BlueCross BlueShield Federal Employee Program.

FEP Medical Policy Manual

FEP 2.01.68 Laboratory Tests Post Transplant and for Heart Failure

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Related Policies:

7.03.08 - Heart/Lung Transplant
7.03.09 - Heart Transplant

Laboratory Tests Post Transplant and for Heart Failure

Description

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Clinical assessment and noninvasive imaging of chronic heart failure can be limited in accurately diagnosing patients with heart failure because symptoms and signs can poorly correlate with objective methods of assessing cardiac dysfunction. For management of heart failure, clinical signs and symptoms (eg, shortness of breath) are relatively crude markers of decompensation and occur late in the course of an exacerbation. Thus, circulating biomarkers have potential benefit in heart failure diagnosis and management.

Noninvasive Heart Transplant Rejection Tests

Presage ST2 Assay

In addition to its use as a potential aid to predict prognosis and manage therapy of heart failure, elevated serum ST2 levels have also been associated with an increased risk of antibody-mediated rejection (AMR) following a heart transplant. For this reason, ST2 has also been proposed as a prognostic marker post heart transplantation and as a test to predict acute cellular rejection (graft-versus-host disease). The Presage ST2 Assay, described above, is a commercially available sST2 test that has been investigated as a biomarker of heart transplant rejection.

Heartsbreath Test

The Heartsbreath test, a noninvasive test that measures breath markers of oxidative stress, has been developed to assist in the detection of heart transplant rejection. In heart transplant recipients, oxidative stress appears to accompany allograft rejection, which degrades membrane polyunsaturated fatty acids and evolving alkanes and methylalkanes that are, in turn, excreted as volatile organic compounds in breath. The

Heartsbreath test analyzes the breath methylated alkane contour, which is derived from the abundance of C4 to C20 alkanes and monomethylalkanes and has been identified as a marker to detect grade 3 (clinically significant) heart transplant rejection.

HeartCare

Cell-free DNA (cfDNA), released by damaged cells, is normally present in healthy individuals.[23,] In patients who have received transplants, donor-derived cfDNA (dd-cfDNA) may be also present. It is proposed that allograft rejection, which is associated with damage to transplanted cells, may result in an increase in dd-cfDNA. HeartCare (CareDx) is a commercially-available test that combines AlloMap gene expression profiling with a next-generation sequencing assay that quantifies the fraction of dd-cfDNA in cardiac transplant recipients relative to total cfDNA. The AlloMap score, AlloMap score variability, and AlloSure % dd-cfDNA are reported.

Prospera

Prospera Heart (Natera) is a commercially available assay that uses massively multiplexed PCR (mmPCR) followed by next-generation sequencing (NGS) to quantify the fraction of dd-cfDNA in transplant recipients. Donor versus recipient cfDNA is differentiated via an advanced bioinformatics analysis of >13,000 single-nucleotide polymorphisms (SNPs) without the need for prior recipient or donor genotyping or computational adjustments for related donors.[24,] The Prospera Heart test reports the dd-cfDNA fraction in the patient's blood as a predictor of acute rejection, although the optimal dd-cfDNA cut-point is not described by the manufacturer.

myTAI$_{HEART}$

Using proprietary myTAI$_{HEART}$ software (TAI Diagnostics), the myTAI$_{HEART}$ test uses multiplexed, high-fidelity amplification followed by allele-specific qPCR of a panel of 94 highly informative bi-allelic single nucleotide polymorphisms (SNPs) and 2 controls to quantitatively genotype cfDNA in the patient's plasma after cardiac transplant, and accurately distinguish dd-cfDNA originating from the engrafted heart from cfDNA originating from the recipient's native cells.[25,] The ratio of dd-cfDNA to total cfDNA is reported as the donor fraction (%) and categorizes the patient as at low or increased risk of moderate (grade 2R) to severe (grade 3R) ACR : low donor fractions indicate less damage to the transplanted heart and a lower risk for rejection, while increased donor fractions indicate more damage to the transplanted heart and an increased risk for rejection. Testing with myTAI$_{HEART}$ does not require a donor specimen. TAI Diagnostics suspended production of the myTAI$_{HEART}$ test in 2020. As of September 2022, TAI Diagnostics appears to no longer be operational and it is unclear if myTAI$_{HEART}$ will be available through another company in the future.

AlloMap

Another approach has focused on patterns of gene expression of immunomodulatory cells, as detected in the peripheral blood. For example, microarray technology permits the analysis of the expression of thousands of genes, including those with functions known or unknown. Patterns of gene expression can then be correlated with known clinical conditions, permitting a selection of a finite number of genes to compose a custom multigene test panel, which then can be evaluated using polymerase chain reaction techniques. AlloMap (CareDx) is a commercially available molecular expression test that has been developed to detect acute heart transplant rejection or the development of graft dysfunction. The test involves expression measurement of a panel of genes derived from peripheral blood cells and applies an algorithm to the results. The proprietary algorithm produces a single score that considers the contribution of each gene in the panel. The score ranges from 0 to 40. The AlloMap website states that a lower score indicates a lower risk of graft rejection; the website does not cite a specific cutoff for a positive test.[26,] All AlloMap testing is performed at the CareDx reference laboratory in California.

Other laboratory-tested biomarkers of heart transplant rejection have been evaluated. They include brain natriuretic peptide, troponin, and soluble inflammatory cytokines. Most have had low accuracy in diagnosing rejection. Preliminary studies have evaluated the association between heart transplant rejection and micro-RNAs or high-sensitivity cardiac troponin in cross-sectional analyses but the clinical use has not been evaluated.[27,28,]

Renal Transplant Rejection

Allograft dysfunction is typically asymptomatic and has a broad differential, including graft rejection. Diagnosis and rapid treatment are recommended to preserve graft function and prevent loss of the transplanted organ. For a primary kidney transplant from a deceased donor (accounting for about 70% of kidney donors), graft survival at 1 year is 93% ; at 5 years, graft survival is 74%.[29,30,]

Surveillance of transplant kidney function relies on routine monitoring of serum creatinine, urine protein levels, and urinalysis.[31,] Allograft dysfunction may also be demonstrated by a drop in urine output or, rarely, as pain over the transplant site. With clinical suspicion of allograft dysfunction, additional noninvasive workup including ultrasonography or radionuclide imaging may be used. A renal biopsy allows a definitive assessment of graft dysfunction and is typically a percutaneous procedure performed with ultrasonography or computed tomography guidance. Biopsy of a transplanted kidney is associated with fewer complications than biopsy of a native kidney because the allograft is typically transplanted more superficially than a native kidney. Renal biopsy is a low-risk invasive procedure that may result in bleeding complications; loss of a renal transplant, as a complication of renal biopsy, is rare.[32,]

Kidney biopsies allow for diagnosis of acute and chronic graft rejection, which may be graded using the Banff Classification.[33,34,] Pathologic assessment of biopsies demonstrating acute rejection allows clinicians to further distinguish between ACR and AMR, which are treated differently.

Noninvasive Renal Transplant Rejection Tests

AlloSure

AlloSure Kidney (CareDx) is a commercially available, next-generation sequencing assay that quantifies the fraction of dd-cfDNA in renal transplant recipients relative to total cfDNA by measuring 266 single nucleotide variants. Separate genotyping of the donor or recipient is not required but patients who receive a kidney transplant from a monozygotic (identical) twin are not eligible for this test. The fraction of dd-cfDNA relative to total cfDNA present in the peripheral blood sample is cited in the report. For patients undergoing surveillance, a routine testing schedule is recommended for longitudinal monitoring.

Prospera

Prospera Kidney (Natera) is a commercially available assay that quantifies the fraction of dd-cfDNA in renal transplant recipients. The manufacturer recommends use of the Prospera test when there is clinical suspicion of active rejection and for regular surveillance of subclinical rejection in renal transplant recipients.[35,] In a surveillance scenario, regular testing is recommended at 1, 2, 3, 4, 6, 9, and 12 months after renal transplant or most recent rejection. [36,] Thereafter, the test should be repeated quarterly. The proportion of dd-cfDNA relative to total cfDNA is reported, with detection of ≥1% dd-cfDNA indicating increased risk for active rejection. The percent dd-cfDNA change between tests is also reported.

Lung Transplant Rejection

Despite advances in induction and maintenance immunosuppressive regimens, lung transplant recipients have a median overall survival of 6 years, with more than a third of patients receiving treatment for acute rejection in the first year after transplant.[37,38,] Acute cellular rejection, lymphocytic bronchiolitis, and AMR are all risk factors for subsequent development of chronic lung allograft dysfunction (CLAD). Pathologic grading of ACR is based on the histological assessment of perivascular and interstitial mononuclear cell infiltrates. Antibody-mediated rejection may be clinical (symptomatic or asymptomatic allograft dysfunction) or subclinical (normal allograft function). Key diagnostic criteria established via consensus by the International Society for Heart and Lung Transplantation include the presence of antibodies directed toward donor human leukocyte antigens and characteristic lung histology with or without evidence of complement 4d within the graft.[39,] The most common phenotype of CLAD is a persistent obstructive decline in lung function known as bronchiolitis obliterans syndrome (BOS), which is graded based on the degree of decrease in FEV_1. Approximately 50% of patients develop BOS within 5 years post-transplant. Median survival following a diagnosis of BOS is 3-5 years. Acute rejection may present with non-specific physical symptoms or be asymptomatic. However, the role of surveillance bronchoscopy for screening asymptomatic patients for acute rejection is controversial, and performance of surveillance bronchoscopies varies across transplant centers.

Noninvasive Lung Transplant Rejection Tests

AlloSure

AlloSure Lung (CareDx) is a commercially available, NGS assay that quantifies the fraction of dd-cfDNA in lung transplant patients relative to total cfDNA by measuring single nucleotide polymorphisms. The test is intended to provide a direct, noninvasive measure of organ injury in lung transplant patients who are undergoing surveillance. Suggested thresholds for severe injury, injury, and quiescence are >0.9%, >0.5 to ≤0.9%, and <0.5%, respectively.[40,]

Prospera

Prospera Lung (Natera) is a commercially available assay that uses the same methodology as Propera Heart and Prospera Kidney to quantify the fraction of dd-cfDNA in transplant recipients. The Prospera Lung test reports the dd-cfDNA fraction in the patient's blood as a predictor of acute rejection, chronic rejection, or infection although the optimal dd-cfDNA cut-point for each outcome is not described by the manufacturer.[41,]

OBJECTIVE

The objective of this evidence review is to determine whether the measurement of various selected biomarkers improves the detection of allograft rejection in transplant patients or in the diagnosis and management of heart failure, thus improving net health outcomes.

POLICY STATEMENT

The use of the Presage ST2 Assay to evaluate the prognosis of individuals diagnosed with chronic heart failure is considered **investigational**.

The use of the Presage ST2 Assay to guide management (eg, pharmacologic, device-based, exercise) of individuals diagnosed with chronic heart failure is considered **investigational**.

The use of the Presage ST2 Assay in the post cardiac transplantation period, including but not limited to predicting prognosis and predicting acute cellular rejection, is considered **investigational.**

The measurement of volatile organic compounds to assist in the detection of moderate grade 2R (formerly grade 3) heart transplant rejection is considered **investigational**.

The use of peripheral blood measurement of dd-cfDNA in the post cardiac transplantation period, including but not limited to predicting prognosis and predicting acute cellular rejection, is considered **investigational.**

The use of peripheral blood gene expression profile tests alone or in combination with peripheral blood measurement of donor-derived cell-free DNA (dd-cfDNA) in the management of individuals after heart transplantation, including but not limited to the detection of acute heart transplant rejection or heart transplant graft dysfunction, is considered **medically necessary.**

The use of peripheral blood measurement of dd-cfDNA in the management of individuals after renal transplantation, including but not limited to the detection of acute renal transplant rejection or renal transplant graft dysfunction, is considered **investigational**.

The use of peripheral blood measurement of dd-cfDNA in the management of individuals after lung transplantation, including but not limited to the detection of acute lung transplant rejection or lung transplant graft dysfunction, is considered **investigational**.

POLICY GUIDELINES

Frequency of Testing:

For Plans choosing to provide coverage, the following information is provided to guide decisions on frequency of testing.

The U.S. Food and Drug Administration has indicated the following (see Rationale Section):

- Heartsbreath (Menssana Research) test is only for use as an aid in the diagnosis of grade 3 (now known as grade 2R) heart transplant rejection in patients who have received heart transplants within the preceding year and who have had endomyocardial biopsy within the previous month.
- AlloMap test is to be used in conjunction with clinical assessment, for aiding in the identification of heart transplant recipients with stable allograft function and a low probability of moderate-to-severe transplant rejection. It is intended for patients at least 15 years old who are at least 2 months post transplant.

American Society of Transplant Surgeons (see Supplemental Information Section):

Does not recommend a specific test for dd-cfDNA surveillance in kidney transplant recipients. Recommends testing monthly during months 1 to 4 post-transplant, then quarterly thereafter. The position supports ongoing surveillance because the risk of allograft injury or acute rejection persists and states that testing should continue indefinitely at some frequency, but it does not specify a long-term testing interval.

International Society for Heart and Lung Transplantation (see Supplemental Information Section):

Gene expression profiling (GEP) (ie, AlloMap) may be used in low-risk patients 2 months to 5 years after heart transplant to identify adult recipients at low risk of current acute cellular rejection and thereby reduce the frequency of endomyocardial biopsy. In pediatric patients, available data are insufficient to support a general recommendation for routine use of GEP at this time.

Medicare:

For heart, lung, and kidney transplants, a proposed (not yet finalized) Local Coverage Determination would recommend the following maximum number of surveillance timepoints in the first year post-transplant: kidney (4), heart (12), and lung (12), with surveillance thereafter limited to 2 tests per year.

BENEFIT APPLICATION

Experimental or investigational procedures, treatments, drugs, or devices are not covered (See General Exclusion Section of brochure).

State or federal mandates (eg, Federal Employee Program) may dictate that certain U.S. Food and Drug Administration-approved devices, drugs, or biologics may not be considered investigational, and thus these devices may be assessed only by their medical necessity.

FDA REGULATORY STATUS

The U.S. Food and Drug Administration (FDA) has cleared multiple biomarker tests for the detection of heart and renal allograft rejection. Table 1 provides a summary of the biomarker tests currently included in this policy that have FDA clearance.

Table 1. Select Biomarker Tests for Detection of Heart or Renal Allograft Rejection Cleared by the U.S. Food and Drug Administration

Test	Manufacturer	FDA Clearance Type, Product Number	FDA Clearance Date	Indicated Use
Heartsbreath™	Menssana Research	Humanitarian device exemption, H030004	2004	To aid in diagnosing grade 3 heart transplant rejection in patients who have received heart transplants within the preceding year. The device is intended as an adjunct to, and not as a substitute for, endomyocardial biopsy and is also limited to patients who have had endomyocardial biopsy within the previous month.
AlloMap Molecular Expression Testing	CareDx, formerly XDx	510(k), k073482	2008	The test is to be used in conjunction with clinical assessment, for aiding in the identification of heart transplant recipients with stable allograft function and a low probability of moderate-to-severe transplant rejection. It is intended for patients at least 15 years old who are at least 2 months post transplant.
Presage ST2 Assay kit	Critical Diagnostics	510(k), k093758	2011	For use with clinical evaluation as an aid in assessing the prognosis of patients diagnosed with chronic heart failure

FDA: Food and Drug Administration.

Laboratory Developed Tests

There are also commercially available laboratory-developed biomarker tests for the detection of heart and renal allograft rejection. Clinical laboratories may develop and validate tests in-house and market them as a laboratory service; laboratory-developed tests must meet the general regulatory standards of the Clinical Laboratory Improvement Amendments. The AlloSure (CareDx), and Prospera (Natera) dd-cfDNA tests are regulated under the Clinical Laboratory Improvement Amendments standards.

myTAI$_{HEART}$ is also a laboratory developed test (LDT) developed for clinical diagnostic performance exclusively in the College of American Pathologists (CAP) and Clinical Laboratory Improvement Amendment (CLIA) accredited TAI Diagnostics Clinical Reference Laboratory.[25,] This test was developed and its performance characteristics were determined by TAI Diagnostics.

These LDTs have not been cleared or approved by the FDA.

Other Tests

Other commercially available LDTs without FDA clearance or approval for use have been excluded from this evidence review when studies reporting on the clinical validity of the marketed version of the test could not be identified and/or where the test is marketed for research use only. Excluded tests and their descriptions are summarized for reference purposes in Table 2.

This policy does not address urine-based tests for allograft dysfunction (eg, One Lambda Laboratories" CXCL10).

Table 2. Biomarker Tests Excluded from Review

Test	Manufacturer	Technology	Indications for Use
AllosurePlus	CareDx	dd-cfDNA	A post-transplant rejection risk prediction algorithm that incorporates AlloSure dd-cfDNA in an artificial intelligence supported model. The test is intended for use post-transplant to provide a personalized probability score and rejection status as a complement to conventional biopsy and standard clinical assessment.
KidneyCare	CareDx	dd-cfDNA and GEP	Available as a research tool through the OKRA Registry.
AlloSeq HCT	CareDx	NGS	To aid in the assessment of engraftment following HCT via NGS analysis of 202 biallelic SNPs. The fraction of recipient and donor genomic DNA is reported. The test is marketed for research use only.
AlloSeq Tx17	CareDx	NGS	An NGS test utilizing Hybrid Capture Technology conducted pre-transplant to identify optimal transplant matches. The test sequences full HLA genes and other transplant-associated genes (KIR, MICA/B, C4, HPA, ABO). This test is marketed for research use only.
Viracor TRAC	Eurofins	dd-cfDNA	To aid in the diagnosis of solid organ transplant rejection via NGS analysis. The fraction of dd-cfDNA is reported.[1]
TruGraf Kidney	Eurofins	NGS	Identifies immune quiescence vs. risk for subclinical rejection in stable kidney transplant recipients to guide when a biopsy can be safely avoided.
OmniGraf Liver	Eurofins	NGS	Identifies immune quiescence vs. risk for subclinical rejection in stable liver transplant recipients to guide when a biopsy can be safely avoided.
VitaGraf™ Kidney	Oncocyte Corporation	dd-cfDNA	Detection of kidney allograft injury and acute rejection based on dd-cfDNA fraction.
VitaGraf™ Liver	Oncocyte Corporation	dd-cfDNA	Detection of liver allograft injury and acute rejection based on dd-cfDNA fraction.
MMDx Heart	Kashi Clinical Laboratories/Thermo Fisher	Tissue-based microarray	Tissue-based microarray mRNA gene expression test of 1283 genes post-transplant to provide a probability score of rejection as a complement to conventional biopsy

			processing. The test is not marketed to provide information for the diagnosis, prevention, or treatment of disease or to aid in the clinical decision-making process.
MMDx Kidney	Kashi Clinical Laboratories/Thermo Fisher	Tissue-based microarray	Tissue-based microarray mRNA gene expression test of 1494 genes post-transplant to provide a probability score of rejection as a complement to conventional biopsy processing. The test is not marketed to provide information for the diagnosis, prevention, or treatment of disease or to aid in the clinical decision-making process.
MMDx Lung	Kashi Clinical Laboratories/Thermo Fisher	Tissue-based microarray	Tissue-based microarray mRNA gene expression test post-transplant to provide a probability score of rejection as a complement to conventional biopsy processing. The test is not marketed to provide information for the diagnosis, prevention, or treatment of disease or to aid in the clinical decision-making process.

dd-cfDNA: donor-derived cell-free DNA; GEP: gene expression profiling; HCT: hematopoietic cell transplantation; HLA: human leukocyte antigen; MMDx: molecular microscope diagnostic system; NGS: next-generation sequencing; OKRA: Outcomes in KidneyCare in Renal Allografts; SNP: single-nucleotide polymorphism; TRAC: transplant rejection allograft check.

[1] Published studies reporting on the clinical validity of the marketed version of the test were not identified.

RATIONALE

Summary of Evidence

For individuals who have chronic heart failure who receive the sST2 assay to determine prognosis and/or to guide management, the evidence includes correlational studies and 2 meta-analyses. Relevant outcomes are OS, quality of life, and hospitalization. Most of the evidence is from reanalysis of existing randomized controlled trials (RCTs) and not from studies specifically designed to evaluate the predictive accuracy of sST2, and prospective and retrospective cross-sectional studies made up a large part of 1 meta-analysis. Studies have mainly found that elevated sST2 levels are statistically associated with an elevated risk of mortality. A pooled analysis of study results found that sST2 significantly predicted overall mortality and cardiovascular mortality. Several studies, however, found that sST2 test results did not provide additional prognostic information compared with N-terminal pro B-type natriuretic peptide levels. Moreover, no comparative studies were identified on the use of the sST2 assay to guide the management of patients diagnosed with chronic heart failure. The evidence is insufficient to determine that the technology results in an improvement in the net health outcome.

For individuals who have heart transplantation who receive sST2 assay to determine prognosis and/or to predict acute cellular rejection (ACR), the evidence includes a small number of retrospective studies on the Presage ST2 Assay. Relevant outcomes are overall survival (OS), morbid events, and hospitalization. No prospective studies were identified that provide high-quality evidence on the ability of sST2 to predict transplant outcomes. One retrospective study (n=241) found that sST2 levels were associated with ACR and mortality; another study (n=26) found that sST2 levels were higher during an acute rejection episode than before rejection. The evidence is insufficient to determine that the technology results in an improvement in the net health outcome.

For individuals who have a heart transplant who receive a measurement of volatile organic compounds to assess cardiac allograft rejection, the evidence includes a diagnostic accuracy study. Relevant outcomes are OS, test validity, morbid events, and hospitalizations. The published study found that, for identifying grade 3 (now grade 2R) rejection, the negative predictive value (NPV) of the breath test the study evaluated (97.2%) was similar to endomyocardial biopsy (96.7%) and the sensitivity of the breath test (78.6%) was better than that for biopsy (42.4%). However, the breath test had a lower specificity (62.4%) and a lower positive predictive value (PPV) (5.6%) in assessing grade 3 rejection than a biopsy (specificity, 97%; PPV, 45.2%). The breath test was also not evaluated for grade 4 rejection. This single study is not sufficient to determine the clinical validity of the test measuring volatile organic compounds and no studies on clinical utility were identified. The evidence is insufficient to determine that the technology results in an improvement in the net health outcome.

For individuals who have a heart transplant who receive dd-cfDNA testing to determine acute rejection, the evidence includes diagnostic accuracy studies. Relevant outcomes are OS, test validity, morbid events, and hospitalizations. Evidence from 3 studies suggests that the dd-cfDNA fraction is elevated in acute rejection, but optimal fraction cut-offs for detection of acute rejection have not been established. Using dd-cfDNA thresholds ranging from 0.12% to 0.32% resulted in NPVs ranging from 82% to 98% and area under the curve ranging from 0.61 to 0.86 in 3 studies. At present, no studies evaluating the clinical utility for the measurement of dd-cfDNA for heart transplant rejection have been identified. The evidence is insufficient to determine that the technology results in an improvement in the net health outcome.

For individuals who have a heart transplant who receive GEP to assess cardiac allograft rejection, the evidence includes 2 diagnostic accuracy studies and several RCTs evaluating clinical utility. Relevant outcomes are OS, test validity, morbid events, and hospitalizations. The 2 studies, Cardiac Allograft Rejection Gene Expression Observation (CARGO, CARGO II) examining the diagnostic performance of GEP for detecting moderate-to-severe rejection lacked a consistent threshold for defining a positive GEP test (ie, 20, 30, or 34) and reported a low number of positive cases. In the available studies, although the NPVs were relatively high (ie, at least 88%), the performance characteristics were only calculated based on few cases of rejection; therefore, performance data may be imprecise. Moreover, the PPV in CARGO II was only 4.0% for patients who were at least 2 to 6 months post transplant and 4.3% for patients more than 6 months post transplant. The threshold indicating a positive test that seems to be currently accepted (a score of 34) was not prespecified; rather it evolved partway through the data collection period in the Invasive Monitoring Attenuation through Gene Expression (IMAGE) study. In addition, the IMAGE study had several methodologic limitations (eg, lack of blinding); further, the IMAGE study failed to provide evidence that GEP offers an incremental benefit over biopsy performed on the basis of clinical exam or echocardiography. Patients at the highest risk of transplant rejection are patients within 1 year of the transplant, and, for that subset, there remains insufficient data on which to evaluate the clinical utility of GEP. The evidence is insufficient to determine that the technology results in an improvement in the net health outcome.

For individuals who have a heart transplant who receive GEP with testing of dd-cfDNA to assess cardiac allograft rejection, the evidence includes 2 retrospective analys of the HeartCare test and 1 diagnostic accuracy study of the AlloSure dd-cfDNA component of the HeartCare test. Relevant outcomes are OS, test validity, morbid events, and hospitalizations. The HeartCare analysis reported a 12.7% reduction in endomyocardial biopsy volume among patients undergoing routine surveillance. However, this observation is limited by lack of reporting on long-term health outcomes and incomplete assessment of diagnostic performance for combined testing, as patients with negative dd-cfDNA scores did not undergo biopsy regardless of GEP score per study protocol. A registry study found that combining GEP testing with AlloMap and dd-cfDNA testing with AlloSure Heart enhanced performance for acute cellular rejection surveillance compared to using either test alone, and a subsequent analysis of this registry found that linked dual-positive results were associated with a higher risk of graft dysfunction or cardiovascular death. The study also reported a reduction in biopsies performed between 2 to 12 months post-transplant, decreasing from 5.9 to 5.3 per patient, along with a decline in second-year biopsy rates from 1.5 to 0.9 per patient. A single-center study found that dual-positive and elevated dd-cfDNA with negative GEP were associated with significantly greater odds of biopsy-confirmed rejection and greater mortality. The evidence is insufficient to determine that the technology results in an improvement in the net health outcome.

For individuals with a renal transplant who are undergoing surveillance or have clinical suspicion of allograft rejection who receive testing of dd-cfDNA to assess renal allograft rejection, the evidence includes diagnostic accuracy studies. Relevant outcomes are OS, test validity, morbid events, and hospitalizations. Two studies examined the diagnostic performance of dd-cfDNA for detecting moderate-to-severe rejection; the NPV was moderately high (75% to 84%) with performance characteristics were calculated on cases of active transplant rejection. In 1 study, the threshold indicating a positive test was not prespecified. A subsequent smaller single-center study that explored variation in clinical validity based on different rejection mechanisms found the strongest performance characteristics for AlloSure with antibody-mediated rejection (AMR). Using dd-cfDNA threshold values from ≥0.5% to ≥1%, the Allosure test established a range of sensitivities from 19% to 86% and specifities of 72% to 100% for the detection of graft rejection. This corresponded to PPVs ranging from 21% to 77% and NPVs from 75% to 86%. A retrospective single-center study of the Prospera dd-cfDNA test reported a PPV and NPV of 52% and 95%, respectively, for detection of active rejection among a combined cohort of patients undergoing surveillance or for-cause biopsies, using the 1% dd-cfDNA threshold previously proposed for the AlloSure test. A second, retrospective study using the same dd-cfDNA threshold found a sensitivity and specificity of 77% and 84%, respectively, with an AUC of 0.88 for all rejection types. A prospective Prospera study reported PPVs of 68% and 71% and NPVs 91% and 83% using combined dd-cfDNA fraction and absolute quantity compared with 2 different reference standards. Larger prospective studies validating the dd-cfDNA thresholds for active rejection are needed to develop conclusions for each test. The evidence is insufficient to determine that the technology results in an improvement in the net health outcome.

For individuals with a lung transplant who receive testing of dd-cfDNA to assess lung allograft rejection, the evidence includes 5 small diagnostic accuracy studies. Relevant outcomes are OS, test validity, morbid events, and hospitalizations. One study examined the diagnostic performance of dd-cfDNA testing at a threshold of 0.87% for detecting acute cellular rejection, yielding a PPV of 34.1% and an NPV of 85.5%. A second study reported a PPV of 43.3% and NPV of 83.6% for an aggregate rejection cohort composed of patients with acute cellular rejection, AMR, and chronic lung allograft dysfunction (CLAD). In the third study, using a dd-cfDNA cut-off of 1.0%, PPV was 51.9% and NPV was 97.3% for acute rejection, and 43.6%, and 91.0% for acute rejection, CLAD/neutrophilic-responsive allograft dysfunction, or infection. One study that used dd-cfDNA testing as part of a home surveillance program found a PPV 43.4% and NPV 96.5% for detection of ACR, AMR, or infection, but when limited to patients with a contemporaneous reference standard surveillance bronchoscopy independent of dd-cfDNA level, PPV 66.7% and NPV was 79.2%. One study assessed dd-cfDNA for detecting acute rejection following single lung transplantation, using a cut-off adjusted for single-organ transplantation; the study reported a sensitivity of 77.8% and specificity of 84.6%, with a PPV of 38.3%, NPV of 96.8%, and an AUC of 0.85. All 5 studies were limited by small sample sizes, and no clinical utility studies were identified. The evidence is insufficient to determine that the technology results in an improvement in the net health outcome.

SUPPLEMENTAL INFORMATION

Practice Guidelines and Position Statements

Guidelines or position statements will be considered for inclusion in 'Supplemental Information' if they were issued by, or jointly by, a US professional society, an international society with US representation, or National Institute for Health and Care Excellence (NICE). Priority will be given to guidelines that are informed by a systematic review, include strength of evidence ratings, and include a description of management of conflict of interest.

American College of Cardiology et al

In 2022, the American College of Cardiology, American Heart Association, and Heart Failure Society issued an updated guideline for the management of heart failure.[8,] The 2022 guideline replaced a 2013 guideline[1,] and a 2017 focused guideline update.[105,] The guideline states measurement of natriuretic peptide levels may be useful for diagnosis, risk stratification, and prognosis of heart failure. The use of soluble suppression of tumorigenicity-2 is not discussed specifically, though the guideline notes that "a widening array of biomarkers including markers of myocardial injury, inflammation, oxidative stress, vascular dysfunction, and matrix remodeling have been shown to provide incremental prognostic information over natriuretic peptides but remain without evidence of an incremental management benefit."

American Society of Transplant Surgeons

In 2024, the American Society of Transplant Surgeons (ASTS) issued a position statement on the role of dd-cfDNA in kidney transplant surveillance.[106,] The authors summarized, "The accumulated evidence supporting the use of dd-cfDNA argues that its use should no longer be considered investigational. The use of dd-cfDNA is evidence-based and validated, and a prime example of much needed innovation in transplant organ surveillance. It has demonstrated utility in the early detection of allograft injury and assistance with clinical decision-making regarding allograft biopsy and treatment initiation." Regarding the timing of dd-cfDNA testing in kidney transplant, the ASTS states, "This protocol recommended testing at monthly intervals from month 1-4, and then quarterly thereafter. There are no data to support cessation of surveillance testing at any timepoint after transplant, and it is intuitively obvious that, as the risk of allograft injury or acute rejection does not disappear, testing should continue indefinitely at some frequency."

The following recommendations regarding the clinical utility and decision analysis were issued:

- "The most data have been accumulated in adult transplant recipients, and these recommendations are therefore most applicable to adult patient populations.
- We suggest that clinicians consider measuring serial dd-cfDNA levels in kidney transplant recipients with stable renal allograft function to exclude the presence of subclinical antibody-mediated rejection.
- We recommend that clinicians measure dd-cfDNA levels in kidney transplant recipients with acute allograft dysfunction to exclude the presence of rejection, particularly antibody-mediated rejection (ABMR).
- We do not recommend the use of blood gene expression profiling (GEP) in kidney transplant recipients for the purpose of diagnosing or excluding sub-clinical rejection, as adequate evidence supporting such use is still lacking.
- We do not recommend the use of blood GEP to diagnose or exclude the presence of acute graft rejection in kidney transplant recipients with acute allograft dysfunction given the paucity of data to support this practice.
- We recommend that dd-cfDNA may be utilized to rule out subclinical rejection in heart transplant recipients.
- We recommend that clinicians utilize peripheral blood GEP as a non-invasive diagnostic tool to rule out acute cellular rejection in stable, low-risk, adult heart transplant recipients who are over 55 days status post heart transplantation."

"Caveats and recommendations for future studies:

- None of these recommendations should be construed as recommending one biomarker over another in the same diagnostic niche.
- We strongly recommend ongoing clinical studies to clarify the scenarios in which molecular diagnostic studies should be utilized.
- We specifically recommend that studies be carried out to evaluate the potential role of dd-cfDNA surveillance in kidney transplant recipients to improve long-term allograft survival.
- We recommend studies be carried out to evaluate the potential global cost effectiveness of clinical use of dd-cfDNA testing in surveillance and for-cause settings."

European Association of Urology

The European Association of Urology (2024) provided guidance on renal transplantation including post-transplatation follow-up.

Table 3. Guidelines for Renal Transplant Recipient Follow-up

Recommendation	SOR
Provide lifelong regular post-transplant follow-up by an experienced and trained transplant specialist at least every six to twelve months.	Strong
Advise patients on appropriate lifestyle changes, potential complications, and the importance of adherence to their immunosuppressive regimen.	Strong
Regularly monitor (approximately every four to eight weeks) serum creatinine, estimated glomerular filtration rate, blood pressure, urinary protein excretion, immunosuppression and complications after renal transplantation. Changes in these parameters over time should trigger further diagnostic work-up including renal biopsy, a search for infectious causes and anti-HLA antibodies.	Strong
Perform an ultrasound of the graft, in case of graft dysfunction, to rule out obstruction and renal artery stenosis.	Strong
In patients with interstitial fibrosis and tubular atrophy undergoing calcineurin inhibitor (CNI) therapy and/or with histological signs suggestive for CNI toxicity (e.g. arteriolar hyalinosis, striped fibrosis) consider CNI reduction or withdrawal.	Strong
Initiate appropriate medical treatment, e.g. tight control of hypertension, diabetes, proteinuria, cardiac risk factors, infections, and other complications according to current guidelines	Strong

SOR: strength of recommendation.

European Society for Organ Transplantation

In 2024, the European Society for Organ Transplantation issued updated guidelines for the care of heart transplant recipients.[107,] The guidelines included the following recommendations including a strong recommendation for GEP based on a moderate level of evidence (see Table 4).

- 'Peripheral blood GEP assay (marketed in United States as Allomap) is a reliable non-invasive diagnostic tool to rule out acute cellular rejection in stable, low-risk heart transplant recipients >15 years of age who are >55 days post HT.'

Table 4. Guidelines for Renal Transplant Recipient Follow-up

PICO question	Recommendation	Level of evidence	
In heart transplant patients with stable graft function, are GEP and dd-cfDNA reliable surveillance tools for subclinical acute rejection monitoring, compared to endomyocardial biopsy?	GEP	Strong for	Moderate
	Cf-ddDNA	Weak for	Low
In heart transplant patients, are dd-cfDNA and GEP reliable methods to monitor for cardiac allograft vasculopathy as compared with standard diagnostic methods?		Weak against	Very Low
In heart transplant patients with stable graft function, is dd-cfDNA or GEP a reliable marker to stratify prognosis as compared to standard clinical classifiers?		Weak against	Very Low
Is dd-cfDNA a reliable marker to diagnose/monitor a) clinical and subclinical acute rejection or b) infection of the graft in lung transplant patients, compared with standard diagnostic methods (surveillance bronchoscopy with TBB for histopathology and bronchoalveolar lavage for microbiology testing)?	5A	Weak for	Low
	5B	Weak for	Very Low
Is dd-cfDNA a reliable therapeutic marker to monitor treatment response for acute rejection or infection of the graft in lung transplant patients, compared with standard diagnostic methods (i.e., follow-up surveillance TBBx)?		Weak against	Very Low

Is dd-cfDNA a reliable marker to stratify prognosis of lung transplant recipients for chronic lung allograft dysfunction (CLAD), as compared to standard clinical classifiers?		Weak for	Very Low

International Society of Heart and Lung Transplantation

In 2022, the International Society of Heart and Lung Transplantation issued updated guidelines for the care of heart transplant recipients. Key recommendations relevant to rejection surveillance and noninvasive testing are summarized in Table 5.[108, 11]). A subsequent 2025 update discusses the potential utility of dd-cfDNA and Molecular Microscope (MMDx) findings in the evaluation and management of post-transplant donor-specific antibodies (DSA), but does not assign a specific class of recommendation or level of evidence.[109,]

'Several variables should be considered to treat patients with post-transplant DSA, including cardiac dysfunction by imaging, abnormal hemodynamics, an increasing MFI trend in DSA, the presence of early DSA, antibody attributes (C1q+ binding, detection of DSA in dilution assay), biopsy-proven pAMR ≥2, and abnormal molecular findings (dd-cfDNA and/or the MMDx) as available.'

Table 5. Guidelines for Postoperative Care of Heart Transplant Recipients

Recommendation	COR	LOE
"It is reasonable to perform periodic EMB during the first 3 to 12 postoperative months for surveillance of HT rejection."	IIa	C
"After the first post-operative year, it is reasonable to continue EMB surveillance in patients who are at higher risk for late acute rejection..."	IIa	C
"Gene Expression Profiling (GEP) (i.e., AlloMap) of peripheral blood can be used in low-risk patients between 2 months and 5 years after HT to identify adult recipients who have low risk of current ACR to reduce the frequency of EMB. Data in children does not allow a general recommendation of GEP as a routine tool at present."	IIa	B

ACR: acute cellular rejection; COR: class of recommendation; EMB: endomyocardial biopsy; HT: heart transplant; LOE: level of evidence.

Kidney Disease Improving Global Outcomes

The Kidney Disease Improving Global Outcomes (2009) issued guidelines for the care of kidney transplant recipients.[110,] The guidelines included the following recommendations (see Table 6).

Table 6. Guidelines for Biopsy in Renal Transplant Recipients

Recommendation	SOR	LOE
"We recommend kidney allograft biopsy when there is a persistent, unexplained increase in serum creatinine."	Level 1	C
"We suggest kidney allograft biopsy when serum creatinine has not returned to baseline after treatment of acute rejection."	Level 2	D
"We suggest kidney allograft biopsy every 7-10 days during delayed function."	Level 2	C
"We suggest kidney allograft biopsy if expected kidney function is not achieved within the first 1-2 months after transplantation."	Level 2	D
"We suggest kidney allograft biopsy when there is new onset of proteinuria."	Level 2	C

"We suggest kidney allograft biopsy when there is unexplained proteinuria ≥3.0 g/g creatinine or ≥3.0 g per 24 hours."	Level 2	C

LOE: level of evidence; SOR: strength of recommendation.

U.S. Preventive Services Task Force Recommendations

Not applicable.

Medicare National Coverage

The Centers for Medicare & Medicaid Services (2008) issued a noncoverage decision for the Heartsbreath test.[111,] The Centers determined that the evidence did not adequately define the technical characteristics of the test; nor did it demonstrate that Heartsbreath testing could predict heart transplant rejection, and therefore the test would not improve health outcomes in Medicare beneficiaries.

For AlloMap, HeartCare, AlloSure, Prospera, myTAI$_{HEART,}$ and the Presage ST2 Assay there are no national coverage determinations. In the absence of a national coverage determination, coverage decisions are left to the discretion of local Medicare carriers.

REFERENCES

1. Yancy CW, Jessup M, Bozkurt B, et al. 2013 ACCF/AHA guideline for the management of heart failure: a report of the American College of Cardiology Foundation/American Heart Association Task Force on Practice Guidelines. J Am Coll Cardiol. Oct 15 2013; 62(16): e147-239. PMID 23747642
2. Roger VL, Go AS, Lloyd-Jones DM, et al. Heart disease and stroke statistics--2011 update: a report from the American Heart Association. Circulation. Feb 01 2011; 123(4): e18-e209. PMID 21160056
3. Virani SS, Alonso A, Benjamin EJ, et al. Heart Disease and Stroke Statistics-2020 Update: A Report From the American Heart Association. Circulation. Mar 03 2020; 141(9): e139-e596. PMID 31992061
4. Rohde LE, Beck-da-Silva L, Goldraich L, et al. Reliability and prognostic value of traditional signs and symptoms in outpatients with congestive heart failure. Can J Cardiol. May 15 2004; 20(7): 697-702. PMID 15197422
5. Marcus GM, Gerber IL, McKeown BH, et al. Association between phonocardiographic third and fourth heart sounds and objective measures of left ventricular function. JAMA. May 11 2005; 293(18): 2238-44. PMID 15886379
6. Stevenson LW, Perloff JK. The limited reliability of physical signs for estimating hemodynamics in chronic heart failure. JAMA. Feb 10 1989; 261(6): 884-8. PMID 2913385
7. Gaggin HK, Januzzi JL. Biomarkers and diagnostics in heart failure. Biochim Biophys Acta. Dec 2013; 1832(12): 2442-50. PMID 23313577
8. Heidenreich PA, Bozkurt B, Aguilar D, et al. 2022 AHA/ACC/HFSA Guideline for the Management of Heart Failure: A Report of the American College of Cardiology/American Heart Association Joint Committee on Clinical Practice Guidelines. Circulation. May 03 2022; 145(18): e895-e1032. PMID 35363499
9. McMurray JJ, Adamopoulos S, Anker SD, et al. ESC guidelines for the diagnosis and treatment of acute and chronic heart failure 2012: The Task Force for the Diagnosis and Treatment of Acute and Chronic Heart Failure 2012 of the European Society of Cardiology. Developed in collaboration with the Heart Failure Association (HFA) of the ESC. Eur J Heart Fail. Aug 2012; 14(8): 803-69. PMID 22828712
10. Lindenfeld J, Albert NM, Boehmer JP, et al. HFSA 2010 Comprehensive Heart Failure Practice Guideline. J Card Fail. Jun 2010; 16(6): e1-194. PMID 20610207
11. Savarese G, Trimarco B, Dellegrottaglie S, et al. Natriuretic peptide-guided therapy in chronic heart failure: a meta-analysis of 2,686 patients in 12 randomized trials. PLoS One. 2013; 8(3): e58287. PMID 23472172
12. Troughton RW, Frampton CM, Brunner-La Rocca HP, et al. Effect of B-type natriuretic peptide-guided treatment of chronic heart failure on total mortality and hospitalization: an individual patient meta-analysis. Eur Heart J. Jun 14 2014; 35(23): 1559-67. PMID 24603309
13. Bhardwaj A, Januzzi JL. ST2: a novel biomarker for heart failure. Expert Rev Mol Diagn. May 2010; 10(4): 459-64. PMID 20465500
14. Chowdhury P, Kehl D, Choudhary R, et al. The use of biomarkers in the patient with heart failure. Curr Cardiol Rep. Jun 2013; 15(6): 372. PMID 23644993
15. Ciccone MM, Cortese F, Gesualdo M, et al. A novel cardiac bio-marker: ST2: a review. Molecules. Dec 11 2013; 18(12): 15314-28. PMID 24335613
16. Daniels LB, Bayes-Genis A. Using ST2 in cardiovascular patients: a review. Future Cardiol. Jul 2014; 10(4): 525-39. PMID 25301315
17. Dieplinger B, Mueller T. Soluble ST2 in heart failure. Clin Chim Acta. Mar 30 2015; 443: 57-70. PMID 25269091
18. Mueller T, Dieplinger B. The Presage() ST2 Assay: analytical considerations and clinical applications for a high-sensitivity assay for measurement of soluble ST2. Expert Rev Mol Diagn. Jan 2013; 13(1): 13-30. PMID 23256700

19. Shah RV, Januzzi JL. ST2: a novel remodeling biomarker in acute and chronic heart failure. Curr Heart Fail Rep. Mar 2010; 7(1): 9-14. PMID 20425491
20. Xu D, Chan WL, Leung BP, et al. Selective expression of a stable cell surface molecule on type 2 but not type 1 helper T cells. J Exp Med. Mar 02 1998; 187(5): 787-94. PMID 9480988
21. Weinberg EO, Shimpo M, De Keulenaer GW, et al. Expression and regulation of ST2, an interleukin-1 receptor family member, in cardiomyocytes and myocardial infarction. Circulation. Dec 03 2002; 106(23): 2961-6. PMID 12460879
22. Stewart S, Winters GL, Fishbein MC, et al. Revision of the 1990 working formulation for the standardization of nomenclature in the diagnosis of heart rejection. J Heart Lung Transplant. Nov 2005; 24(11): 1710-20. PMID 16297770
23. Celec P, Vlkov B, Laukov L, et al. Cell-free DNA: the role in pathophysiology and as a biomarker in kidney diseases. Expert Rev Mol Med. Jan 18 2018; 20: e1. PMID 29343314
24. Natera. Prospera Heart Overview. 2024; https://www.natera.com/organ-health/prospera-heart-transplantation-assessment. Accessed October 22, 2025.
25. North PE, Ziegler E, Mahnke DK, et al. Cell-free DNA donor fraction analysis in pediatric and adult heart transplant patients by multiplexed allele-specific quantitative PCR: Validation of a rapid and highly sensitive clinical test for stratification of rejection probability. PLoS One. 2020; 15(1): e0227385. PMID 31929557
26. CareDx. Overview: AlloMap Testing: Answering Unmet Needs in Heart Transplant Surveillance. n.d.; https://caredx.com/products-and-services/transplant-services/heart/allomap/. Accessed October 22, 2025.
27. Duong Van Huyen JP, Tible M, Gay A, et al. MicroRNAs as non-invasive biomarkers of heart transplant rejection. Eur Heart J. Dec 01 2014; 35(45): 3194-202. PMID 25176944
28. Patel PC, Hill DA, Ayers CR, et al. High-sensitivity cardiac troponin I assay to screen for acute rejection in patients with heart transplant. Circ Heart Fail. May 2014; 7(3): 463-9. PMID 24733367
29. Organ Procurement and Transplantation Network. National Data. 2023; https://optn.transplant.hrsa.gov/data/view-data-reports/national-data/#. Accessed October 22, 2023.
30. Hariharan S, Israni AK, Danovitch G. Long-Term Survival after Kidney Transplantation. N Engl J Med. Aug 19 2021; 385(8): 729-743. PMID 34407344
31. Goldberg RJ, Weng FL, Kandula P. Acute and Chronic Allograft Dysfunction in Kidney Transplant Recipients. Med Clin North Am. May 2016; 100(3): 487-503. PMID 27095641
32. Ahmad I. Biopsy of the transplanted kidney. Semin Intervent Radiol. Dec 2004; 21(4): 275-81. PMID 21331139
33. Solez K, Colvin RB, Racusen LC, et al. Banff 07 classification of renal allograft pathology: updates and future directions. Am J Transplant. Apr 2008; 8(4): 753-60. PMID 18294345
34. Haas M. The Revised (2013) Banff Classification for Antibody-Mediated Rejection of Renal Allografts: Update, Difficulties, and Future Considerations. Am J Transplant. May 2016; 16(5): 1352-7. PMID 26696524
35. Natera. Prospera Transplant Assessment: Indications for Use. 2019; https://www.natera.com/wp-content/uploads/2020/12/PRO_OS_IndicationsForUse_20191011_Rev2_DWNLD.pdf. Accessed October 22, 2023.
36. Natera. Prospera Transplant Assessment: Clinician's Guide to Results. 2019; https://www.natera.com/wp-content/uploads/2020/12/PRO_OS_CGR_20190821_NAT-801994_DWNLD.pdf. Accessed October 22, 2023.
37. Parulekar AD, Kao CC. Detection, classification, and management of rejection after lung transplantation. J Thorac Dis. Sep 2019; 11(Suppl 14): S1732-S1739. PMID 31632750
38. Yusen RD, Edwards LB, Kucheryavaya AY, et al. The Registry of the International Society for Heart and Lung Transplantation: Thirty-second Official Adult Lung and Heart-Lung Transplantation Report--2015; Focus Theme: Early Graft Failure. J Heart Lung Transplant. Oct 2015; 34(10): 1264-77. PMID 26454740
39. Levine DJ, Glanville AR, Aboyoun C, et al. Antibody-mediated rejection of the lung: A consensus report of the International Society for Heart and Lung Transplantation. J Heart Lung Transplant. Apr 2016; 35(4): 397-406. PMID 27044531
40. CareDx. Allosure Lung. 2024; https://caredx.com/products-and-services/transplant-services/lung/ Accessed October 22, 2025
41. Natera. Prospera Lung Overview. 2024; https://www.natera.com/organ-health/prospera-lung-transplantation-assessment. Accessed October 22, 2025.
42. Cohn JN, Tognoni G. A randomized trial of the angiotensin-receptor blocker valsartan in chronic heart failure. N Engl J Med. Dec 06 2001; 345(23): 1667-75. PMID 11759645
43. O'Connor CM, Whellan DJ, Lee KL, et al. Efficacy and safety of exercise training in patients with chronic heart failure: HF-ACTION randomized controlled trial. JAMA. Apr 08 2009; 301(14): 1439-50. PMID 19351941
44. Kjekshus J, Apetrei E, Barrios V, et al. Rosuvastatin in older patients with systolic heart failure. N Engl J Med. Nov 29 2007; 357(22): 2248-61. PMID 17984166
45. Januzzi JL, Rehman SU, Mohammed AA, et al. Use of amino-terminal pro-B-type natriuretic peptide to guide outpatient therapy of patients with chronic left ventricular systolic dysfunction. J Am Coll Cardiol. Oct 25 2011; 58(18): 1881-9. PMID 22018299
46. Ky B, French B, McCloskey K, et al. High-sensitivity ST2 for prediction of adverse outcomes in chronic heart failure. Circ Heart Fail. Mar 2011; 4(2): 180-7. PMID 21178018
47. Bayes-Genis A, de Antonio M, Galn A, et al. Combined use of high-sensitivity ST2 and NTproBNP to improve the prediction of death in heart failure. Eur J Heart Fail. Jan 2012; 14(1): 32-8. PMID 22179033
48. Broch K, Ueland T, Nymo SH, et al. Soluble ST2 is associated with adverse outcome in patients with heart failure of ischaemic aetiology. Eur J Heart Fail. Mar 2012; 14(3): 268-77. PMID 22302661
49. Felker GM, Fiuzat M, Thompson V, et al. Soluble ST2 in ambulatory patients with heart failure: Association with functional capacity and long-term outcomes. Circ Heart Fail. Nov 2013; 6(6): 1172-9. PMID 24103327

50. Gaggin HK, Motiwala S, Bhardwaj A, et al. Soluble concentrations of the interleukin receptor family member ST2 and β-blocker therapy in chronic heart failure. Circ Heart Fail. Nov 2013; 6(6): 1206-13. PMID 24114865
51. Anand IS, Rector TS, Kuskowski M, et al. Prognostic value of soluble ST2 in the Valsartan Heart Failure Trial. Circ Heart Fail. May 2014; 7(3): 418-26. PMID 24622243
52. Zhang R, Zhang Y, An T, et al. Prognostic value of sST2 and galectin-3 for death relative to renal function in patients hospitalized for heart failure. Biomark Med. 2015; 9(5): 433-41. PMID 25985174
53. Dupuy AM, Curinier C, Kuster N, et al. Multi-Marker Strategy in Heart Failure: Combination of ST2 and CRP Predicts Poor Outcome. PLoS One. 2016; 11(6): e0157159. PMID 27311068
54. Aimo A, Vergaro G, Passino C, et al. Prognostic Value of Soluble Suppression of Tumorigenicity-2 in Chronic Heart Failure: A Meta-Analysis. JACC Heart Fail. Apr 2017; 5(4): 280-286. PMID 27816512
55. Januzzi JL, Horne BD, Moore SA, et al. Interleukin receptor family member ST2 concentrations in patients following heart transplantation. Biomarkers. May 2013; 18(3): 250-6. PMID 23557127
56. Pascual-Figal DA, Garrido IP, Blanco R, et al. Soluble ST2 is a marker for acute cardiac allograft rejection. Ann Thorac Surg. Dec 2011; 92(6): 2118-24. PMID 22035779
57. Phillips M, Boehmer JP, Cataneo RN, et al. Heart allograft rejection: detection with breath alkanes in low levels (the HARDBALL study). J Heart Lung Transplant. Jun 2004; 23(6): 701-8. PMID 15366430
58. Khush KK, Patel J, Pinney S, et al. Noninvasive detection of graft injury after heart transplant using donor-derived cell-free DNA: A prospective multicenter study. Am J Transplant. Oct 2019; 19(10): 2889-2899. PMID 30835940
59. Richmond ME, Deshpande SR, Zangwill SD, et al. Validation of donor fraction cell-free DNA with biopsy-proven cardiac allograft rejection in children and adults. J Thorac Cardiovasc Surg. Feb 2023; 165(2): 460-468.e2. PMID 35643770
60. Kim PJ, Olymbios M, Siu A, et al. A novel donor-derived cell-free DNA assay for the detection of acute rejection in heart transplantation. J Heart Lung Transplant. Jul 2022; 41(7): 919-927. PMID 35577713
61. Rodgers N, Gerding B, Cusi V, et al. Comparison of two donor-derived cell-free DNA tests and a blood gene-expression profile test in heart transplantation. Clin Transplant. Jun 2023; 37(6): e14984. PMID 37036133
62. Richmond ME, Zangwill SD, Kindel SJ, et al. Donor fraction cell-free DNA and rejection in adult and pediatric heart transplantation. J Heart Lung Transplant. May 2020; 39(5): 454-463. PMID 31983667
63. CareDx. HeartCare Interpretive Guide. April 2021; https://caredx.com/wp-content/uploads/2021/09/LK-10482-Heartcare-Interpretive-Guide.pdf. Accessed October 22, 2025.
64. Blue Cross Blue Shield Technology Evaluation Center (TEC). Gene expression profiling as a noninvasive method to monitor for cardiac allograft rejection. TEC Assessment Program. 2011;26(8).
65. Deng MC, Eisen HJ, Mehra MR, et al. Noninvasive discrimination of rejection in cardiac allograft recipients using gene expression profiling. Am J Transplant. Jan 2006; 6(1): 150-60. PMID 16433769
66. Crespo-Leiro MG, Stypmann J, Schulz U, et al. Clinical usefulness of gene-expression profile to rule out acute rejection after heart transplantation: CARGO II. Eur Heart J. Sep 01 2016; 37(33): 2591-601. PMID 26746629
67. Kanwar MK, Khush KK, Pinney S, et al. Impact of cytomegalovirus infection on gene expression profile in heart transplant recipients. J Heart Lung Transplant. Feb 2021; 40(2): 101-107. PMID 33341360
68. Gondi KT, Kao A, Linard J, et al. Single-center utilization of donor-derived cell-free DNA testing in the management of heart transplant patients. Clin Transplant. May 2021; 35(5): e14258. PMID 33606316
69. Khush K, Hall S, Kao A, et al. Surveillance with dual noninvasive testing for acute cellular rejection after heart transplantation: Outcomes from the Surveillance HeartCare Outcomes Registry. J Heart Lung Transplant. Sep 2024; 43(9): 1409-1421. PMID 38759766
70. Kim PJ, Alam AH, Teuteberg JJ, et al. Donor-Derived Cell-Free DNA in Antibody-Mediated Rejection: An Analysis of the Surveillance HeartCare Outcomes Registry. JACC Heart Fail. Oct 21 2025: 102716. PMID 41123513
71. Teuteberg J, Patel S, Baran DA, et al. Multimodal molecular testing provides prognostic value for heart transplant recipients. J Heart Lung Transplant. Nov 24 2025. PMID 41297735
72. Moeller CM, Valledor AF, Oren D, et al. Clinical Utility of Combined Donor-Derived Cell-Free DNA and Peripheral Gene-Expression-Profiling in Heart Transplant Recipients. Clin Transplant. Oct 2025; 39(10): e70340. PMID 41085484
73. Kobashigawa J, Patel J, Azarbal B, et al. Randomized pilot trial of gene expression profiling versus heart biopsy in the first year after heart transplant: early invasive monitoring attenuation through gene expression trial. Circ Heart Fail. May 2015; 8(3): 557-64. PMID 25697852
74. Pham MX, Deng MC, Kfoury AG, et al. Molecular testing for long-term rejection surveillance in heart transplant recipients: design of the Invasive Monitoring Attenuation Through Gene Expression (IMAGE) trial. J Heart Lung Transplant. Aug 2007; 26(8): 808-14. PMID 17692784
75. Pham MX, Teuteberg JJ, Kfoury AG, et al. Gene-expression profiling for rejection surveillance after cardiac transplantation. N Engl J Med. May 20 2010; 362(20): 1890-900. PMID 20413602
76. CareDx. Allosure Kidney. 2024; https://caredx.com/products-and-services/transplant-services/kidney/allosure/. Accessed October 22, 2025.
77. Pai A, Swan JT, Wojciechowski D, et al. Clinical Rationale for a Routine Testing Schedule Using Donor-Derived Cell-Free DNA After Kidney Transplantation. Ann Transplant. Jul 02 2021; 26: e932249. PMID 34210952
78. Wijtvliet VPWM, Plaeke P, Abrams S, et al. Donor-derived cell-free DNA as a biomarker for rejection after kidney transplantation: a systematic review and meta-analysis. Transpl Int. Dec 2020; 33(12): 1626-1642. PMID 32981117
79. Xiao H, Gao F, Pang Q, et al. Diagnostic Accuracy of Donor-derived Cell-free DNA in Renal-allograft Rejection: A Meta-analysis. Transplantation. Jun 01 2021; 105(6): 1303-1310. PMID 32890130
80. Xing Y, Guo Q, Wang C, et al. Donor-derived cell-free DNA as a diagnostic marker for kidney-allograft rejection: A systematic review and meta-analysis. Biomol Biomed. Feb 20 2024; 24(4): 731-740. PMID 38386614

81. Bloom RD, Bromberg JS, Poggio ED, et al. Cell-Free DNA and Active Rejection in Kidney Allografts. J Am Soc Nephrol. Jul 2017; 28(7): 2221-2232. PMID 28280140
82. Huang E, Sethi S, Peng A, et al. Early clinical experience using donor-derived cell-free DNA to detect rejection in kidney transplant recipients. Am J Transplant. Jun 2019; 19(6): 1663-1670. PMID 30725531
83. Stites E, Kumar D, Olaitan O, et al. High levels of dd-cfDNA identify patients with TCMR 1A and borderline allograft rejection at elevated risk of graft injury. Am J Transplant. Sep 2020; 20(9): 2491-2498. PMID 32056331
84. Sawinski DL, Mehta S, Alhamad T, et al. Association between dd-cfDNA levels, de novo donor specific antibodies, and eGFR decline: An analysis of the DART cohort. Clin Transplant. Sep 2021; 35(9): e14402. PMID 34184326
85. Jordan SC, Bunnapradist S, Bromberg JS, et al. Donor-derived Cell-free DNA Identifies Antibody-mediated Rejection in Donor Specific Antibody Positive Kidney Transplant Recipients. Transplant Direct. Sep 2018; 4(9): e379. PMID 30234148
86. Mayer KA, Doberer K, Tillgren A, et al. Diagnostic value of donor-derived cell-free DNA to predict antibody-mediated rejection in donor-specific antibody-positive renal allograft recipients. Transpl Int. Sep 2021; 34(9): 1689-1702. PMID 34448270
87. Puliyanda DP, Swinford R, Pizzo H, et al. Donor-derived cell-free DNA (dd-cfDNA) for detection of allograft rejection in pediatric kidney transplants. Pediatr Transplant. Mar 2021; 25(2): e13850. PMID 33217125
88. Dandamudi R, Gu H, Goss CW, et al. Longitudinal Evaluation of Donor-Derived Cellfree DNA in Pediatric Kidney Transplantation. Clin J Am Soc Nephrol. Nov 2022; 17(11): 1646-1655. PMID 36302566
89. Bu L, Gupta G, Pai A, et al. Clinical outcomes from the Assessing Donor-derived cell-free DNA Monitoring Insights of kidney Allografts with Longitudinal surveillance (ADMIRAL) study. Kidney Int. Apr 2022; 101(4): 793-803. PMID 34953773
90. Moein M, Settineri JP, Suleiman H, et al. Application of Combined Donor-Derived Cell-Free DNA and Transcriptome in Diagnosis of Kidney Transplant Rejection. Transplant Proc. 2024; 56(6): 1259-1263. PMID 39019762
91. Aubert O, Ursule-Dufait C, Brousse R, et al. Cell-free DNA for the detection of kidney allograft rejection. Nat Med. Aug 2024; 30(8): 2320-2327. PMID 38824959
92. Aravamudhan A, Krug K, Stoffel M, et al. Evaluating the Diagnostic Utility of dd-cfDNA in Renal Allograft Surveillance: A Single-Center Perspective. Genes (Basel). Jun 21 2025; 16(7). PMID 40725381
93. Bromberg JS, Brennan DC, Taber DJ, et al. Donor-derived cell-free DNA significantly improves rejection yield in kidney transplant biopsies. Am J Transplant. Dec 2025; 25(12): 2529-2542. PMID 40780562
94. Hogan J, George R, Hayes C, et al. Donor-derived Cell-free DNA as a Noninvasive Biomarker of Kidney Allograft Rejection in Pediatric Kidney Transplantation. Transplantation. Sep 01 2025; 109(9): 1520-1525. PMID 40200407
95. Sigdel TK, Archila FA, Constantin T, et al. Optimizing Detection of Kidney Transplant Injury by Assessment of Donor-Derived Cell-Free DNA via Massively Multiplex PCR. J Clin Med. Dec 23 2018; 8(1). PMID 30583588
96. Bunnapradist S, Homkrailas P, Ahmed E, et al. Using both the Fraction and Quantity of Donor-Derived Cell-Free DNA to Detect Kidney Allograft Rejection. J Am Soc Nephrol. Oct 2021; 32(10): 2439-2441. PMID 34162734
97. Halloran PF, Reeve J, Madill-Thomsen KS, et al. Combining Donor-derived Cell-free DNA Fraction and Quantity to Detect Kidney Transplant Rejection Using Molecular Diagnoses and Histology as Confirmation. Transplantation. Dec 01 2022; 106(12): 2435-2442. PMID 35765145
98. Halloran PF, Reeve J, Madill-Thomsen KS, et al. The Trifecta Study: Comparing Plasma Levels of Donor-derived Cell-Free DNA with the Molecular Phenotype of Kidney Transplant Biopsies. J Am Soc Nephrol. Feb 2022; 33(2): 387-400. PMID 35058354
99. Bromberg JS, Bunnapradist S, Samaniego-Picota M, et al. Elevation of Donor-derived Cell-free DNA Before Biopsy-proven Rejection in Kidney Transplant. Transplantation. Sep 01 2024; 108(9): 1994-2004. PMID 38595232
100. Sayah D, Weigt SS, Ramsey A, et al. Plasma Donor-derived Cell-free DNA Levels Are Increased During Acute Cellular Rejection After Lung Transplant: Pilot Data. Transplant Direct. Oct 2020; 6(10): e608. PMID 33062841
101. Khush KK, De Vlaminck I, Luikart H, et al. Donor-derived, cell-free DNA levels by next-generation targeted sequencing are elevated in allograft rejection after lung transplantation. ERJ Open Res. Jan 2021; 7(1). PMID 33532456
102. Keller M, Sun J, Mutebi C, et al. Donor-derived cell-free DNA as a composite marker of acute lung allograft dysfunction in clinical care. J Heart Lung Transplant. Apr 2022; 41(4): 458-466. PMID 35063338
103. Rosenheck JP, Ross DJ, Botros M, et al. Clinical Validation of a Plasma Donor-derived Cell-free DNA Assay to Detect Allograft Rejection and Injury in Lung Transplant. Transplant Direct. Apr 2022; 8(4): e1317. PMID 35372675
104. Arunachalam A, Anjum F, Rosenheck J, et al. Donor-derived cell-free DNA is a valuable monitoring tool after single lung transplantation: Multicenter analysis. JHLT Open. November 2024; Volume 6
105. Yancy CW, Jessup M, Bozkurt B, et al. 2017 ACC/AHA/HFSA Focused Update of the 2013 ACCF/AHA Guideline for the Management of Heart Failure: A Report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines and the Heart Failure Society of America. J Card Fail. Aug 2017; 23(8): 628-651. PMID 28461259
106. American Society of Transplant Surgeons (ASTS). ASTS Statement on donor-derived cell-free DNA (dd-cf-DNA). 2023. Accessed on October 4, 2025. Available at: https://asts.org/docs/default-source/position-statements/dd-cfdna-position-statement.pdf?sfvrsn=143d4bd3
107. Nikolova A, Agbor-Enoh S, Bos S, et al. European Society for Organ Transplantation (ESOT) Consensus Statement on the Use of Non-invasive Biomarkers for Cardiothoracic Transplant Rejection Surveillance. Transpl Int. 2024; 37: 12445. PMID 38962472
108. Velleca A, Shullo MA, Dhital K, et al. The International Society for Heart and Lung Transplantation (ISHLT) guidelines for the care of heart transplant recipients. J Heart Lung Transplant. May 2023; 42(5): e1-e141. PMID 37080658
109. Kobashigawa J, Zuckermann A, Zeevi A, et al. Summary of the International Society for Heart and Lung Transplantation consensus conference on emerging understanding of antibodies and antibody-mediated rejection in heart transplantation. J Heart Lung Transplant. Aug 2025; 44(8): e1-e20. PMID 40498433
110. Kasiske BL, Zeier MG, Chapman JR, et al. KDIGO clinical practice guideline for the care of kidney transplant recipients: a summary. Kidney Int. Feb 2010; 77(4): 299-311. PMID 19847156

111. Centers for Medicare & Medicaid Services (CMS). National Coverage Determination for HEARTSBREATH Test for Heart Transplant Rejection (260.10). 2008; https://www.cms.gov/medicare-coverage-database/details/ncd-details.aspx?NCDId=325. Accessed October 22, 2025.

POLICY HISTORY - THIS POLICY WAS APPROVED BY THE FEP® PHARMACY AND MEDICAL POLICY COMMITTEE ACCORDING TO THE HISTORY BELOW:

Date	Action	Description
June 2012	New policy	
June 2013	Replace policy	Policy updated with literature review through March 5, 2015. References 2-3 and 12 added. Policy statements unchanged.
June 2014	Replace policy	Policy updated with literature review through March 4, 2014; reference 9 added. Policy statements unchanged.
June 2015	Replace policy	Policy updated with literature review, References removed and renumbered. Policy statements unchanged.
December 2017	Replace policy	Policy updated with literature review through August 28, 2017; no references added; reference 2 updated. In first policy statement, "grade 3€š changed to "grade 2R/grade 3€š due to updated ISHLT rejection grades and brand name of test removed; intent of statements unchanged. Policy statement corrected from "not medically necessary€š to "investigational€š.
December 2018	Replace policy	Policy updated with literature review through August 22, 2018; references 5-9, 18, 20, and 22 added. Policy statement added that "The use of peripheral blood measurement of donor-derived cell-free DNA in the management of patients after renal transplantation, including but not limited to the detection of acute renal transplant rejection or renal transplant graft dysfunction, is considered investigational.€š Title expanded to include kidney transplant rejection.
December 2019	Replace policy	Policy updated with literature review through August 5, 2019; no references added. Policy statements unchanged.
December 2020	Replace Policy	Policy updated with literature review through August 25, 2020. references added. Content from policy 2.04.130 (Molecular Testing for Chronic Heart Failure and Heart Transplant) was merged into this policy and the title was changed to "Laboratory Tests Post Transplant and for Heart Failure".
December 2021	Replace Policy	Policy updated with literature review through October 20, 2021; references added. New investigational policy statement regarding dd-cfDNA testing in lung transplantation was added. Investigational policy statement for GEP testing (ie, AlloMap) in heart transplantation was updated to include use alone or in combination with dd-cfDNA testing (ie, HeartCare).
March 2022	Replace policy	Administrative formatting.
December 2022	Replace policy	Policy updated with literature review through August 24, 2022; references added. New investigational policy statement regarding dd-cfDNA testing in heart transplantation was added. Other changes to policy statements reflect minor editorial refinements; intent unchanged.
December 2023	Replace policy	Policy updated with literature review through August 21, 2023; references added. Policy statements unchanged.
July 2024	Replace policy	Off cycle FEP Policy review to update policy statement for Allomap laboratory test post transplant and heart failure.
March 2025	Replace policy	Policy updated with literature review through November 22, 2024; references added. Policy statements unchanged.
March 2026	Replace policy	Policy updated with literature review through November 17, 2025; references added. Policy statements unchanged.