinical use of these tests is discouraged until further evidence supports
improved clinical outcomes (EGAPP, 2010).

The U.S. Preventive Services Task Force (USPSTF) recommendations on the use of
nontraditional risk factors in coronary heart disease risk assessment do not address
genetic/genomic markers (USPSTF, 2009).

Professional Societies

American College of Cardiology (ACC)
ACC guidelines do not address gene expression profiling specifically; however, the guidelines do
state that genotype testing for coronary heart disease risk assessment in asymptomatic adults is
not recommended as it offers no proven benefit (Greenland et al. 2010). (Level of Evidence: B –
data derived from a single randomized trial or nonrandomized studies.)

American Heart Association (AHA)
Although evidence exists linking common variants to complex cardiovascular disease, studies are
not yet available to inform the clinical benefit of providing such genetic information to patients.
Skepticism remains about the clinical utility for risk prediction. It is not always clear how the
genotype results can or should influence clinical management. In addition, no clinical trials have
been performed that demonstrate the benefit of genotyping in influencing clinical outcomes
(Ashley et al., 2012).

In a published scientific statement on the relevance of genetics and genomics for the prevention
and treatment of cardiovascular disease (CVD), the AHA states that RNA gene expression
profiling shows great promise. However, further results from large, patient cohorts are needed to
determine the clinical utility of this methodology. The statement also proposes several
recommendations to guide future research (Arnett et al. 2007).
In a separate scientific statement, the AHA proposes a set of criteria for evaluating novel markers of cardiovascular risk and determining their clinical value (Hlatky et al., 2009).

**U.S. FOOD AND DRUG ADMINISTRATION (FDA)**


Microarrays and next-generation sequencing represent core technologies in pharmacogenomics and toxicogenomics; however, before these technologies can successfully and reliably be used in clinical practice and regulatory decision-making, standards and quality measures need to be developed. The MicroArray Quality Control (MAQC) project is helping improve the microarray and next-generation sequencing technologies and foster their proper applications in discovery, development and review of FDA regulated products. Additional information is available at: [http://www.fda.gov/ScienceResearch/BioinformaticsTools/MicroarrayQualityControlProject/default.htm](http://www.fda.gov/ScienceResearch/BioinformaticsTools/MicroarrayQualityControlProject/default.htm). Accessed February 20, 2014.

The original version of the Tissue of Origin Test (Pathwork® Diagnostics) received FDA approval (K080896) on July 30, 2008. A second version of the test (K092967) was approved on June 8, 2010. The test is an in-vitro diagnostic intended to measure the degree of similarity between the RNA expression patterns in a patient's formalin fixed, paraffin embedded (FFPE) tumor and the RNA expression patterns in a database of fifteen tumor types (poorly differentiated, undifferentiated and metastatic cases) that were diagnosed according to then current clinical and pathological practice.

The test is not intended to do any of the following:

- establish the origin of tumors that cannot be diagnosed according to current clinical and pathological practice
- subclassify or modify the classification of tumors that can be diagnosed by current clinical and pathological practice
- predict disease course, survival or treatment efficacy
- distinguish primary from metastatic tumor.

See the following websites for more information:


**Additional Products**

ColoPrint®

**CENTERS FOR MEDICARE AND MEDICAID SERVICES (CMS)**

Medicare does not have a National Coverage Determination (NCD) for gene expression tests. Local Coverage Determinations (LCDs) exist. Refer to the LCDs for Gene Expression Profiling Panel for use in the Management of Breast Cancer Treatment and MyPRS Genetic Expression Profile Testing. (Accessed January 31, 2014)
REFERENCES


POLICY HISTORY/REVISION INFORMATION

<table>
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| 05/01/2014 | • Reorganized policy content  
             • Added benefit considerations language for Essential Health Benefits for Individual and Small Group plans to indicate:  
             • For plan years beginning on or after January 1, 2014, the Affordable Care Act of 2010 (ACA) requires fully insured non-grandfathered individual and small group plans (inside and outside of Exchanges) to provide coverage for ten categories of Essential Health Benefits (“EHBs”)  
             • Large group plans (both self-funded and fully insured), and small group ASO plans, are not subject to the requirement to offer coverage for EHBs; however, if such plans choose to provide coverage for benefits which are deemed EHBs (such as maternity benefits), the ACA requires all dollar limits on those benefits to be removed on all Grandfathered and Non-Grandfathered plans  
             • The determination of which benefits constitute EHBs is made on a state by state basis; as such, when using this guideline, it is important to refer to the enrollee’s specific plan document to determine benefit coverage  
             • Updated coverage rationale:  
               o Added language to indicate if service is “medically necessary” or “not medically necessary” to applicable proven/unproven statement  
               o Replaced reference to “Pathwork® Diagnostics” with “ResponseDX”  
             • Updated supporting information to reflect the most current description of services, clinical evidence, FDA and CMS information, and references  
             • Archived previous policy version 2014T0552D |