August 12, 2016

National Government Services
LCD Comments
P.O. Box 7108
Indianapolis, IN 46207-7108
PartBLCDComments@anthem.com

Re: Draft Local Coverage Determination Molecular Pathology Procedures (DL35000), for MAC jurisdiction 6 and jurisdiction K.

Dear Dr. Clark and Dr. Cunningham,

Thank you for the opportunity to comment on this draft local coverage determination policy. The Association for Molecular Pathology (AMP) is an international medical and professional association representing approximately 2,300 physicians, doctoral scientists, and medical technologists who perform or are involved with laboratory testing based on knowledge derived from molecular biology, genetics, and genomics. Membership includes professionals from the government, academic medicine, private and hospital-based clinical laboratories, and the in vitro diagnostics industry.

The College of American Pathologists (CAP) is a national medical specialty society representing more than 18,000 physicians who practice anatomic and/or clinical pathology. College members practice their specialty in clinical laboratories, academic medical centers, research laboratories, community hospitals and federal and state health facilities.

Members of both AMP and CAP are experts in molecular pathology and the implementation of this coverage policy will directly impact their practices. We are submitting joint comments because at this time both of our organizations share the same concerns regarding this draft LCD.

Before commenting on the specifics of the proposed policy, we would like to make a recommendation to inform NGS' future policies. This policy as drafted is extremely broad in scope, covering the entire field of molecular pathology. The breadth of this policy makes it difficult for AMP and CAP to develop a meaningful response. We request that NGS consider developing future policies by disease state, narrowing the scope of both the policy being developed and our response. Another benefit to this change is NGS would not have to revise this entire broad policy as this field evolves. Given the rapid advancement of the science, NGS is faced with revising one general policy to address discrete changes that may be confined to disease state rather than a more targeted policy.

We thank NGS considering previous comments, for adding indications for therapeutic decision making (eg PDGFRA in GIST) and for making appropriate exceptions to one in a lifetime testing in appropriate clinical situations (Exceptions include clinical scenarios whereby repeat testing of somatically-acquired mutations (for example , pre- and post- therapy) may be required to inform appropriate therapeutic decision-making."

The following comments illustrate several significant coverage concerns about this dLCD and its potential negative impact on patient care. We have additionally included an appendix of background data specific to

Acute Leukemia and Myeloid Neoplasm/Leukemia to provide additional evidence and support for the requests listed below. We request that NGS consider the recommendations outlined in this letter.

I. INDICATIONS

dLCD "Indications" statement: Molecular pathology procedures (Tier1 and Tier 2) may be eligible for coverage when ALL of the following criteria are met:

Bullet #4: Results of the testing must directly impact treatment or management of the Medicare beneficiary;

Recommendation: Appropriate patient management is absolutely dependent on a correct DIAGNOSIS. We therefore recommend adding the term "diagnosis" to this indications requirement to read:

"*Results of the testing must directly impact DIAGNOSIS, treatment or management of the Medicare beneficiary*;"

In a previous NGS response to DL35000 comments (see Response to Comments: Molecular pathology Procedures A54825) "NGS requires that the results of the testing must directly impact the treatment of management of the Medicare beneficiary, not to determine a diagnosis." This position appears to be in conflict with Title XVIII of the Social Security Act (SSA) which states "Section 1862(a)(1)(A) excludes expenses incurred for items or services which are not reasonable and necessary for the DIAGNOSIS or treatment of illness or injury or to improve the functioning of a malformed body member (see LCD DL35000)."

We contend that an accurate diagnosis is required for therapeutic decision making and provide the following example for consideration. Note that other examples can be found in the text of this letter.

Equivocal or non-diagnostic morphological results from a tissue biopsy or fine needle aspirate (eg skin biopsy):

In instances when morphology (pathologist review of tissue on slide) fails to provide a diagnosis, ancillary testing is necessary to make the diagnosis which will guide therapeutic decision making
1). A diagnosis of lymphoma is required to determine the appropriate therapy (see NCCN Lymphoma guidelines). A positive TRG@ (T cell antigen receptor gamma) assay supports a diagnosis of lymphoma (eg Mycosis Fungoides). Failure to make a diagnosis delays treatment and could result in tumor progression from local to disseminated disease. Localized disease (eg skin) can be treated with Topical corticosteroids whereas metastatic disseminated disease (eg sezary disease) requires systemic treatment (eg chemotherapy) with a low probability of cure. The negative impact of withholding a diagnosis would be severe for the patient in this instance.

II. TIER 1 AND TIER 2 INDICATIONS AND LIMITATIONS OF COVERAGE:

CPT Code 81170
ABL1 (ABL proto-oncogene 1, non-receptor tyrosine kinase) (eg, acquired imatinib tyrosine kinase inhibitor resistance), gene analysis, variants in the kinase domain is considered medically necessary in patients with acute lymphoblasic leukemia (ALL) and chronic lymphoblastic leukemia (CLL) to guide therapeutic decision making.

Recommendation. ABL kinase domain sequencing is indicated in patients with ALL and chronic myeloid leukemia (chronic "lymphoblastic" leukemia is presumably a typographic error).

CPT Codes 81206, 81207, and 81208 (BCR/ABL)

dLCD Statement: BCR/ABL is considered medically necessary in the evaluation of individuals with chronic myelogenous leukemia or BCR-ABL positive acute lymphoblastic leukemia to evaluate treated individuals who

manifest suboptimal response to initial tyrosine kinase inhibitor therapy or loss of response to tyrosine kinase inhibitor therapy.

Recommendation: To conform to current practice, we request that this statement be amended to read:

"BCR/ABL is considered medically necessary in the evaluation of individuals with chronic myelogenous leukemia or acute myeloid Leukemia or BCR-ABL positive acute lymphoblastic leukemia to evaluate treated individuals **response** to tyrosine kinase inhibitor therapy or identify loss of response to tyrosine kinase inhibitor therapy." BCR/ABL is considered medically necessary to identify patients who should be treated with a tyrosine kinase inhibitor (eg imatinib)

Molecular testing is necessary to distinguish acute myeloid leukemia with BCR-ABL1 from other myeloproliferative diseases and leukemias because these patients may benefit from tyrosine-kinase inhibitor (TKI) therapy (see 2016 WHO revision).

Recommendation: Besides providing coverage for BCR-ABL testing using CPT codes 81206, 81207, and 81208, we recommend that NGS provide coverage for BCR-ABL testing (and all of the other single genes that could be part of a standard of care evaluation for the diagnosis of a myeloproliferative diseases) when they are included in a multi-gene panel. In addition to BCR-ABL, the other genes that are minimally necessary for a comprehensive evaluation of a patient with a suspected myeloproliferative disease (or MPN/MDS disease like CMML) include BCR-ABL, JAK2 (81270), CALR (81219), MPL (81402), CSF3R (81479), and SETBP1 (81479) , as per the 2016 revision to the World Health Organization (WHO) classification of myeloid neoplasms and acute leukemia (Arber et al, Blood First Edition Paper, prepublished online April 11, 2016; DOI 10.1182/blood-2016-03-643544).

Given that multi-gene analysis by next-generation sequencing (rather than sequential or parallel single-gene testing) is often performed as a cost-effective, time-saving, tissue-saving method for the evaluation of a patient with a suspected myeloproliferative disorder, we therefore strongly recommend that CPT code 81450 (5-50 gene hematolymphoid targeted genomic sequencing) be removed from group 5 (non-covered genomic sequencing procedures) and added to group 2 (procedures that require individual review). Consequently, please add coverage for:

C92.20	Atypical chronic myeloid leukemia, BCR/ABL-negative, not having achieved remission
C92.21	Atypical chronic myeloid leukemia, BCR/ABL-negative, in remission
C92.22	Atypical chronic myeloid leukemia, BCR/ABL-negative, in relapse
D45	Polycythemia vera
D47.1	Chronic myeloproliferative disease
D47.3	Essential (hemorrhagic) thrombocythemia
D75.1	Secondary polycythemia
D75.81	Myelofibrosis
D75.89	Other specified diseases of blood and blood-forming organs

CPT Code 81210 (BRAF)

Additional clinical indications beyond those listed in the draft LCD require BRAF testing. The LCD defines the Tier 1 and Tier 2 indications and limitations of coverage. Guidelines from professional societies (including NCCN) include additional indications which directly impact diagnosis, treatment, and management of the Medicare beneficiary. Published peer reviewed medical literature is cited below to support the use of molecular assays in these situations. Further, molecular testing may sometimes be performed more than once in a lifetime especially in instances when residual disease monitoring of somatic mutations is used to measure response to therapy. We request additional indications and limitations of coverage be added to LCD DL3500. We offer the following rationale and literature references to support the request for each individual CPT code (please see details listed below by CPT code).

BRAF testing is also critical in the diagnosis and treatment of other malignancies besides melanoma and NSCLC. The list of other allowable covered diagnoses (including "rule out" diagnoses) needs to minimally include:

- Colon (and other intestinal) cancers (as per NCCN guidelines):
- Thyroid cancer (papillary thyroid carcinoma) in equivocal thyroid cytology specimens (as per NCCN and American Thyroid Association Management guidelines)
- Intraductal Papillary Mucinous Neoplasm (IPMN) of the pancreas: (per NCCN guidelines).

Details are provided below.

1. NCCN Colon Version 2,2016 guidelines state small bowel and appendiceal carcinoma can be treated with systemic chemotherapy according to NCCN guidelines for colon cancer. Therefore, the same molecular testing required for colon cancer will be required for these cancers. For example a patient with BRAF positive appendiceal carcinoma would not be eligible for therapies targeting EGFR (e.g. cetuximab) (NCCN Oncology Guidelines: Colon Cancer, 2016).

2. BRAF V600* positive status in microsatellite positive colon cancer is used to differentiate between sporadic inactivation of the mismatch repair gene MLH1 versus inactivation of MLH1 by a hereditary germ line mutation (Lynch syndrome). Lynch syndrome colon cancer patients have different surgical management and follow up monitoring than patients with sporadic microsatellite positive colon cancer (NCCN Guidelines in Oncology: Genetic/Familial High-Risk Assessment: Colorectal, 2015).

3. Patients with BRAF V600* positive disease other than melanoma (e.g. lung adenocarcinoma, Erdheim-Chester disease, PLCH, glioblastoma, multiple myeloma) may be eligible for off-label use of BRAF V600 inhibitors (Andrulis, 2013; Haroche, 2013; Robinson, 2014).

4. BRAF V600* or BRAF K601* positivity is used to confer a positive diagnosis (e.g. carcinoma) in equivocal surgical biopsies, or cytology specimens (fine needle aspirates) or equivocal flow cytometry specimens (blood/bone marrow) (NCCN non-Hodgkin's lymphoma guidelines, 2016).

5.

a) A positive diagnosis of papillary thyroid carcinoma in equivocal thyroid cytology specimens (2016 ver 1 NCCN Guidelines for Oncology: Thyroid Carcinoma; Nikiforova, 2011).

   A BRAF mutation confers a positive diagnosis of papillary thyroid carcinoma in fine needle aspirates (FNA) showing equivocal thyroid cytology (eg atypical/suspicious ,morphology).36, 39 Mutation profiling in patients with equivocal thyroid cytology guides clinical decision making. 1) Patients who are negative for a mutation will be monitored and can be spared a thyroidectomy improving quality of life. Patients who are positive for a mutation will receive a complete thyroidectomy (see 2016 NCCN guidelines THYR-3,4, PAP-1), . Patients who are denied mutation testing are monitored, subjected to multiple FNA procedures and if multiple FNAs are equivocal will receive a partial thyrodectomy (lobectomy) (see 2016 NCCN guidelines THYR-4),. If the lobectomy shows a carcinoma, the patient will undergo a second surgery to remove the remaining thyroid tissue at added expense (see 2016 NCCN guidelines PAP-2, FOLL-1, HURT-1). If the partial thyrodectomy does not show a carcinoma, the surgery might be considered an unnecessary cost and reduction in the patient's quality of life. (2016 ver 1 NCCN Guidelines for Oncology: Thyroid Carcinoma; Haugen et al, 2015, Nikiforova, 2011).

b) A positive diagnosis of Pulmonary Langerhans histiocytosis in equivocal lung biopsy of fine needle aspirate (Berres, 2011; Roden, 2014).

c) A positive diagnosis of Erdheim-Chester disease in equivocal specimens (Diamond, 2014).

d) A positive diagnosis of hairy cell leukemia in specimens with equivocal flow cytometry results (i.e. to distinguish classical hairy cell leukemia (HCL) from HCL-variant which has a poor prognosis and requires a different treatment approach)(NCCN Oncology Guidelines: Non Hodgkins Lymphoma, 2016; Shao, 2013; Verma, 2012). (Current LCD includes an indication for BRAF testing in HCL, should we delete this? LH

e) A positive diagnosis of Intraductal Papillary Mucinous Neoplasm (IPMN) in pancreatic cyst fluid with equivocal cytology results. NCCN guidelines state “Endoscopic Ultrasound plays a role in better characterizing cystic pancreatic lesions due to the ability to aspirate the cyst contents for cytologic, biochemical and molecular analysis.”NCCN guidelines state "Intraductal papillary mucinous neoplasms (IPMNs) of the pancreas are cystic lesions"..."with the risk malignancy at around 62%" (NCCN Guidelines: Pancreatic Adenocarcinoma, 2014; Schonleblen, 2008; Thiruvengadam, 2015). 45,51

f) A positive diagnosis of a biliary tract neoplasm in equivocal cytology specimens (e.g biliary tract brushing) (Borger, 2012; Kipp, 2012).

g) BRAF testing can distinguish hepatocellular carcinoma from hepatocellular adenoma. These entities have different surgical management (Nault, 2013).

CPT Codes 81211, 81212, 81213, 81214, 81215, 81217 (BRCA1/2)

BRCA1/2 testing is now considered essential for predicting sensitivity to the new PARP inhibitor drugs. The FDA has mandated testing of BRCA1/2 as part of the labeling for the PARP inhibitor drug olaparib. Olaparib was approved by the U.S. Food and Drug Administration in 2014 for treatment of patients with advanced ovarian cancer who specifically have a BRCA1/2 mutation.

Other tumors also respond to PARP inhibitors, including: melanoma, prostate, lung, and other advanced solid tumors (Mateo, 2015; Kaufman, 2015).

Clinical trial enrollment for various PARP inhibitor therapies is now common in so-called "basket" trials for patients with a BRCA1/2 mutation, regardless of their cancer type (Kaufman, 2015).

Recommendation: We recommend the following edits to your current draft in order to better address the needs of patients in regard to BRCA 1/2 testing:

In situations where patients with a personal history of breast or another BRCA-related malignancy have been adopted or do not otherwise have access to accurate family health information, we recommend clarification on coverage for BRCA1 and BRCA2 testing. These individuals should be covered for this testing.

We recommend that BRCA1 testing needs to occur in a variety of tumor types other than just breast and ovarian cancer to predict responses to PARP inhibitor therapies. At a minimum, this list should include melanoma, colon, prostate, and pancreatic cancer.

CPT Code 81218 (CEBPA)

Please consider adding coverage for CEBPA testing for Myelodysplastic Syndrome. Please consider adding coverage for the following ICD-10 codes (Wen, 2015):
D46.9 Myelodysplastic Syndrome, unspecified
D46.C Myelodysplastic Syndrome with Isolated del(5q) chromsoml abnlt
D46.Z Other myelodysplastic syndromes.

Recommendation: Amend the policy to allow add coverage for Myelodysplastic Syndrome.

CPT Code 81225 (CYP2C6 19-cytochrome P450 CYP2C6 P450)

We support the proposal to cover for CYP2C619 to identify patients who are poor metabolizers of clopidogrel, particularly those with acute coronary syndrome or who are undergoing percutaneous coronary intervention.

CPT Code 81226 (CYP2D6)

We agree with your decision to cover CYP2D6 testing. As you recognize, patients requiring doses above 50 mg per day should be genotyped for the drug metabolizing enzyme CYP2D6 to determine if the patient is a poor metabolizer (PM) or a normal/extensive metabolizer (NM/EM). People with CYP2D6 poor metabolizer genotypes should be treated with lower doses.

CPT Code 81227 (G9143 CYP2C9 and/or VKORC1 Gene Testing for Warfarin Response)

We agree with NGS' proposal to cover testing for warfarin response in accordance with NCD 90.1.

CPT Code 81235 (EGFR)

We appreciate NGS' coverage of EGFR testing. First, second and third generation EGFR tyrosine kinase inhibitors with different activities against acquired resistance mutations are now available. Consequenlty, we believe this is an area where allowing repeat testing is critical to ensure that patients receive the most appropriate therapy. We also recommend the NGS provide coverage for EGFR testing in patients with brain cancer, particularly glioblastoma (Zadeh, 2013).

Recommendation: Add coverage for patients with brain cancer, particularly glioblastoma (Zadeh, 2013).

CPT Codes 81245, 81246 (FLT3)

When FLT3 and other common mutations in AML are positive at diagnosis, post-treatment testing of residual disease has been shown to be predictive of long-term outcomes and the need for more aggressive therapy (ie stem cell transplantation) (Klco, 2015).

We also believe that FLT3 ITD should be covered for patients with Chronic Myelomonocytic Leukemia, as it, along with KIT and JAK2, for which coverage is also proposed, are utilized as drug targets in this patient population (Strati, 2013; Huang, 2009).

Recommendation: The policy should be amended to cover FLT3 testing in patients with Chronic Myelomonocytic Leukemia.

CPT Codes 81272 and 81273 (KIT)

We agree with your decision to cover KIT testing in GIST, AML and melanoma to guide therapeutic decision making.

KIT mutation testing is also often critical to assist in the diagnosis of systemic mastocytosis (Valent, 2013). Please add coverage for the following related ICD-10 codes:

- Q82.2 Mastocytosis
- C96.2 Malignant mast cell tumor
- D47.0 Histiocytic and mast cell tumors of uncertain behavior

CPT Code 81275 (KRAS codon 12/13) and 81276 (KRAS codon 61/146)

KRAS testing is critical in the diagnosis and treatment of other malignancies besides lung and colon cancer, and we recommend adding the following additional diagnoses to the "covered" indications list:

1. NCCN Colon Version 2,2016 guidelines state small bowel and appendiceal carcinoma can be treated with systemic chemotherapy according to NCCN guidelines for colon cancer. Therefore, the same molecular testing required for colon cancer will be required for these cancers. For example a patient with KRAS positive appendiceal carcinoma would not be eligible for therapies targeting EGFR (e.g. cetuximab) (NCCN Guidelines: Colon, 2016).

2. KRAS mutation status is used in patients with acute myeloid leukemia (AML) to guide therapeutic decision making. In AML patients with a KRAS mutation at diagnosis the KRAS mutation levels are used to monitor response to therapy (e.g. induction chemotherapy, immuno-suppression therapy after transplant) and to identify loss of response. Oncologists use this information to manage chemotherapy, type of transplant approach (auto-transplant, allotransplant, matched unrelated donor) or modifications in the immuno-suppression or targeted therapy after transplant. NCCN 2015 guidelines state there is "an undeniable need for monitoring" minimal residual disease (MRD) (Klco, 2015; Ley, 2013; NCCN Guidelines: Acute Myeloid Leukemia, 2015).

3. KRAS mutation status can be used in patients with multiple myeloma (MM) to guide therapeutic decision in situations when the IGH gene rearrangement (CPT codes 81261-81264) is negative at diagnosis and a KRAS mutation is detected at diagnosis. Multiple myeloma patients with a KRAS mutation but not NRAS mutation respond to bortezomib therapy (Mulligan, 2014). NCCN guidelines ver2.2016 state "Patients on treatment should be monitored for response to therapy, for response to primary therapy, and for symptoms related to disease and/or treatment. It is recommended to re-evaluate (after 2 cycles) with the laboratory test, bone survey and bone marrow aspiration and biopsy to determine treatment response, or whether the primary disease is progressive (Mulligan 2014, NCCN Guidelines: Multiple Myeloma, 2016).

4. KRAS G12* or KRAS G13** positivity is used to confer a positive diagnosis (e.g. carcinoma, MDS) in equivocal surgical biopsies, or cytology specimens (fine needle aspirates) or equivocal flow cytometry specimens (blood/bone marrow).

    a. A positive diagnosis of papillary thyroid carcinoma in equivocal thyroid cytology specimens (see thyroid carcinoma comments for BRAF above)_ (NCCN Guidelines: Thyroid, 2015; Nikiforova, 2011).

    b. A positive diagnosis of Intraductal Papillary Mucinous Neoplasm (IPMN) or Mucinous cystic neoplasm in pancreatic cyst fluid with equivocal cytology results (Nikiforova, 2013; Thiruvengadam, 2015).

c. A positive diagnosis of a bilary tract neoplasm in equivocal cytology specimens (e.g. bilary tract brushing) (Borger, 2012; Kipp, 2010).

d. A positive diagnosis of myelodysplastic neoplasm in equivocal bone marrow for which a definitive diagnosis of MDS cannot be made by morphological review, flow cytometry, or cytogenetic karyotype (Cazzola, 2013; NCCN Myelodysplastic Syndrome, 2016).

5. We recommend that chronic myelomonocytic leukemia (CMML) be added as an indication to the KRAS CPT codes in Group 1 in the policy (covered procedures, without the need for individual review) because KRAS testing for CMML is required or recommended by 2016 WHO revision and is needed to determine if a Medicare covered therapy is a reasonable option given the individual's specific clinical presentation.

CPT Code 81287 (MGMT)

The neuro-oncology community has recently come to recognize the concept of pseudo-progression in the treatment course of high grade gliomas. In particular, pseudo-progression is defined as apparent post-treatment radiographically-identified disease progression followed by subsequent improvement or stabilization without any additional treatment. Pseudo-progression is a transient phenomenon that likely represents a local tissue reaction to the therapy, and its presence has actually been shown to improve overall survival (Da Cruz, 2011). Distinguishing pseudo-progression from its radiographic mimic, true tumor-specific disease progression, is thus critical, given that the best treatment option for pseudo-progression is to continue the current therapy, while the exact opposite management, discontinuation of the current therapy, is the best treatment option for true disease progression. Although current radiographic imaging methods cannot distinguish (Da Cruz, 2011) these two disparate diagnoses with radically different treatment ramifications, it has recently been determined that gliomas with MGMT promoter methylation have a significantly higher prevalence of pseudo-progression than non-methylated tumors (Brandes 2008). In this study, 91% of patients with methylated MGMT had pseudo-progression (versus 41% of patients without methylated MGMT, P = .0002), and were best managed by continuing the current therapy. The determination of MGMT promoter methylation status in post-treatment patients with imaging consistent with progression/psuedo-progression is thus clinically critical to ensure that effective therapies are not inappropriately terminated under the false assumption of disease progression (versus the alternative diagnosis of transient good-prognosis pseudo-progression).

Recommendation: MGMT testing should be covered for all glioma patients with a post-treatment imaging study suggesting progression/pseudo-progression and that any ICD-10 codes relating to this diagnosis be added to this policy. Please add coverage for the following associated codes:

C71.0 Malignant neoplasm of cerebrum, except lobes and ventricles
C71.1 Malignant neoplasm of frontal lobe
C71.2 Malignant neoplasm of temporal lobe
C71.3 Malignant neoplasm of parietal lobe
C71.4 Malignant neoplasm of occipital lobe
C71.5 Malignant neoplasm of cerebral ventricle
C71.6 Malignant neoplasm of cerebellum
C71.7 Malignant neoplasm of the brain stem
C71.8 Malignant neoplasm of overlapping sites of brain
C71.9 Malignant neoplasm of the brain, unspecified
C70.0 Malignant neoplasm of cerebral meninges
C70.0 Malignant neoplasm of meninges, unspecified
C72.0 Malignant neoplasm of spinal cord
C72.1 Malignant neoplasm of cauda equina
C70.1 Malignant neoplasm of spinal meninges

CPT Code 81310 (NPM1)

NPM1 testing has also been shown to be actionable in patients with Myelodysplastic Syndrome (Schnittinger, 2011; Bains, 2011).

Recommendation: NGS should provide NPM1 testing coverage for patients with Myelodysplastic Syndrome.

CPT Code 81311 (NRAS)

NRAS (neuroblastoma RAS viral [v-ras] oncogene homolog) (e.g., colorectal carcinoma), gene analysis, variants in exon 2 (e.g., codons 12 and 13) and exon 3 (e.g., codon 61) is considered medically necessary in patients with colorectal cancer or CMML when needed to determine if a Medicare covered therapy is a reasonable option given the individual's specific clinical presentation.

An accurate diagnosis of chronic myelomonocytic leukemia (CMML) is required to initiate therapy for this neoplasm. The 2016 WHO revision states diagnostic criteria for CMML (see WHO 2016 Guidelines; table 11) and includes "a molecular genetic abnormality is present in hemopoietic cells." "The presence of mutations in genes often associated with CMML (eg TET2, SRSF2, ASXL1 in >80% of cases and SETBP1, **NRAS/KRAS**, RUNX1, CBL and EZH2 at a lower frequency) in the proper clinical context can be used to support a diagnosis. Other molecular criteria for a CMML diagnosis include no evidence of BCR-ABL , PDGFRA, PDGFRB, FGFR1, PCM1-JAK2 translocations or JAK2, MPL1, CALR mutations (see 2016 WHO revision , Table 11 Diagnostic criteria for CMML).

An NRAS mutation often confers a positive diagnosis of papillary thyroid carcinoma in fine needle aspirates (FNA) showing equivocal thyroid cytology (eg atypical/suspicious,morphology). (NCCN Guidelines in Oncology: Thyroid, Nikiforov 2011) Mutation profiling in patients with equivocal thyroid cytology guides clinical decision making. 1) Patients who are negative for a mutation will be monitored and can be spared a thyrodectomy improving quality of life. Patients who are positive for a mutation will receive a complete thyrodectomy. Patients who are denied mutation testing are monitored, subjected to multiple FNA procedures and if multiple FNAs are equivocal will receive a partial thyrodectomy. If the thyrodectomy shows a carcinoma, the patient will undergo a second surgery to remove the remaining thyroid tissue. If the partial thyrodectomy does not show a carcinoma, the surgey might be considered an unnecessary cost and reduction in the patient's quality of life.

Recommendation: Chronic myelomonocytic leukemia (CMML) and thyroid specimens with equivocal morphology be added as an indication to the **CPT Code 81311 (NRAS)** in Group 1 in the policy (covered procedures, without the need for individual review) because NRAS testing for CMML is required or recommended by 2016 WHO revision and is needed to determine if a Medicare covered therapy is a reasonable option given the individual's specific clinical presentation.

.

CPT Code 81313 (PCA3)

dLCD Statement: When the physician plans to biopsy the prostate, NGS will consider a PCA3 test as not medically necessary, and thus, not a covered Medicare benefit. NGS considers all other indications for PCA3 not reasonable and necessary. Medical record documentation must indicate the rationale to perform a PCA3 assay.

Comment: Medical record documentation showing patients had negative biopsy and clinical suspicion of prostate cancer should be sufficient rationale for performing PCA3 test. Additional documentation should not be necessary (Roobal, 2004; Vickers, 2009).

CPT Code 81479 (ROS and MET; unlisted molecular pathology procedure)

We support coverage for testing of ROS and MET, but also believe that the procedures listed below are clinically appropriate for CPT 81479 are examples, and not an exhaustive list, of molecular procedures required or recommended by current NCCN guidelines or well-established peer-reviewed literature.

1. **81479** ERBB2 (v-erb-b2 avian erythroblastic leukemia viral oncogene homolog 2) mutation or gene amplification and 81479 RET (ret proto-oncogene) translocation or mutation (if not covered by 81404, 81405, 81406) should be considered medically necessary for non-small cell lung cancer therapeutic decision-making. NCCN Non-Small Cell Lung Cancer guidelines state “Other driver mutations and gene rearrangements (i.e driver events are being identified such as HER2 (also known as ERBB2) and BRAF V600E mutations, ROS1 and RET gene rearrangements, and high-level MET amplification or MET exon skipping mutation." NCCN guidelines also state “Afatinib is an oral TDI that inhibits the entire ErbB/HER family of receptors including EGFR and HER2.”

   “For the 2016 update, the NCCN panel added a recommendation of a dabrafenib/trametinib regimen for patients with BRAF V600E mutations." "In addition, the recommendation for cabozantinib for RET rearrangements was revised to category 2A (from category 2B).” Trastuzumab and afatinib (both for HER2 mutations) are category 2B recommendations” (NCCN Guidelines: Non-Small Cell Lung Cancer, 2016).

2. **81479** PIK3CA (phosphatidylinositol-4,5-bisphosphate 3-kinase, catalytic subunit alpha) mutations are associated with resistance to EGFR therapies (e.g. cetuximab) in colorectal carcinoma (Therkildsen, 2014).

3. **81479** HRAS (Harvey rat sarcoma viral oncogene homolog) should be considered medically necessary for the diagnosis of equivocal spitz neoplasms. HRAS mutations are associated with atypical spitz nevi but not associated with spitz melanoma. Clinical management is different for these entities. A diagnosis of melanoma requires lymph node dissection and may require chemotherapy whereas a resection of the spitz nevi is sufficient (Van Engen – Van Grunsven, 2010).

4. **81479** PTPN11 mutations aid in AML/MDS diagnosis, and MDS prognosis contributing to therapeutic decision making, PTPN11 is associated with poor survival in MDS (Bejar, 2014; NCCN Guidelines: Myelodysplastic Syndrome, 2016).

5. **81479** ASXL1 mutations aid in the diagnosis and prognosis of MDS, MPN, and MDS/MPN, and contribute to therapeutic decision making. ASXL1 is a negative prognostic marker in AML, MDS, and PMF and has been shown to negate the good response of TET2 mutations to hypomethylating therapy in MDS (Bejar, 2014), see NCCN guidelines on myleodysplastic syndromes, especially chart (Meggendorfer, 2016; NCCN Guidelines: Myelodysplastic Syndrome, 2016).

   Identification of ASXL1 mutations is necessary for therapeutic decision making in AML since they define a poor risk category defining choices for Treatment Induction and Post-remission Therapy in AML patients with a normal or abnormal karyotype (see 2016 WHO supplementary Table 2; 2016 ver2 NCCN guidelines AML-10,). ASXL1 is a predictor of aggressive disease behavior and has been incorporated into a prognostic scoring system for CMML, alongside karyotype and clinicopathologic parameters (2016 WHO revision). An accurate diagnosis of chronic myelomonocytic leukemia is required to initiate therapy for this neoplasm. The 2016 WHO revision states diagnostic criteria for CMML (see 2016 WHO revision. Table 11) includes "a molecular genetic abnormality is present in hemopoietic cells." "The presence of mutations in genes often associated with CMML (eg TET2, SRSF2, ASXL1 in >80% of cases and SETBP1, NRAS/KRAS, RUNX1, CBL and EZH2 at a lower frequency) in the proper clinical

context can be used to support a diagnosis. Other molecular criteria for a CMML diagnosis include no evidence of BCR-ABL, PDGFRA, PDGFRB, FGFR1, PCM1-JAK2 translocations or JAK2, MPL1, CALR mutations (see r2016 WHO Table 11 Diagnostic criteria for CMML). Identification of ASXL1 mutations is necessary for therapeutic decision in MDS since this mutation is independently associated with a poor prognosis and defines a high risk group within the intermediate risk IPSS-R prognostic category changing patient Treatment from low risk options (eg hypomethylating agents or lenalidomide) to high risk options (eg chemotherapy and HCT), see (see MDS-9,10,11 2016 NCCN ver1).

An accurate diagnosis of MDS is required to initiate therapy for this neoplasm. Molecular testing for ASXL1 mutations is medically necessary in appropriate clinical contexts, specifically in instances of stable cytopenia with "non-diagnostic morphology". In these situations, the presence of a ASXL1 mutation acts as co-criteria to support diagnosis of MDS and rules a benign cytopenia (see 2016 WHO revision and NCCN ver1 guidelines MDS-2).

6. **81479** CBL mutations aid in AML MDS, MDS/MPN diagnosis contributing to therapeutic decision making. CBL testing is specifically recommended to make the diagnosis of CMML (Meggendorfer, 2014) see NCCN guidelines on myleodysplastic syndromes, especially chart MDS-7 (NCCN Guidelines: Myelodysplastic Syndrome, 2016). 81479 CBL (SET-binding protein) mutation: An accurate diagnosis of chronic myelomonocytic leukemia is required to intiate therapy for this neoplasm. The 2016 WHO revision states diagnostic criteria for CMML (see 2016 WHO. Table 11) includes "a molecular genetic abnormality is present in hemopoietic cells". "The presence of mutations in genes often associated with CMML (eg TET2, SRSF2, ASXL1 in >80% of cases and SETBP1, NRAS/KRAS, RUNX1, CBL and EZH2 at a lower frequency) in the proper clinical context can be used to support a diagnosis. Other molecular criteria for a CMML diagnosis include no evidence of BCR-ABL , PDGFRA, PDGFRB, FGFR1, PCM1-JAK2 translocations or JAK2, MPL1, CALR mutations (see 2016 WHO., Table 11 Diagnostic criteria for CMML).

7. **81479** MLL mutations aid in AML/MDS diagnosis and prognosis, contributing to therapeutic decision making. MLL alterations are specifically mentioned in the WHO diagnostic “gold standard” as an adverse prognostic marker in normal karyotype AML which can be negated by allogeneic stem cell transplantation (2016 WHO Classification of Myeloid Neoplasms; Patel, 2012; Mrozek, 2012.) This has led to treatment schemes suggesting transplantation in patients with MLL-PTD (Mrozek, 2007).

8. **81479** U2AF1 mutations aid in MDS diagnosis, contributing to therapeutic decision making (found with high frequency in MDS, NOT found with high frequency in age-related clonal hematopoiesis) (for comparisons, see Yoshida, 2011; McKerell, 2015; Jasiwal, 2014; Geneovese, 2014), see NCCN guidelines on myleodysplastic syndromes, especially chart MDS-7 (NCCN Guidelines: Myelodysplastic Syndrome, 2016). An accurate diagnosis of MDS is required to initiate therapy for this neoplasm. Molecular testing for U2AF1 mutations is medically necessary in appropriate clinical contexts, specifically in instances of stable cytopenia with "non-diagnostic morphology". In these situations, the presence of a U2AF1 mutation acts as co-criteria to support a diagnosis of MDS and rules out a benign cytopenia (see 2016 WHO revision and NCCN ver1 guidelines MDS-2).

9. **81479** ZRSR2, mutations aid in MDS diagnosis contributing to therapeutic decision making (found with high frequency in MDS, NOT found with high frequency in age-related clonal hematopoiesis) (for comparisons, see Yoshida, 2011; McKerell, 2015; Jasiwal, 2014; Geneovese, 2014).

10. **81479** RUNX1 (runt related transcription factor 1) mutation testing is necessary to distinguish acute myeloid leukemia with mutated RUNX1 from other AML classifications because these patients may benefit from therapy designed for high-risk AML (2016 WHO revision, 2016 NCCN guidelines). The

2016 WHO classification states "This new provisional disease category appears to represent a biologically distinct group with a possibly worse prognosis than other AML types." The 2016 NCCN guidelines say "AML with RUNX1 is associated with a poorer prognosis". Based on current NCCN guidelines, both therapeutic decisions for "Treatment Induction" and "Post Remission Therapy" depend on risk stratification based on recurrent genetic abnormalities. Identification of RUNX1 mutations is necessary for therapeutic decision making since they define an poor risk category defining choices for Treatment Induction and Post-remission Therapy in AML patients with a normal or abnormal karyotype (see 2016 WHO supplementary Table 2, 2016 rev2 NCCN guidelines AML-10,).

An accurate diagnosis of chronic myelomonocytic leukemia (CMML) is required to initiate therapy for this neoplasm. The 2016 WHO revision states diagnostic criteria for CMML (see 2016 WHO. Table 11) includes "a molecular genetic abnormality is present in hemopoietic cells." "The presence of mutations in genes often associated with CMML (eg TET2, SRSF2, ASXL1 in >80% of cases and SETBP1, NRAS/KRAS, RUNX1, CBL and EZH2 at a lower frequency) in the proper clinical context can be used to support a diagnosis. Other molecular criteria for a CMML diagnosis include no evidence of BCR-ABL , PDGFRA, PDGFRB, FGFR1, PCM1-JAK2 translocations or JAK2, MPL1, CALR mutations (see 2016 WHO., Table 11 Diagnostic criteria for CMML).

An accurate diagnosis of MDS is required to initiate therapy for this neoplasm. Molecular testing for RUNX1mutations is medically necessary in appropriate clinical contexts, specifically in instances of stable cytopenia with non-diagnostic morphology. In these situations, the presence of a RUNX1 mutation acts as co-criteria to support a diagnosis of MDS and rules out a benign cytopenia (see 2016 WHO revision and NCCN ver1 guidelines MDS-2).

Identification of RUNX1 mutations is necessary for therapeutic decision in MDS since this mutation is independently associated with a poor prognosis and defines a high risk group within the intermediate risk IPSS-R prognostic category changing patient treatment from low risk options (eg hypomethylating agents or lenalidomide) to high risk options (eg chemotherapy and HCT), see (see MDS-9,10,11 2016 NCCN ver1).

11. **81479** TET2 (tet methylcytosine dioxygenase 2) mutations: Identification of TET2 mutations is necessary for therapeutic decision making since they define an intermediate risk category defining choices for treatment Induction and post-remission therapy in AML patients with a normal karyotype, positive for NPM1 mutation and negative for FLT3-ITD mutation (see 2016 WHO supplementary Table 2, 2016; ver2 NCCN guidelines AML-10,).

An accurate diagnosis of chronic myelomonocytic leukemia (CMML) is required to initiate therapy for this neoplasm. The 2016 WHO revision states diagnostic criteria for CMML (see 2016 WHO. Table 11) includes "a molecular genetic abnormality is present in hemopoietic cells." The presence of mutations in genes often associated with CMML (eg TET2, SRSF2, ASXL1 in >80% of cases and SETBP1, NRAS/KRAS, RUNX1, CBL and EZH2 at a lower frequency) in the proper clinical context can be used to support a diagnosis. Other molecular criteria for a CMML diagnosis include no evidence of BCR-ABL , PDGFRA, PDGFRB, FGFR1, PCM1-JAK2 translocations or JAK2, MPL1, CALR mutations (see 2016 WHO, Table 11 Diagnostic criteria for CMML).

Since oncologists have a choice between immunosuppressive therapy and hypomethylating agents for MDS patients in a low/intermediate prognostic category (see 2016 NCCN guidelines MDS-10) knowledge of the TET2 mutation status affects therapeutic decision making for MDS patients. MDS NCCN guidelines state “TET2 mutations have been shown to effect the response to hypomethylating agents. Patients with mutated TET2 had an 82% response rate to AzaC compared to 45% of patients with wildtype TET2 (P=0.007).”

An accurate diagnosis of MDS is required to initiate therapy for this neoplasm. Molecular testing for TET2 mutations is medically necessary in appropriate clinical contexts, specifically in instances of stable cytopenia with "non-diagnostic morphology". In these situations, the presence of a TET2 mutation acts as co-criteria to support a diagnosis of MDS and rules out a benign cytopenia (see 2016 WHO revision and NCCN ver1 guidelines MDS-2).

12. **81479** WT1 (Wilms tumor 1) mutations: Identification of WT1 mutations is necessary for therapeutic decision making in AML since they define a poor risk category defining choices for treatment Induction and post-remission therapy in AML patients with a normal or abnormal karyotype (see 2016 WHO supplementary Table 2; 2016 ver 2 NCCN guidelines AML-10,).

13. **81479** SF3B1 (splicing factor 3b subunit 1) mutations: An accurate diagnosis of MDS/MPN with ringed sideroblasts is required to initiate treatment for this neoplasm. 2016 WHO criteria for diagnosis of MDS/MPN with ringed sideroblasts and thrombocytosis "is strongly supported by the presence of SF3B1 mutation together with a mutation in JAK2 V617F, CALR or MPL genes." Other molecular criteria for a MDS/MPN with ringed sideroblasts diagnosis include no evidence of BCR-ABL , PDGFRA, PDGFRB, FGFR1, PCM1-JAK2, t(3;3)(q21q26), inv(3)(q21q26) translocations or del(5q) or JAK2, MPL1, CALR mutations (see WHO 2016, Table 13 Diagnostic criteria for MDS/MPN with ringed sideroblasts).

14. **81479** CSF3R (colony stimulating factor 3) mutation: An accurate diagnosis of chronic neutrophilic leukemia (CNL) is required to initiate treatment for this neoplasm. CSF3R mutations are strongly associated with CNL and represent a clonal marker for CNL diagnosis as detailed specifically in the 2016 WHO diagnostic criteria (see 2016 WHO Table 3 Diagnostic criteria for CNL). Other molecular criteria for a CNL diagnosis include no evidence of BCR-ABL , PDGFRA, PDGFRB, FGFR1, PCM1-JAK2 translocations or JAK2, MPL1, CALR mutations (see WHO 2016, Table 3 Diagnostic criteria for CNL, Maxson, 2013).

15. **81479** SRSF2 (Serine/arginine-rich splicing factor 2) mutations: An accurate diagnosis of chronic myelomonocytic leukemia is required to initiate therapy for this neoplasm. The 2016 WHO revision states diagnostic criteria for CMML (see 2016 WHO revision. Table 11) says "a molecular genetic abnormality is present in hemopoietic cells." The presence of mutations in genes often associated with CMML (eg TET2, SRSF2, ASXL1 in >80% of cases and SETBP1, NRAS/KRAS, RUNX1, CBL and EZH2 at a lower frequency) in the proper clinical context can be used to support a diagnosis. Other molecular criteria for a CMML diagnosis include no evidence of BCR-ABL , PDGFRA, PDGFRB, FGFR1, PCM1-JAK2 translocations or JAK2, MPL1, CALR mutations (see 2016 WHO revision, Table 11 Diagnostic criteria for CMML).

    An accurate diagnosis of MDS is required to initiate therapy for this neoplasm. Molecular testing for SRSF2 mutations is medically necessary in appropriate clinical contexts, specifically in instances of stable cytopenia with "non-diagnostic morphology." In these situations, the presence of a SRSF2 mutation acts as co-criteria to support a diagnosis of MDS and rules out a benign cytopenia (see 2016 WHO revision and NCCN ver1 guidelines MDS-2).

16. **81479** SETBP1 (SET-binding protein) mutation: An accurate diagnosis of chronic myelomonocytic leukemia is required to initiate therapy for this neoplasm. The 2016 WHO revision states diagnostic criteria for CMML (see 016 WHO revision Table 11) includes "a molecular genetic abnormality is present in hemopoietic cells". The presence of mutations in genes often associated with CMML (eg TET2, SRSF2, ASXL1 in >80% of cases and SETBP1, NRAS/KRAS, RUNX1, CBL and EZH2 at a

lower frequency) in the proper clinical context can be used to support a diagnosis. Other molecular criteria for a CMML diagnosis include no evidence of BCR-ABL , PDGFRA, PDGFRB, FGFR1, PCM1-JAK2 translocations or JAK2, MPL1, CALR mutations (see 016 WHO revision, Table 11 diagnostic criteria for CMML).

17. **81479** EZH2 (enhancer of zeste 2 polycomb repressive complex 2 subunit) mutation: An accurate diagnosis of chronic myelomonocytic leukemia is required to initiate therapy for this neoplasm. The 2016 WHO revision states diagnostic criteria for CMML (see r016 WHO revision Table 11) includes "a molecular genetic abnormality is present in hemopoietic cells." "The presence of mutations in genes often associated with CMML (eg TET2, SRSF2, ASXL1 in >80% of cases and SETBP1, NRAS/KRAS, RUNX1, CBL and EZH2 at a lower frequency) in the proper clinical context can be used to support a diagnosis. Other molecular criteria for a CMML diagnosis include no evidence of BCR-ABL , PDGFRA, PDGFRB, FGFR1, PCM1-JAK2 translocations or JAK2, MPL1, CALR mutations (see 2016 WHO revision, Table 11 diagnostic criteria for CMML).

An accurate diagnosis of MDS is required to initiate therapy for this neoplasm. Molecular testing for EZH2 mutations is medically necessary in appropriate clinical contexts, specifically in instances of stable cytopenia with "non-diagnostic morphology". In these situations, the presence of a EZH2 mutation acts as co-criteria to support a diagnosis of MDS and rules out a benign cytopenia (see 2016 WHO revision and NCCN ver1 guidelines MDS-2).

Identification of EZH2 mutations is necessary for therapeutic decision in MDS since this mutation is independently associated with a poor prognosis and defines a high risk group within the intermediate risk IPSS-R prognostic category changing patient treatment from low risk options (eg hypomethylating agents or lenalidomide) to high risk options (eg chemotherapy and HCT), see (see MDS-9,10,11 2016 NCCN ver1).

18. **81479** ETV6 (ETS variant 6) mutation: Identification of ETV6 mutations is necessary for therapeutic decision in MDS since this mutation is independently associated with a poor prognosis and defines a high risk group within the intermediate risk IPSS-R prognostic category changing patient treatment from low risk options (eg hypomethylating agents or lenalidomide) to high risk options (eg chemotherapy and HCT), see (see MDS-9,10,11 2016 NCCN ver1).

81445 - Targeted genomic sequence analysis panel, solid organ neoplasm DNA and RNA analysis when performed, 5-50 genes (eg ALK, BRAF, CDKN2A, EGFR, ERBB2, KIT, KRAS, NRAS, MET, PDGFRA, PDGFRB, PGR, PIK3CA, PTEN, RET) seq variants and copy# or rearrangements if performed

Additional clinical indications beyond non-small cell lung carcinoma require testing of 5 or more genes. The CPT 81445 offers an appropriate and cost effective, tissue-saving, time-saving approach to providing this service. Guidelines from professional societies (including NCCN) include additional indications which directly impact diagnosis, treatment, and management of the Medicare beneficiary. We request that CPT **81445** be added to Group 1 in the policy (covered procedures, without the need for individual review) based on Professional guidelines, Guidelines, published peer reviewed medical literature and recognized clinical utility for the following indications:

1) Therapeutic decision making for thyroid cancer patients in the appropriate clinical context, specifically molecular testing when a fine needle aspirate (FNA) shows atypia of undetermined significance/follicular lesion of undetermined significance (AUS/FLUS).
A diagnosis of thyroid cancer is required to initiate therapy for this neoplasm. Professional guidelines (2016

ver1 NCCN Thyroid Carcinoma guidelines, and 2015 American Thyroid Association Management Guidelines) call for an FNA in patients with thyroid nodules (see THYR-1). In some instances morphological review (pathologist review of cells on slide) yields equivocal findings (AUS/FLUS). When there is not a high clinical and/or radiographic suspicion of malignancy both guidelines recommend molecular diagnostics to direct therapeutic decisions (see NCCN THYR-3). Negative molecular results call for observation or lobectomy whereas positive molecular results lead to a total thyroidectomy (see NCCN THYR-4, PAP-1, FOLL-1).

Specifically, molecular diagnostic testing to detect individual mutations in 6 or more genes (eg BRAF V600E, RET/PTC, RAS [KRAS, NRAS, HRAS], PAX8/PPARg [peroxisome proliferator-activated receptors gamma]) can be used to evaluate FNA samples that are indeterminate to assist in management decisions (Table X, 2016 ver1 NCCN Thyroid Carcinoma guidelines, and 2015 American Thyroid Association Management Guidelines, Nikiforova 2013).

Patients who are denied mutation testing are monitored, subjected to multiple FNA procedures and if multiple FNAs are equivocal will receive a partial thyrodectomy (lobectomy) (see 2016 NCCN guidelines THYR-4),. If the lobectomy shows a carcinoma, the patient will undergo a second surgery to remove the remaining thyroid tissue at added expense (see 2016 NCCN guidelines PAP-2, FOLL-1, HURT-1), If the partial thyrodectomy does not show a carcinoma, the surgery might be considered an unnecessary cost and reduction in the patient's quality of life. Total thyroidectomy avoids a second surgery but a lobectomy avoids hormone replacement therapy.

Relative frequency of gene mutations in Thyroid carcinoma.

Gene Mutation	PTC	FTC	HCTC	MTC	ATC	F/Ad	H/Ad
AKT1 (%)	<1	<1	5	<1	5	5	<1
APC (%)	12	<1	-	<1	16	0	-
BRAF (%)	40-52	0-1	0-1	0-7	26	0-1	<1
CDH1 (%)	6	-	-	<1	-	-	-
CDKN2A(%)	7	14	-	<1	9	<1	-
CTNNB1(%)	3	-	-	<1	29-60	-	-
GNAS (%)	4	3	-	<1	-	6	-
HRAS (%)	2-6	6-14	7	9-18	5	5-12	0-2
IDH1 (%)	10-20	5-25	14	<1	8-30	<1	<1
KRAS (%)	1-4	4-7	2	3-7	8	1-6	0 0
NRAS (%)	4-10	16-29	5	0-1	15	6-22	2-11
PIK3CA(%)	1-2	5-15	10	<1	12-25	0-5	<1
PTEN (%)	1-2	7-15	<1	<1	10-20	0	0
RET (%)	<1*	<1	<1	40-50	<1	-	<1
SMAD4 (%)	14	-	<50	<1	-	0-45	0-40
TP53 (%)	4	10	-	<1	58-80	6	-

PTC, Papillary Carcinoma FTC, Follicular Carcinoma
HCTC, Hurthle Cell Carcinoma MTC, Medullary Carcinoma
ATC, Anaplastic Carcinoma
F/Ad, Follicular Adenoma H/Ad, Hurthle Cell Adenoma

2) Therapeutic decision making for patients with pancreatic cysts in the appropriate clinical context, specifically molecular testing when imaging,studies and endoscopic ultrasound (EUS) are equivocal and fine needle aspirate (EUS-FNA) produces negative or non-diagnostic morphological findings.

Since pancreatic cysts can be benign or malignant, proper management requires accurate classification. Cystic lesions of the pancreas include intraductal papillary mucinous neoplasms (IPMNs) and mucinous cystic neoplasms (MCN) with a risk malignancy at around 62% and <15% respectively (2016 ver 1 NCCN Guidelines: Pancreatic Adenocarcinoma). A diagnosis of malignancy triggers surgery (eg whipple), which carries significant morbidity and mortality. Current management relies on endoscopic ultrasound to make this diagnosis since it has "the ability to aspirate the cyst contents for cytologic, biochemical and molecular analysis. " (2016 ver 1 NCCN guideline, Al-Haddad M 2015) In some instances imaging studies and endoscopic ultrasound produce ambiguous findings and cytologic review (pathologist review of cells on slide) yields negative or equivocal findings. In these instances molecular testing can direct therapeutic decisions. A positive molecular result will trigger periodic imaging surveillance. Surgery is triggered when the molecular profile supports a high risk cytologic or imaging classification. Negative molecular results in combination with other clinical findings can rule out a neoplastic process. Mutation analysis of 6 or more genes (eg BRAF, CTNNB1, CDKN2A, KRAS, GNAS, PIK3CA, RNF43, VHL, SMAD4, TP53) has been shown to identify nonmucinous and mucinous cysts that would by imaging, cytologic, and CEA criteria be classified as benign (Table Y, Schonleblen 2008, Wu 2011, Nikiforov 2011, Singhi 2014, Thiruvengadam 2015, Jones M 2016) . This is important because mucinous cysts have malignant potential and thus require rigorous follow-up (Tanaka M, 2012). Further, SMAD4, TP53 mutations are associated with progression and support a diagnosIs of high grade dysplasia and invasive carcinoma (Hruban 2004, Jones 2008, Murphy 2013, Amato 2014, Lennon 2014, Jones 2016).

Table Y: Relative frequency of gene mutations in Pancreatic Neoplasms.

Gene Mutation	PDAC	IPMN	ITPN	MCN	SCA	SPN	CRC	Apdx Ca
APC (%)	1-6	2-10	-	<1	<1	<1	43-79	-
ATM (%)	4-5	<1	-	<1	<1	<1	33	-
BRAF (%)	1-5	**1-5**	<1	1-2	<1	<1	14-22	-
CDKN2A(%)	2-23	**2-40**	-	-	<1	<1	2	-
CTNNB1(%)	0-2	<1	<1	<1	<1	**95**	20	<1
ERBB2 (%)	1	<2	-	<1	<1	<1	7	-
ERBB4 (%)	2	<1	-	<1	<1	<1	14	-
FBXW7 (%)	2-3	<1	-	<1	<1	<1	21-29	-
FGFR2 (%)	<1	2-5	-	<1	<1	<1	10	-
GNAS (%)	1	**40-75**	-	<1	<1	<1	4	46
KRAS (%)	70-96	**63-81**	<1	**40-80**	<1	<1	36-46	68
PIK3CA(%)	1-3	1-2	**10-20**	<1	<1	<1	23-43	-
SMAD4 (%)	18-22	1-2*****	-	1-2*	<1	<1	25-31	14
STK11 (%)	0-2	5-25	-	<1	<1	<1	7	-
TP53 (%)	39-52	0-5*****	-	0-5*	<1	<1	49-66	50-99
VHL (%)	<1	<1	-	<1	**70-80**	<1	17	-

* Mutations are frequently seen in carcinoma arising from IPMN or MCN (SMAD4 = 25%, TP53 = 50%).

PDAC - Pancreatic Adenocarcinoma
IPMN - Intraductal Papillary Mucinous Neoplasm
ITPN - Intraductal Tubulopapillary Neoplasm
MCN - Mucinous Cystic Neoplasm
SCA - Serous Cystadenoma
SPN - Solid-Pseudopapillary Neoplasm
CRC - Colorectal Adenocarcinoma
ApdxCa - Appendiceal Carcinoma

III. TIER 1 CODES NOT LISTED AS COVERED MOLECULAR PATHOLOGY PROCEDURES

Recommendation: We recommend the following CPT codes be added to Group 1 in the policy (covered procedures, without the need for individual review) because it is required or recommended by current WHO classifications and NCCN guidelines, as indicated

CPT Code 81315 - PML/RARALPHA, (T(15;17)), (PROMYELOCYTIC LEUKEMIA/RETINOIC ACID RECEPTOR ALPHA) (EG,PROMYELOCYTIC LEUKEMIA) TRANSLOCATION ANALYSIS; COMMON BREAKPOINTS (EG, INTRON 3 AND
INTRON 6), QUALITATIVE OR QUANTITATIVE
CPT Code 81316 - PML/RARALPHA, (T(15;17)), (PROMYELOCYTIC LEUKEMIA/RETINOIC ACID RECEPTOR ALPHA) (EG, PROMYELOCYTIC LEUKEMIA) TRANSLOCATION ANALYSIS; SINGLE BREAKPOINT (EG, INTRON 3, INTRON
6 OR EXON 6), QUALITATIVE OR QUANTITATIVE

Molecular testing is necessary to distinguish Acute Promeylocytic leukemia (APL) from other AML classifications because these patients benefit from a different induction therapy (ie ATRA) and consolidation therapy (ie Arsenic trioxide) than other AML classifications. 2016 WHO classifies AML with PML-RARA as a separate classification. The 2016 ver2 NCCN guidelines state APL classification should be based on "APL morphology and (+) for t(15;17) by cytogenetics or PML/RARA by molecular testing (see AML-2 algorithm). Further 2016 NCCN ver2 guidelines require the documentation of "molecular remission after consolidation. Monitor by PCR for up to 2 year."

PML/RARA molecular testing for residual disease is also necessary for therapeutic decision making in APL patients. Current NCCN guidelines state "PCR should be performed on a marrow sample at completion of consolidation to document molecular remission." "In patients receiving the ATRA/arsenic regimen, consider earlier sampling at 3-4 months after consolidation. Subsequent monitoring by PCR can be done with peripheral blood, although marrow is a more sensitive monitoring technique and may give earlier signs of relapse." 2016 ver2 NCCN guidelines provide therapeutic options for patients who show molecular evidence of relapse (see AML-5, AML-6)

IV. New CPT 2016 CPT codes

We appreciate NGS efforts to add new CPT codes to Group 1 in the policy (eg 81170, 81219, 81276).

We also request the following new CPT codes that were available as of January 1, 2016, be added to Group 1 in the policy (covered procedures, without the need for individual review), based on abundant evidence of clinical utility, as a predictor of PARP inhibitor sensitivity, as detailed above.

1. **81162** BRCA1, BRCA2 full sequence analysis and full duplication/deletion analysis (Mateo, 2015; Kaufman, 2015).

V. Tier 2 Codes not listed as covered molecular pathology procedures

We request that NGS add the following CPT codes to Group 1 in the policy (covered procedures, without the need for individual review) base on abundant evidence of clinical utility. These CPT codes are examples of molecular procedures required or recommended by current NCCN guidelines or well- established peer-reviewed

literature, and do not represent an exhaustive list of CPT codes that should be covered.

1. **81403** CTNNB1 (catenin (cadherin-associated protein),beta 1, 88kDa) is considered medically necessary for the diagnosis desmoid fibromatosis and to guide therapeutic decision making (Lazar, 2008; NCCN Guidelines: Soft Tissue Sarcoma, 2015; Tejpar, 1999). In the previous NGS response to DL35000 comments (see Response to Comments: Molecular pathology Procedures A54825) NGS indicated it "will add only CTTNNB1 to Group 2 – Molecular Pathology procedures that require review – only for individuals who have desmoids tumors to guide therapeutic decision making." We request that a statement confirming the CTNNB1 indication is covered be added to the Group 2 (subject to review) paragraph to clarify this question.

2. **81403** IDH1 (isocitrate dehydrogenase 2) and **81403** IDH2 (isocitrate dehydrogenase 2) The current LCD states "Removed IDH1 (isocitrate dehydrogenase) and IDH2 (reported with code 81403) from the TIER 2 NON-COVERED MOLECULAR PATHOLOGY PROCEDURES (now subject to individual review)." We appreciate NGS' decision to make this change. We would request that a statement confirming the IDH1/2 indication be added to the Group 2 (subject to review) paragraph to better clarify when this testing is medically

   Identification of IDH1 mutations is necessary for therapeutic decision making since they define an intermediate risk category defining choices for Treatment Induction and Post-remission Therapy in AML patients with a normal karyotype, positive for NPM1 mutation and negative for FLT3-ITD mutation (see WHO supplementary Table 2, NCCN guidelines AML-10).

   Both 81403 IDH1 and IDH2 should be a covered molecular pathology procedure to aid the diagnosis myelodysplastic syndromes in patients with clinical diagnostic criteria but lacking diagnostic morphology (eg. equivocal morphology) needed to establish a diagnosis of MDS. See rationale listed for Group 5 Paragraph Non-covered genomic sequencing procedure 81450.31 These mutations also lead to the production of a novel onco-metabolite 2-hydroxy-glutarate that is the target of new anti-cancer drugs that are showing substantial activity in clinical trials (many results listed at clinicaltrials.gov).*(*2016 NCCN Guidelines in Oncology: Myelodysplastic Syndromes)

3. **81403** DNMT3A, tsa AML/ MDS diagnosis/ prognosis. DNMT3A mutations are very common in myeloid malignancies, assist in the diagnosis of clonal disease (MPN/ MDS) from benign reactive conditions, and inform the appropriate dose of chemotherapy for optimal outcomes in AML (Sehgal, 2015; NCCN Guidelines: Myelodyspastic Syndrome, 2016).

   Identification of **DNMT3A** mutations is necessary for therapeutic decision making in **AML** since they define an intermediate risk category defining choices for Treatment Induction and Post-remission Therapy in AML patients with a normal karyotype, positive for NPM1 mutation and negative for FLT3-ITD mutation (see 2016 WHO supplementary Table 2, 2016 ver2 NCCN guidelines AML-10).

   An accurate diagnosis of **MDS** is required to initiate therapy for this neoplasm. Molecular testing for **DNMT3A** mutations is medically necessary in appropriate clinical contexts, specifically in instances of stable cytopenia with "non-diagnostic morphology". In these situations, the presence of a DNMT3A mutation acts as co-criteria to support a diagnosis of MDS and rules out a benign cytopenia (see 2016 WHO revision and NCCN ver1 guidelines MDS-2).

VI. Tier 2 Non-Covered Molecular Pathology Procedures:

We request that NGS add the following Tier 2 CPT codes to Group 1 in the policy (covered procedures, without

the need for individual review) based on abundant evidence of clinical utility. These CPT codes are examples of molecular procedures required or recommended by current NCCN guidelines or well- established peer-reviewed literature, and do not represent an exhaustive list of CPT codes that should be covered.

1. **81401** DEK-NUP214 Please restore DEK-NUP214 to TIER 2 COVERED MOLECULAR PATHOLOGY PROCEDURES. Identification of DEK-NUP214 translocations are necessary for therapeutic decision making since they define a poor risk category defining choices for Treatment Induction and Post-remission Therapy in AML patients (see 2016 WHO supplementary Table 2, 2016 NCCN guidelines AML-10). While this translocation is more common in children, this recurrent genetic abnormality is observed in 1% of adult AML (see Table B below).

2. **81401** MLL/MLLT3 or MLLT3-KMT2A Identification of MLL/MLLT3 or MLLT3-KMT2A translocations are necessary for therapeutic decision making since they define an intermediate risk category defining choices for Treatment Induction and Post-remission Therapy in AML patients (see 2016 WHO supplementary Table 2, 2016 NCCN guidelines AML-10,).

3. **81404** TP53 mutations should be a covered molecular pathology procedure for the following reasons:

   Identification of TP53 mutations is necessary for therapeutic decision making in AML since they define an poor risk category defining choices for Treatment Induction and Post-remission Therapy in AML patients with a normal or abnormal karyotype (see 2016 WHO supplementary Table 2, 2016 ver2 NCCN guidelines AML-10).

   An accurate diagnosis of MDS is required to initiate therapy for this neoplasm. Molecular testing for TP53 mutations is medically necessary in appropriate clinical contexts, specifically in instances of stable cytopenia with "non-diagnostic morphology". In these situations, the presence of a TP53 mutation acts as co-criteria to support a diagnosis of MDS and rules out a benign cytopenia (see 2016 WHO revision and 2016 NCCN ver1 guidelines MDS-2).

   Identification of TP53 mutations is necessary for therapeutic decision in MDS since this mutation is independently associated with a poor prognosis and defines a high risk group within the intermediate risk IPSS-R prognostic category changing patient Treatment from low risk options (eg hypomethylating agents or lenalidomide) to high risk options (eg chemotherapy and HCT), see MDS-9,10,11 2016 NCCN ver1. Further, WHO 2016 revision states TP53 mutation "appears to predict poorer response to lenalidomide in [MDS] patient with del(5q)." Evaluation for TP53 mutation is recommended in patients with MDS with isolated del(5q) to help identify an adverse prognostic subgroup in this generally favorable prognosis MDS entity." (WHO 2016). MDS patients with del(5q) and TP53 mutation will receive a different treatment (see MDS-9,10,11 2016 NCCN ver1).

   TP53 mutation status identifies chronic lymphocytic leukemia (CLL) patients who will benefit from Ibrutinib since this therapy is active in TP53-aberrant chronic lymphocytic leukemia. “Patients with CLL who harbor mutation and/or deletion of TP53 tumor suppressor gene respond poorly to chemoimmunotherapy and frequently succumb to relapse”. Ibrutinib is FDA-approved for patients with relapsed or refractory CLL, including patients with deletion of 17p13.1, which contains TP53. In a phase 2 study Farooqui et al provide evidence supporting ibrutinib therapy for patients with a TP53 mutation in the absence of 17p13.1 deletion. Further, TP53 mutations are associated with poor prognosis in CLL and oncologists use this information in therapeutic decision making. (Kandoth, 2013; Farooqui, 2015; Rossi, 2014; T, 2010).

VII. Group 5 Paragraph: Non-Covered Genomic Sequencing Procedures

81450 - TARGETED GENOMIC SEQUENCE ANALYSIS PANEL, HEMATOLYMPHOID NEOPLASM OR DISORDER, DNA AND RNA ANALYSIS WHEN PERFORMED, 5-50 GENES (EG, BRAF, CEBPA, DNMT3A, EZH2, FLT3, IDH1,IDH2, JAK2, KRAS, KIT, MLL, NRAS, NPM1, NOTCH1), INTERROGATION FOR SEQUENCE VARIANTS, AND COPY NUMBER VARIANTS OR REARRANGEMENTS, OR ISOFORM EXPRESSION OR MRNA EXPRESSION

We request that CPT **81450** be added to Group 1 in the policy (covered procedures, without the need for individual review) based on 2016 NCCN guidelines the 2016 WHO revision and recognized clinical utility for the following indications:

1. Molecular testing to aid Diagnosis for Therapeutic Decision making in Acute Leukemia (see AML background section and 2016 WHO revision and 2016 ver 2 NCCN AML guidelines). The correct therapy for acute leukemia depends on correct classification of the leukemia. Morphology alone (pathologist review of tissue on slide) is not sufficient to provide determine the classification. 2016 WHO guidelines define 11 acute myeloid leukemia classifications defined by a recurrent genetic abnormality. Since testing can involve greater than 5 genes including RUNX1/RUNX1T1, CBFB-MYH11, PML-RARA, MLL/MLLT3, DEK-NUP214, BCR-ABL1, NPM1, CEBPA, RUNX1 (not an an exhaustive or comprehensive list) the CPT 81450 offers an appropriate and cost effective approach to providing this service when testing is available.

2. Molecular testing to aid Risk Classification for Therapeutic Decision making in Acute Myeloid Leukemia (see AML background section and 2016 WHO revision and 2016 ver 2 NCCN AML guidelines). Testing for recurrent genetic abnormalities is necessary for therapeutic decision making in AML since these mutations/translocations define a patient risk categories which define choices for Treatment Induction and Post-remission Therapy in AML patients (see Table B, 2016 WHO supplementary Table 2, 2016 ver2 NCCN guidelines AML-10,). Since testing involves greater than 5 genes including KIT, FLT-ITD, NPM1, CEBPA, TP53, ASXL1, RUNX1, WT1 (not an an exhaustive or comprehensive list) the CPT 81450 offers an appropriate and cost effective approach to providing this service when testing is available

3. Molecular testing to aid Diagnosis for Therapeutic Decision making in chronic myelomonocytic leukemia (CMML) (see MDS/MPN background section and 2016 WHO revision and 2016 ver 1 NCCN MDS/MPN guidelines) A diagnosis of (CMML) is required to intiate therapy for this neoplasm. The 2016 WHO revision states diagnostic criteria for CMML (see 2016 WHO revision Table 11) includes "a molecular genetic abnormality is present in hemopoietic cells." The presence of mutations in genes often associated with CMML (eg TET2, SRSF2, ASXL1 in >80% of cases and SETBP1, NRAS/KRAS, **RUNX1**, CBL and EZH2 at a lower frequency) in the proper clinical context can be used to support a diagnosis. Other molecular criteria for a CMML diagnosis include no evidence of BCR-ABL , PDGFRA, PDGFRB, FGFR1, PCM1-JAK2 translocations or JAK2, MPL1, CALR mutations (see 2016 WHO revision. Table 11 Diagnostic criteria for CMML). Since testing involves greater than 5 genes including KIT, FLT-ITD, NPM1, CEBPA, TP53, ASXL1, RUNX1, WT1 (not an an exhaustive or comprehensive list) the CPT 81450 offers an appropriate and cost effective approach to providing this service when testing is available

4. Molecular testing to aid Diagnosis for Therapeutic Decision making in Myeloproliferative Neoplasms/Myelodysplastic Syndromes (see MDS/MPN background section and 2016 WHO revision and 2016 ver 1 NCCN MDS/MPN guidelines) A diagnosis of MDS is required to initiate therapy for this neoplasm. Molecular testing for recurrent MDS mutations is medically necessary in appropriate clinical contexts, specifically in instances of stable cytopenia with "non-diagnostic morphology". In these situations, the presence of a recurrent MDS mutations acts as co-criteria to support a diagnosis of MDS and rules out benign cytopenia (see Table MDS-7, 2016 WHO revision and NCCN ver1 guidelines MDS-2). Since testing can involve greater than 5 genes including SF3B1, TET2, SRSF2, ASXL1, DNMT3A, RUNX1, U2AF1, TP53 and EZH2 (not an an exhaustive or comprehensive list) the CPT 81450 offers an appropriate and cost effective approach to providing this service when testing is available. Because no one single gene is present in more than 30% of patients with myelodysplastic syndrome, multiple genes must be tested to have a reasonable certainty a neoplastic process can be

ruled in or out. The CPT code 81450 was designed exactly for this purpose and is the appropriate assay in this context. Furthermore, current NCCN guidelines state “mutations in some non-MDS genes may indicate the presence of neoplasms that can mimic MDS. These mimic conditions” can include CALR mutations associated with primary myelofibrosis, CSF3R mutations associated with atypical CML and chronic neutrophilic leukemia, and STAT3 mutations associated with LGL leukemia."

5. Molecular testing to aid Risk Classification for Therapeutic Decision making in MDS/MPN (see MDS/MPN background section and 2016 WHO revision and 2016 ver 1 NCCN MDS guidelines). The correct therapy for MPN/MDS depends on correct risk classification of the neoplasm. NCCN guidelines recommend different "Treatment" based on prognostic categories that range from very low to intermediate to very high risk. For patients who fall into the intermediate risk category molecular testing for gene mutations (TP53, ASXL1, ETV6, RUNX1, and EZH2) is necessary for therapeutic decision in MDS since these mutations are independently associated with a poor prognosis and defines a high risk group within the intermediate risk IPSS-R prognostic category. The presence of these mutations changes patient Treatment from low risk options (eg hypomethylating agents or lenalidomide) to high risk options (eg chemotherapy and HCT), see (see MDS-9,10,11 2016 NCCN ver1). Since testing involves 5 genes including TP53, ASXL1, ETV6, RUNX1, and EZH2 the CPT 81450 offers an appropriate and cost effective approach to providing this service when testing is available.

Table MDS-7 (adapted from NCCN guidelines)

Mutated Gene	Overall Incidence	Clinical Significance
TET2	20-25%	Associated with normal karyotypes. More frequency in CMML (40-60%)
DNMT3A	12-18%	Associated with poor prognosis
TP53	8-12%	Independently associated with poor prognosis. More frequent with complex karyotypes (50)% and del(5q) (15-20%)
SF3B1	18-30%	Strongly associated with ring sideroblast, more frequent in RARAS (90%) Associated with more favorable prognosis
SRSF2	10-15%	More frequent in CMML (40-50%),associated with poor prognosis.
U2AF1	8-12%	Associated with a poor prognosis
ZRSR2	5-10%	Associated with a poor prognosis
ASXL1	15-25%	Independently associated with a poor prognosis in MDS, CMML. More frequent in CMML (40-50%)
RUNX1	10-15%	Independently associated with a poor prognosis in MDS.
EZH2	5-10%	Independently associated with a poor prognosis in MDS,MDS/MPN
NRAS	5-10%	Associated with a poor prognosis, particularly in patients predicted to have lower-risk MDS More frequent in CMML, JMML (~15%)
CBL	<5%	More frequent in CMML (10-20%) and JMML (15%).
JAK2	<5%	More frequent in RARS-T (50%).
SETBP1	<5%	Associated with disease progression. More frequent in CMML (5-10%) and JMML (7%)
IDH1	<5%	More frequent in AML
IDH2	<5%	More frequent in AML
ETV6	<5%	Independently associated with a poor prognosis.

5) Additionally 2015 NCCN guidelines recommend obtaining "marrow to document remission status upon hematologic recovery including cytogenetics and molecular studies as appropriate." Consequently, mutation status must be determined to identify molecular markers to monitor therapeutic response. NCCN guidelines also instruct oncologists to "consider clinical trials for patients with targeted molecular abnormalities." It is only possible follow these NCCN guidelines if gene panels of 5-50 genes are not denied automatically as not medically necessary (Kandoth, 2013; Ley, 2013; NCCN Guidelines: Acute Myeloid Leukemia, 2015).

We respectfully ask that you consider these comments which were prepared by members of AMP and CAP who provide services to Medicare beneficiaries covered by NGS. We are happy to be of assistance in providing additional clinical information, references, contacts, or whatever is needed to assist you with this draft LCD. Please direct your correspondence to Tara Burke, AMP Senior Policy Analyst, at tburke@amp.org or Nonda Wilson, CAP's Manager, Economic and Regulatory Affairs, at nwilson@cap.org.

Sincerely,

Association for Molecular Pathology
College of American Pathologists

References

Al-Haddad M. Role of emerging molecular markers in pancreatic cyst fluid. Endoscopic Ultrasound. 2015;4(4):276-283.

Amato E, Molin MD, Mafficini A, et al. Targeted next-generation sequencing of cancer genes dissects the molecular profiles of intraductal papillary neoplasms of the pancreas. J Pathol 2014;233:217-27.

Andrulis M et al. Targeting the BRAF V600E mutation in multiple myeloma. Cancer Discov. 2013 Aug;3(8):862-9. doi: 10.1158/2159-8290.CD-13-0014. Epub 2013 Apr 23.

Arber DA et al. The Updated WHO Classification of Hematological Malignancies. The 2016 revision of the World Health Organization classification of myeloid neoplasms and acute leukemia. Blood 2016;127(20);2391-2405

Bains A, Luthra R, Medeiros L, Zuo Z. FLT3 and NPM1 Mutations in Myelodysplastic Syndromes: Frequency and Potential Value for Predicting Progression to Acute Myeloid Leukemia. Am J Clin Pathol. 2011; 135: 62-69.

Bejar R, Lord A, Stevenson K, et al. TET2 mutations predict response to hypomethylating agents in myelodysplastic syndrome patients. Blood. 2014;124(17):2705-2712. doi:10.1182/blood-2014-06-582809.

Berres M-L et al. BRAF-V600E expression in precursor versus differentiated dendritic cells defines clinically distinct LCH risk groups. J. Exp. Med. 2014 Vol. 211 No. 4 669-683 www.jem.org/cgi/doi/10.1084/jem.20130977

Birtle AJ. Clinical features of patients who present with metastatic prostate carcinoma and serum prostate-specific antigen (PSA) levels < 10 ng/mL: the "PSA negative" patients. Cancer. 2003 Dec 1;98(11):2362-7.

Boissel et al. Incidence and prognostic impact of c-Kit, FLT3, and Ras gene mutations in core binding factor acute myeloid leukemia (CBF-AML) Leukemia (2006) 20, p. 965-970

Borger D et al. Frequent Mutation of Isocitrate Dehydrogenase (IDH)1 and IDH2 in Cholangiocarcinoma Identified Through Broad-Based Tumor Genotyping. The Oncologist 2012; 17:72-79.

Brandes, A, et al. MGMT Promoter Methylation Status Can Predict the Incidence and Outcome of Pseudoprogression After Concomitant Radiochemotherapy in Newly Diagnosed Glioblastoma Patients. J Clin Oncol 26:2192-2197, 2008.

Cargo CA, Rowbotham N, Evans PA, et al. Targeted sequencing identifies patients with preclinical MDS at high risk of disease progression. Blood 2015. 126(21):2362-2365.

Cazzola M1, Della Porta MG, Malcovati L. The genetic basis of myelodysplasia and its clinical relevance. Blood. 2013 Dec 12;122(25):4021-34. doi:10.1182/blood-2013-09-381665. Epub 2013 Oct 17.

Chen W, Huang Q Detection of FLT3/ITD, JAK2(V617F) and NPM1 Gene Mutations in Chronic Myelomonocytic Leukemia. Leukemia Research 2009:E207-E209.

Da Cruz, H, et al. Pseudoprogression and pseudoresponse: imaging challenges in the assessment of posttreatment glioma. Am J Neuroradiology 2011.

Dave N, Strati P, Jabbour E et al. FLT3 Mutations in Myelodysplastic Syndrome and Chronic Myelomonocytic Leukemia. American Journal of Hematology 2013: 56-59.

Diamond EL et al. Consensus guidelines for the diagnosis and clinical management of Erdheim-Chester disease Blood, 2014;124(4):483-492.

Farooqui et al. Ibrutinib for previously untreated and relapsed or refractory chronic lymphocytic leukaemia with TP53 aberrations: a phase 2, single-arm trial. Lancet. 2015; 16(2):169-176.

Gastrointest Endosc. 2016; 83(1):140-8.

Genovese G et al. Clonal hematopoiesis and blood-cancer risk inferred from blood DNA sequence. N Engl J Med 2014;371(26): 2477-2487.

Genovese, G., Kähler, A. K., Handsaker, R. E., Lindberg, J., Rose, S. A., Bakhoum, S. F., et al. (2014). Clonal Hematopoiesis and Blood-Cancer Risk Inferred from Blood DNA Sequence. New England Journal of Medicine N Engl J Med, 371(26), 2477-2487. doi:10.

Guo J et al. Phase II, Open-Label, Single-Arm Trial of Imatinib Mesylate in Patients With Metastatic Melanoma Harboring c-Kit Mutation or Amplification J Clin Oncol 2011; 29:2904-2909.

Haroche J, et al. Dramatic efficacy of vemurafenib in both multisystemic and refractory Erdheim-Chester disease and Langerhans cell histiocytosis harboring the BRAF V600E mutation. Blood. 2013;121(9):1495- 1500

Haugen et al, 2015 American Thyroid Association Management Guidelines for Adult Patients with Thyroid Nodules and Differentiated Thyroid Cancer The American Thyroid Association Guidelines Task Force on Thyroid Nodules and Differentiated Thyroid Cancer . T

Hodi FS et al. Imatinib for melanomas harboring mutationally activated or amplified KIT arising on mucosal, acral, and chronically sun-damaged skin. J Clin Oncol. 2013 Sep 10;31(26):3182-90. doi: 10.1200/JCO.2012.47.7836.

Hruban RH, Takaori K, Klimstra DS, et al. An illustrated consensus on the classification of pancreatic intraepithelial neoplasia and intraductal papillary mucinous neoplasms. Am J Surg Pathol 2004;28: 977-87.

Ito T et al. Gastrointestinal stromal tumors with exon 8 c-kit gene mutation might occur at extragastric sites and have metastasis-prone nature. Int J Clin Exp Pathol. 2014;7(11): 8024-8031. PMCID: PMC4270532.

Jaiswal S, et al. Age-related clonal hematopoiesis associated with adverse outcomes. N Engl J Med 2014;371(26):2488-2498.

Jaiswal, S., Fontanillas, P., Flannick, J., Manning, A., Grauman, P. V., Mar, B. G.., et. Al.(2014). Age-Related Clonal Hematopoiesis Associated with Adverse Outcomes. New England Journal of Medicine N Engl J Med, 371(26), 2488-2498. doi:10.1056/nejmoa140

Jones M et al. Impact of next-generation sequencing on the clinical diagnosis of pancreatic cysts.

Jones S, Zhang X, Parsons DW, et al. Core signaling pathways in human pancreatic cancers revealed by global genomic analyses. Science 2008;321:1801-6.

Kandoth C et al. Mutational landscape and significance across 12 major cancer types. Nature, 2013; 502:333-339.

Kaufman, B., Shapira-Frommer, R., Schmutzler, R. K., Audeh, M. W., Friedlander, M., Balmana, J., . . . Domchek, S. M. (2014). Olaparib Monotherapy in Patients With Advanced Cancer and a Germline BRCA1/2 Mutation. Journal of Clinical Oncology, 33(3), 244-2

Kindler et al. Efficacy and safety of imatinib in adult patients with c-kit positive acute myeloid leukemia Blood, 2004;103(10) 3644-3653.

Kipp BR et al. Comparison of KRAS Mutation Analysis and FISH for Detecting Pancreatobiliary Tract Cancer in Cytology Specimens Collected During Endoscopic Retrograde Cholangiopancreatography. J Mol Diagn. 2010 Nov; 12(6):780-786. doi: 10.2353/jmoldx.2010.

Klco JM et al. Association between Mutation Clearance after Induction Therapy and Outcomes in Acute Myeloid Leukemia. JAMA. 2015 Aug 25;314(8):811-22. doi: 10.1001/jama.2015.9643

Lazar AJ et al. Specific mutations in the beta-catenin gene (CTNNB1) correlate with local recurrence in sporadic desmoid tumors. Am J Pathol. 2008 Nov; 173(5): 1518-27.

Lennon AM, Wolfgang CL, Canto MI, et al. The early detection of pancreatic cancer: What will it take to diagnose and treat curable pancreatic neoplasia? Cancer Res 2014;74:3381-9.

Ley TJ et al. Cancer Genome Network. Genomic and Epigenomic Landscapes of Adult De Novo Acute Myeloid Leukemia. N Engl J Med 2013;368:2059-74.

Lindsley RC et al. Acute myeloid leukemia ontogeny is defined by distinct somatic mutations. Blood. 2015 Feb 26;125(9):1367-76. doi: 10.1182/blood-2014-11-610543. Epub 2014 Dec 30.

Mateo, J., Sandhu, S., Miranda, S., Carreira, S., Jain, S., Ralph, C., . . . Bono, J. S. (2015). Abstract CT322: DNA repair defects and antitumor activity with PARP inhibition: TOPARP, a phase II trial of olaparib in metastatic castration resistant prosta

Maxson JE et al. Oncogenic CSF3R mutations in chronic neutrophillic leukemia and atypical CML. N.Engl K Med. 2013:368(19):1781-1790.

Mckerrell, T., Park, N., Moreno, T., Grove, C., Ponstingl, H., Stephens, J., . . . Vassiliou, G. (2015). Leukemia-Associated Somatic Mutations Drive Distinct Patterns of Age-Related Clonal Hemopoiesis. Cell Reports, 10(8), 1239-1245. doi:10.1016/j.celrep.

Meggendorfer, Haematologica 2014; 99:e246 and references therein; also Patel, NEJM 2012;366:1079- 1089

Mrozek, K. et al. Prognostic Significance of the European LeukemiaNet Standardized System for Reporting Cytogenetic and Molecular Alterations in Adults With Acute Myeloid Leukemia. 30 J of Clin Onc. 36. DOI: 10.1200/JCO.2012.43.4738

Mrózek, K., Marcucci, G., Paschka, P., Whitman, S. P., & Bloomfield, C. D. (2007). Clinical relevance of mutations and gene-expression changes in adult acute myeloid leukemia with normal cytogenetics: are we ready for a prognostically prioritized molecula

Mulligan G et al. Mutation of NRAS but not KRAS significantly reduces myeloma sensitivity to single- agent bortezomib therapy. Blood, 2014;123:632-639.

Murphy SJ, Hart SN, Lima JF, et al. Genetic alterations associated with progression from pancreatic intraepithelial neoplasia to invasive pancreatic tumor. Gastroenterology 2013;145:1098-109.e1.

Nanri T et al. Imatinib mesylate for refractory acute myeloblastic leukemia harboring inv(16) and a C-KIT

Nault J. C., & Zucman Rossi J. Molecular Classification of Hepatocellular Adenomas. International Journal of Hepatology, 2013, 315947. doi:10.1155/2013/315947

NCCN clinical Practice Guidelines in Oncology (NCCN Guidelines) Acute Lymphoblastic Leukemia 2.2015. NCCN.org

NCCN Clinical Practice Guidelines in Oncology (NCCN Guidelines) Acute Myeloid Leukemia Version 1.2015. NCCN.org

NCCN clinical Practice Guidelines in Oncology (NCCN Guidelines) Colon Version 2.2016. NCCN.org

NCCN clinical Practice Guidelines in Oncology (NCCN Guidelines) Genetic/Familial High-Risk Assessment: Colorectal Version 2.2015. NCCN.org

NCCN clinical Practice Guidelines in Oncology (NCCN Guidelines) Melanoma Version 2.2016. NCCN.org

NCCN clinical Practice Guidelines in Oncology (NCCN Guidelines) Multiple myeloma Version 2.2016. NCCN.org

NCCN clinical Practice Guidelines in Oncology (NCCN Guidelines) Myelodysplastic syndromes. Version 1. 2016 NCCN.org.

NCCN clinical Practice Guidelines in Oncology (NCCN Guidelines) Myelodysplastic syndromes. Version 1. 2016 NCCN.org

NCCN clinical Practice Guidelines in Oncology (NCCN Guidelines) Non-hodgkin's Lymphomas Version 1. 2016. NCCN.org

NCCN clinical Practice Guidelines in Oncology (NCCN Guidelines) Non-Small Cell Lung Cancer , 2016.

NCCN clinical Practice Guidelines in Oncology (NCCN Guidelines) Pancreatic Adenocarcinoma Version 2. 2015. NCCN.org.

NCCN clinical Practice Guidelines in Oncology (NCCN Guidelines) Soft tissue Sarcoma Version 1. 2015. NCCN.org 2. 2016. NCCN.org

NCCN clinical Practice Guidelines in Oncology (NCCN Guidelines) Thyroid Version 2.2015. NCCN.org

Nikiforov et al. Impact of Mutational Testing on the Diagnosis and Management of Patients with Cytologically Indeterminate Thyroid Nodules: A Prospective Analysis of 1056 FNA Samples. J Clin Endocrinol Metab, 2011, 96 (11):3390-3397.

Nikiforov et al. Impact of Mutational Testing on the Diagnosis and Management of Patients with Cytologically Indeterminate Thyroid Nodules: A Prospective Analysis of 1056 FNA Samples. J Clin Endocrinol Metab, 2011, 96 (11):3390-3397.

Nikiforova MN1, Wald AI, Roy S, Durso MB, Nikiforov YE. Targeted next-generation sequencing panel (ThyroSeq) for detection of mutations in thyroid cancer. J Clin Endocrinol Metab. 2013 Nov;98(11):E1852-60. doi: 10.1210/jc.2013-2292.

Nikiforova, et al. Integration of KRAS testing in the diagnosis of pancreatic cystic lesions: a clinical experience of 618 pancreatic cysts. Mod Pathol 2013 Nov; 26(11):1478-87.

Paschka et al. Adverse Prognostic Significance of KIT Mutations in Adult Acute Myeloid Leukemia With inv(16) and t(8;21): A Cancer and Leukemia Group B Study. J. Clin. Oncol. 2006;24:3904-3911.

Patel, J. Prognostic Relevance of Integrated Genetic Profiling in Acute Myeloid Leukemia. N Engl J Med 2012; 366:1079-1089March 22, 2012DOI: 10.1056/NEJMoa1112304.

Prostate-specific antigen velocity at low prostate-specific antigen levels as screening tool for prostate cancer: results of second screening round of ERSPC (ROTTERDAM). AU Roobol MJ, Kranse R, de Koning HJ, Schröder FH

Prostate-specific antigen velocity for early detection of prostate cancer: result from a large, representative, population-based cohort. Vickers AJ, Wolters T, Savage CJ, Cronin AM, O'Brien MF, Pettersson K, Roobol MJ, Aus G, Scardino PT, Hugosson J, Schr

Robinson GW1, Orr BA, Gajjar A. Complete clinical regression of a BRAF V600E-mutant pediatric glioblastoma multiforme after BRAF inhibitor therapy. BMC Cancer. 2014Apr 12;14:258. doi: 10.1186/1471- 2407-14-258.

Roden AC et al. BRAF V600E expression in Langerhans cell histiocytosis: clinical and Immunohistochemical study on 25 pulmonary and 54 extrapulmonary cases. Am J Surg Pathol. 2014 Apr;38(4):548-51. doi: 0.1097/PAS.0000000000000129.

Rossi D. Clinical impact of small TP53 mutated subclones in chronic lymphocytic leukemia. Blood. 2014;123(14):2139-47.

Rossi D. Clinical impact of small TP53 mutated subclones in chronic lymphocytic leukemia. Blood. 2014;123(14):2139-47.

Schnittger S, Bacher U, Haferlach C, et al. Characterization of NPM1-mutated AML with a history of myelodysplastic syndromes or myeloproliferative neoplasms. Leukemia. 2011; 25: 615-621.

Schonleben et al. PIK3CA, KRAS, and BRAF mutations in intraductal papillary mucinous neoplasm/carcinoma (IPMN/C) of the pancreas. Langenbecks Arch Surg. 2008, 393:289-96.

Sehgal, A. R., Gimotty, P. A., Zhao, J., Hsu, J., Daber, R., Morrissette, J. D., et al. (2015). DNMT3A Mutational Status Affects the Results of Dose-Escalated Induction Therapy in Acute Myelogenous Leukemia. Clinical Cancer Research, 21(7), 1614-1620. doi

Shao H et al. Distinguishing hairy cell leukemia variant from hairy cell leukemia: development and validation of diagnostic criteria. Leuk Res. 2013 Apr;37(4):401-9. doi:10.1016/j.leukres.2012.11.021. Epub 2013 Jan 22.

Singhi AD, Nikiforova MN, Fasanella KE, et al. Preoperative GNAS and KRAS testing in the diagnosis of pancreatic mucinous cysts. Clin Cancer Res 2014;20:4381-9.

Systematic review of pretreatment prostate-specific antigen velocity and doubling time as predictors for prostate cancer.

Tanaka M, Fernández-del Castillo C, Adsay V, et al. International consensus guidelines 2012 for the management of IPMN and MCN of the pancreas. Pancreatology 2012;12:183-97.

Tefferi etal, Leukemia (2014) 28, 1407–1413. Documented in current WHO as major criterion for diagnosis (p 44 and 48) Swerdlow SH, Camp E, Harris NL, Jaffe ES, Pileri SA, Stein H, Thiele J, Vardiman JW (Eds): WHO Classification of Tumours of the Hematopoi

Tejpar S et al. Predominance of beta-catenin mutations and beta-catenin dysregulation in sporadic aggressive fibromatosis (desmoid tumor). Oncogene 1999; 18(47):6615

Therkildsen C et al. The predictive value of KRAS , NRAS , BRAF , PIK3CA and PTEN for anti-EGFR treatment in metastatic colorectal cancer: A systematic review and meta-analysis. Acta Oncologica, 2014; 53: 852–864

Thiruvengadam N and Park WG. Systematic Review of Pancreatic Cyst Fluid Biomarkers: The Path Forward. Clinical and Translational Gastroenterology (2015) 6, e88; doi:10.1038/ctg.2015.17

Urology. 2004;63(2):309.

Valent P et al. Guidelines and diagnostic algorithm for patients with suspected systemic mastocytosis: a proposal of the Austrian competence network (AUCNM). Am J Blood Res. 2013 May 5;3(2):174-

van Engen-van Grunsven AC et al. HRAS-mutated Spitz tumors: A subtype of Spitz tumors with distinct features. Am J Surg Pathol. 2010 Oct;34(10):1436-41. doi:10.1097/PAS.0b013e3181f0a749.

Verma S et al. Rapid Detection and Quantitation of BRAF Mutations in Hairy Cell Leukemia Using a Sensitive Pyrosequencing Assay. Am J Clin Pathol 2012;138:153-156, DOI: 10.1309/AJCPL0OPXI9LZITV

Vickers AJ, Savage C, O'Brien MF, Lilja H J Clin Oncol. 2009;27(3):398

Wen, X., Hu, J., Yang, J., Qian, W., Yao, D., Deng, Z., . . . Qian, J. (2015). CEBPA methylation and mutation in myelodysplastic syndrome. Medical Oncology Med Oncol, 32(7). doi:10.1007/s12032-015-0605-z

Wu J, Matthaei H, Maitra A, et al. Recurrent GNAS mutations define an unexpected pathway for pancreatic cyst development. Sci Transl Med 2011;3:92ra66.

Yoshida, K., Frequent pathway mutations of splicing machinery in myelodysplasia. Nature. 2011 Sep 11;478(7367):64-9. doi: 10.1038/nature10496.

Zadeh G, Bhat Krishna PL, Aldape K. EGFR and EGFRvIII in Glioblastoma: Partners in Crime.Cancer Cell. 2013;24:403–404.

Zenz, T et al. TP53 mutation and survival in chronic lymphocytic leukemia. J Clin Oncol. 2010 28(29):4473-9.

APPENDIX A

In addition to the recommendations above, we believe that **Acute Leukemia and Myeloid Neoplasm/Leukemia** warrant substantial additional coverage and considerations. To support our arguments, we have included current evidence, background, and best practices.

Background: Molecular testing to aid Diagnosis for Therapeutic Decision making in Acute Leukemia.
The correct therapy for acute leukemia depends on correct classification of the leukemia. Morphology alone (pathologist review of tissue on slide) is not sufficient to provide determine the classification. 2016 WHO guidelines define 11 acute myeloid leukemia classifications defined by a recurrent genetic abnormality. Consequently molecular testing is needed to classify acute myeloid leukemia (2016 WHO revision, summarized in Table A), see PML/RARA and BCR-ABL1 and RUNX1 below. Codes in red are currently not covered procedures by DL35000.

Table A. AML with recurrent genetic abnormalities (adapted from WHO 2016)
81401 RUNX1/RUNX1T1, AML with t(8;21) (q22;q22.1)
81401 CBFB-MYH11, AML with inv(16)(p13.1q22) or t(16;16)(p13.1,q22)
81315 81316, APL with PML-RARA
81401 MLL/MLLT3 or MLLT3-KMT2A, AML with t(9;11)(p21.3;q23.3)
81401 DEK-NUP214, AML with t(6;9)(p23;q34.1)
81479 GATA2, MECOM, AML with inv(3)(q21.3q26.2) or t(3;3)(q21.3q26.2)
81206, 81207, 81208 AML with BCR-ABL1
81310 AML with mutated NPM1
81218 AML with biallelic mutations of CEBPA
81479 AML with mutated RUNX1 (runt related transcription factor 1)

Background: Molecular testing to aid Risk Classification for Therapeutic Decision making in Acute Leukemia.
The 2016 ver2 AML NCCN guidelines state "Molecular abnormalities (KIT, FLT-ITD, NPM1, CEBPA, and other mutations) are important for prognostication in a subset of patients (category 2A) and may guide therapeutic intervention (category 2B, see AML-A)… if a test is not available at your institution, consult pathology about

preserving material from the original diagnostic sample for future use at an outside reference lab after full cytogenetic data are available."

The correct therapy for acute leukemia depends on correct risk classification of the leukemia. Morphology alone (pathologist review of tissue on slide) is not sufficient to provide determine the risk classification. Current NCCN guidelines use recurrent genetic abnormalities to stratify patients into three risk classifications favorable, intermediate, and poor-risk (see Table B below, NCCN AML-A, WHO Supplemental Table 2). In patients >= 60 years, NCCN guidelines recommend different approaches to "Treatment Induction" and "Post Remission Therapy" based on favorable versus unfavorable molecular markers (see AML-11, AML-13). In patients <60 years, NCCN guidelines recommend different "Post Remission Therapies" based on risk status defined by molecular abnormalities (eg chemotherapy versus matched sibling or alternative donor hematopoietic cell transplantation [HCT]) (see AML-10) Furthermore, NCCN guidelines state "For older patients (age >60 years) with AML the panel recommends using patient performance status, in addition to adverse features (eg de novo AML without unfavorable cytogenetics or molecular markers; therapy-related AML; antecedent hematologic disorder) and comorbid conditions, to select treatment options rather than rely on a patient's chronologic age alone". Consequently, "post remission therapy" for older patients (age >60 years) with good performance status can follow the same algorithm as patients <60 years based on risk classification based on recurrent genetic abnormalities.

Table B. AML RISK status based on validated genetic abnormalities: (adapted from NCCN 2016 ver2, AML-A, WHO 2016 supplemental table 1,2)

RISK STATUS	GENETIC ABNORMALITY	KARYOTYPE	CPT	REF	L35000 STATUS
Favorable	CBFB-MYH11	same	81401	1	Tier 2 code, covered
*Favorable	PML-RARA	same	81315, 81316	1	Tier 1 that require individual review
Favorable	RUNX1/RUNX1T1	t(8;21)	81401	1	Tier 2 code, covered
Favorable	CEBPA mutant (isolated biallelic)	norm/abn	81218	1,2	Tier 1 code, covered
Favorable	NPM1 mutant (FLT3-ITD-)	norm	81310	1,2	Tier 1 code, covered
Intermediate	MLL/MLLT3 or MLLT3-KMT2A	t(9;11)	81401 [a]	1,2	no guidance
Intermediate	DNMT3A mutant (NPM1+/FLT3-ITD-)	norm	81403	2	no guidance
Intermediate	IDH1 mutant (NPM1+/FLT3-ITD-)	norm	81403	2	Tier 2 that require individual review
Intermediate	TET2 mutant (NPM1+/FLT3-ITD-)	norm	81479	2	no guidance
Poor	BCR-ABL1	t(9;22)	81206, 81207, 81208	1	Tier 1 code, new indication
Poor	DEK-NUP214	t(6;9)	81401 [b]	1,2	Tier 2 code, Not covered
Poor	GATA2_MECOM	inv(3)	81479	1,2	no guidance
Poor	ASXL1 mutant	norm/int/abn	81479	2	no guidance
Poor	FLT3-ITD mutant	norm/abn	81245, 81246	1,2	Tier 1 code, covered
Poor	KIT mutant	t(8;21)	81272,81273	1[c],2	Tier 1 code, covered
Poor	RUNX1 mutant	norm/abn	81479	1,2	no guidance
Poor	TP53 mutant	norm/abn	81404	1,2	Tier 2 code, Not covered
Poor	WT1 mutant	norm/abn	81479	2	no guidance

abn = abnormal karyotype (usually not complex karyotype),
norm=normal karyotype
int = intermediate karyotype
[a] (frequency in children versus adults = 9.5% versus 2.0%)
[b] (frequency in children versus adults = 1.7% versus 1.0%)
[c] Discussed in NCCN algorithims and Discussion (AML-1 algorithm)
Ref 1 = 2016 ver2 NCCN, Ref 2 = 2016 WHO revision

Background: Molecular testing to aid Diagnosis for Therapeutic Decision making in Myeloproliferative Neoplasms/Myelodysplastic Syndromes.

A diagnosis of myeloproliferative neoplasms/myelodysplastic syndrome is necessary to initiate treatment. Failure to make a diagnosis holds back treatment and could result in progression from chronic to acute phase. The negative impact of withholding a diagnosis would be severe for the patient in this instance. The 2016 revision to the World Health Organization classification of myeloid neoplasms have added recurrent genetic abnormalities as a diagnostic criteria for chronic neutrophilic leukemia (CNL, gene CSF3R), chronic myelomonocytic leukemia (CMML, genes TET2, SRSF2, ASXL1 in >80% of cases and SETBP1, NRAS/KRAS,

RUNX1, CBL and EZH2 at a lower frequency) and MDS/MPN with ring sideroblasts (gene SF3B1). *Please note that this is not an exhaustive list of genetic abnormalities used as diagnostic criteria in the the 2016 WHO revision.*

A diagnosis of MDS requires stable cytopenia, exclusion of other disorders as a primary reason for cytopenia, and one of three MDS-related (decisive) criteria: 1) dysplasia 2) blast cell 5-19% or 3) MDS karyotype. In instances cytopenia persists yet none of these criteria provide a definitive diagnosis. In these instances, 2016 NCCN version 1 guidelines state that co-criteria may help confirm the diagnosis of MDS. These co-criteria include "molecular marker analysis (to detect or exclude abnormal CD34 antigen expression, fibrosis dysplastic megakaryocytes, atypical localization of immature progenitors)." "MDS-associated gene mutations can establish the presence of clonal hematopoiesis, which can help exclude benign causes of cytopenias in cases with non-diagnostic morphology" (2016 NCCN guidelines). The 2016 WHO revision reports "targeted sequencing of a limited number of genes can detect mutations in 80% to 90% of MDS patients; the most commonly mutated genes in MDS are SF3B1, TET2, SRSF2, ASXL1, DNMT3A, RUNX1, U2AF1, TP53 and EZH2."

Background: Molecular testing to aid RISK Classification for Therapeutic Decision making in MPN/MDS.
In addition to diagnosis of the neoplasm, the correct therapy for MPN/MDS depends on correct risk classification of the neoplasm. 2016 ver1 NCCN guidelines use a combination of MDS subtype (based on diagnosis eg MDS versus CMML) and prognostic scoring systems (eg IPSS, IPSS-R, WPSS) to determine the risk classification of the neoplasm (see NCCN 2016 ver1, MDS-3,4,5,6,7). This approach stratifies MDS patients into five risk classifications very low, low, intermediate, high and very high. NCCN guidelines recommend different "Treatment" based on prognostic category ranging from immune suppression therapy (IST) for low risk, hypomethylating agents or lenalidomide for intermediate risk to chemotherapy followed by HCT for high risk patients (see MDS-9,10,11 2016 NCCN ver1).

For patients who fall into the intermediate risk prognostic category 2016 NCCN guidelines recommend "molecular testing for recurrently mutated genes in this clinically appropriate context"(see NCCN 2016 ver1, MDS-2). Certain gene mutations (TP53, ASXL1, ETV6, RUNX1, and EZH2) can refine the prognosis of MDS in patients risk stratified by the IPSS or IPSS-R and may be helpful in patients predicted to have intermediate risk (see NCCN 2016 ver1, MDS-2,7). Mutations in TP53, ASXL1, ETV6, RUNX1, and EZH2 hold independent prognostic value and predict decreased OS in multivariable models adjusted for IPSS or IPSS-R risk groups in several studies of distinct cohorts (see NCCN 2016 ver1 discussion). "When applied to patients stratified by the IPSS-R, the presence of a mutation in one or more of these five genes was associated with shorter OS for patients in the low and intermediate-risk groups." (see NCCN 2016 discussion). Furthermore, 2016 WHO revision states that "the number and types of specific mutations are strongly associated with disease outcome in MDS, and the addition of mutation data improves the prognostic value of existing risk-stratification schemes in MDS". For example, "TP53 mutation is associated with aggressive disease in MDS in general and appears to predict poorer response to lenalidomide in patient with del(5q). Evaluation for TP53 mutation is recommended in patients with MDS with isolated del(5q) to help identify an adverse prognostic subgroup in this generally favorable prognosis MDS entity." (r2016 WHO revision).Oncologists use this mutation status to determine the most appropriate therapy (eg. chemotherapy alone versus transplant)(Cargo, 2015; Cazzola, 2013; Genovese, 2014; Jaiswal, 2014).

Table C. MDS/MPN RISK status for validated genetic abnormalities: (adapted from NCCN 2016 ver2, MDS-7)

RISK STATUS	GENETIC ABNORMALITY	KARYOTYPE	CPT	REF	L35000 STATUS
low	SF3B1 mutant	good/int	81479	1,2	no guidance
high	ASXL1 mutant	good/int	81479	2	no guidance
high	RUNX1 mutant	good/int	81479	1,2	no guidance
high	TP53 mutant	good/int	81404	1,2	Tier 2 code, not covered
high	EZH2 mutant	good/int	81479	1,2	no guidance
high	ETV6 mutant	good/int	81479	1,2	no guidance

REFERENCES

Arber DA et al. The Updated WHO Classification of Hematological Malignancies. The 2016 revision of the World Health Organization classification of myeloid neoplasms and acute leukemia. Blood 2016;127(20);2391-2405

NCCN Clnical Practice Guidelines in Oncology (NCCN Guidelines) Acute Myeloid Leukemia Version 2.2016 NCCN.org.