

MOLECULAR DIAGNOSTIC TESTING FOR ACUTE REJECTION IN KIDNEY OR LIVER ALLOGRAFTS

List has been updated

See publications in italicized text

Selected Clinical Literature List

Below is the list of selected literature (organized by organ and test) for the Kidney or Liver CAC discussion. For select tests, there is a paucity of evidence. Therefore, additional searches on manufacturer's websites were conducted to ensure that all relevant information was included. Results from these searches and rationale for exclusion are included in the Appendices.

Transplant Organ: Kidney

AlloMap

1. *Cheung R, Xu H, Jin X, et al. Validation of a gene expression signature to measure immune quiescence in kidney transplant recipients in the CLIA setting. Biomark Med. 2022 Jun;16(8):647-661.*
2. *Akalin E, Weir MR, Bunnapradist S, et al. Clinical Validation of an Immune Quiescence Gene Expression Signature in Kidney Transplantation. Kidney360. 2021 Sep 28;2(12):1998-2009.*

AlloSure

1. Bromberg JS, Brennan DC, et. al. Biological Variation of Donor-Derived Cell-Free DNA in Renal Transplant Recipients: Clinical Implications. Journal of Applied Laboratory Medicine (2017, September); 2:02, 1-13.
2. Bloom RD, Bromberg JS, Poggio ED, et al.; Circulating Donor-Derived Cell-Free DNA in Blood for Diagnosing Active Rejection in Kidney Transplant Recipients (DART) Study Investigators. Cell-Free DNA and Active Rejection in Kidney Allografts. J Am Soc Nephrol. 2017 Jul;28(7):2221-2232.
3. 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. 2022 Apr;101(4):793-803.
4. 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. 2018 Aug 20;4(9):e379.

5. 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.* 2020 Sep;20(9):2491-2498.

AlloSure and Prospera

1. Melancon JK, Khalil A, Lerman MJ. Donor-Derived Cell Free DNA: Is It All the Same? *Kidney360.* 2020 Jun 19;1(10):1118-1123.

Prospera

1. 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.* 2018 Dec 23;8(1):19.
2. Halloran PF, Reeve J, Madill-Thomsen KS, et al.; Trifecta Investigators*. Combining Donor-derived Cell-free DNA Fraction and Quantity to Detect Kidney Transplant Rejection Using Molecular Diagnoses and Histology as Confirmation. *Transplantation.* 2022 Jun 29.

Viracor TRAC

The literature review returned no publications that assessed the clinical validity or utility of Viracor TRAC in kidney transplant recipients.

kSORT

1. Roedder S, Sigdel T, Salomonis N, et al. The kSORT assay to detect renal transplant patients at high risk for acute rejection: results of the multicenter AART study. *PLoS Med.* 2014 Nov 11;11(11):e1001759.
2. Crespo E, Roedder S, Sigdel T, et al. Molecular and Functional Noninvasive Immune Monitoring in the ESCAPE Study for Prediction of Subclinical Renal Allograft Rejection. *Transplantation.* 2017 Jun;101(6):1400-1409.
3. Van Loon E, Giral M, Anglicheau D, et al. Diagnostic performance of kSORT, a blood-based mRNA assay for noninvasive detection of rejection after kidney transplantation: A retrospective multicenter cohort study. *Am J Transplant.* 2021 Feb;21(2):740-750.

TruGraf

1. Ang A, Schieve C, Rose S, et al. *Avoiding surveillance biopsy: Use of a noninvasive biomarker assay in a real-life scenario.* *Clin Transplant.* 2021 Jan;35(1):e14145.
2. Friedewald JJ, Kurian SM, Heilman RL, et al; Clinical Trials in Organ Transplantation 08 (CTOT-08). Development and clinical validity of a novel blood-based molecular biomarker for subclinical acute rejection following kidney transplant. *Am J Transplant.* 2019 Jan;19(1):98-109.
3. First MR, Peddi VR, Mannon R, et al. Investigator Assessment of the Utility of the TruGraf Molecular Diagnostic Test in Clinical Practice. *Transplant Proc.* 2019 Apr;51(3):729-733.

4. Marsh CL, Kurian SM, Rice JC, et al. Application of TruGraf v1: A Novel Molecular Biomarker for Managing Kidney Transplant Recipients with Stable Renal Function. *Transplant Proc.* 2019 Apr;51(3):722-728.

OmniGraf

1. Park S, Guo K, Heilman RL, et al. Combining Blood Gene Expression and Cellfree DNA to Diagnose Subclinical Rejection in Kidney Transplant Recipients. *CJASN* Oct 2021, 16 (10) 1539-1551.

QSant

1. Nolan N, Valdivieso K, Mani R, et al. Clinical and Analytical Validation of a Novel Urine-Based Test for the Detection of Allograft Rejection in Renal Transplant Patients. *J Clin Med.* 2020 Jul 22;9(8):2325.
2. Yang JYC, Sarwal RD, Sigdel TK, et al. A urine score for noninvasive accurate diagnosis and prediction of kidney transplant rejection. *Sci Transl Med.* 2020 Mar 18;12(535):eaba2501.

Transplant Organ: Liver

TruGraf

1. *Levitsky J, Kandpal M, Guo K, et al. Donor-derived cell-free DNA levels predict graft injury in liver transplant recipients. Am J Transplant. 2022a Feb;22(2):532-540.*
2. *Levitsky J, Kandpal M, Guo K, et al. Prediction of Liver Transplant Rejection with a Biologically Relevant Gene Expression Signature. Transplantation. 2022b May 1;106(5):1004-1011.*
3. *Levitsky J, Asrani SK, Schiano T, et al; Clinical Trials in Organ Transplantation - 14 Consortium. Discovery and validation of a novel blood-based molecular biomarker of rejection following liver transplantation. Am J Transplant. 2020 Aug;20(8):2173-2183.*

Viracor TRAC – Liver

The literature review returned no publications that reported the performance of Viracor TRAC in liver transplant recipients.

Appendix A

Organ Kidney

Prospera

Due to the paucity of literature on the use of Prospera in kidney transplant recipients, an additional search of Natera's website was conducted. [Natera's Prospera Kidney webpage](#) cites the following additional references:

Rationale for exclusion	Reference
This was an analytical validation study.	Altug Y, Liang N, Ram R, et al. Analytical validation of a single-nucleotide polymorphism-based donor-derived cell-free DNA assay for detecting rejection in kidney transplant patients. <i>Transplantation</i> . 2019;103(12):2657-2665.
This study evaluated AlloSure rather than Prospera.	Bloom RD, Bromberg JS, Poggio ED, et al.; Circulating Donor-Derived Cell-Free DNA in Blood for Diagnosing Active Rejection in Kidney Transplant Recipients (DART) Study Investigators. Cell-Free DNA and Active Rejection in Kidney Allografts. <i>J Am Soc Nephrol</i> . 2017 Jul;28(7):2221-2232.
This was an analytical validation study for the detection of rejection in heart and kidney recipients that was conducted using AlloSure rather than Prospera Heart.	Grskovic M, Hiller DJ, Eubank LA, et al. Validation of a clinical-grade assay to measure donor-derived cell-free DNA in solid organ transplant recipients. <i>J Mol Diagn</i> . 2016;18(6):890-902.
This is a reference for organ transplant data cited.	Data from the U.S. Department of Health & Human Services: Health Resources and Services Administration. Scientific Registry of Transplant Recipients (SRTR): Organ Procurement and Transplantation Network (OPTN)/SRTR.
This is a reference for organ transplant data cited.	Kidney Disease Statistics for the United States. National Institute of Diabetes and Digestive and Kidney Diseases. https://www.niddk.nih.gov/health-information/health-statistics/kidney-disease . Published Dec. 1, 2016.
This is a narrative review.	Stegall MD, Gaston RS, Cosio FG, Matas A. Through a glass darkly: seeking clarity in preventing late kidney transplant failure. <i>J Am Soc Nephrol</i> . 2015 Jan;26(1):20-9.
An evaluation of short-term and long-term renal allograft survival in the US. No assay evaluation was included.	Lamb KE, Lodhi S, Meier-Kriesche HU. Long-term renal allograft survival in the United States: a critical reappraisal. <i>Am J Transplant</i> . 2011 Mar;11(3):450-62.

Rationale for exclusion	Reference
This publication is labeled “Research Letters”.	Bunnapradist S, Homkrailas P, Ahmed E, Fehringer G, Billings PR, Tabriziani H. Using both the Fraction and Quantity of Donor-Derived Cell-Free DNA to Detect Kidney Allograft Rejection. J Am Soc Nephrol. 2021 Oct;32(10):2439-2441.

Viracor TRAC

The literature review returned no publications that reported the performance of Viracor TRAC in kidney transplant recipients. An additional search of Eurofins’ website was performed.

[Eurofins’ Viracor TRAC Kidney dd-cfDNA webpage](#) cites the following references:

Rationale for exclusion	Reference
This is an abstract, no full text publication was found. In the Establishment of Reference Range section, cutoff values and performance indices are displayed.	Kleiboeker S, Grantham J, Mickey K, Cowden S, Bixler E, Sinha R, Altrich M. Clinical Performance of a Donor-Derived Cell-Free DNA Assay for Detection of Rejection in Kidney Transplant Recipients [abstract]. Am J Transplant. 2020; 20 (suppl 3). Clinical Performance of a Donor-Derived Cell-Free DNA Assay for Detection of Rejection in Kidney Transplant Recipients - ATC Abstracts (atcmeetingabstracts.com) . Accessed 10/07/22.
This study evaluated AlloSure (not Viracor TRAC).	Bromberg JS, Brennan DC, et. al. Biological Variation of Donor-Derived Cell-Free DNA in Renal Transplant Recipients: Clinical Implications. Journal of Applied Laboratory Medicine (2017, September); 2:02, 1-13.
This study is a narrative review. It is labeled as a “Minireview” and discusses dd cf DNA measurement methods and several observational studies.	Gielis EM, Ledeganck KJ, De Winter BY, et. al. Cell-Free DNA: An Upcoming Biomarker in Transplantation. American Journal of Transplantation (2015); 15: 2541-2551.
This study evaluated lung (not kidney) transplant recipients and measured dd cf DNA by shotgun sequencing. Eurofins states that Viracor TRAC Kidney “analyzes NGS and genome-wide recipient genotype data to determine the percentage of dd cfDNA present...”, which is a different measurement method.	De Vlaminck I, Martin L, Kertesz M, et. al. Noninvasive monitoring of infection and rejection after lung transplantation. Proceedings of the National Academy of Sciences (2015, October 27); 112:43, 13336-13341.

Rationale for exclusion	Reference
This was an analytical validation study for the detection of rejection in heart and kidney recipients and conducted using AlloSure rather than Viracor TRAC Heart.	Grskovic M, Hiller DJ, Eubank LA, et. al. Validation of a Clinical-Grade Assay to Measure Donor-Derived Cell-Free DNA in Solid Organ Transplant Recipients. The Journal of Molecular Diagnostics (2016, November); 18:6, 890-902.

kSORT

An additional search of [Immuncor's kSORT webpage](#) was conducted. The following additional publications were cited:

Rationale for exclusion	Reference
This is an abstract.	Sarwal, Minnie; Vincenti, Flavio; Schroeder, Andrew; Hseish, Szu-Chuan; Liberto, Juliane; Towfighi, Parhom; Koh, Crystal; Sigdel, Tara. The Results of the PRISM (Prediction of Rejection In Sensitized patient blood saMples) Trial with a Novel Bioassay. Transplantation: July 2018 - Volume 102 - Issue - p S128.
This is an abstract.	Ekberg J, Jespersen B, Skov K, Sarwal M, Sigdel T, Hsieh S, Lindner P. A Non-Invasive Blood Transcriptional Assay, Ksort, Monitors Alloimmune Response in the Sailor Randomized Multicenter Trial. Am J Transplant. 2016;16 (suppl 3).

TruGraf

An additional search of [Eurofins TruGraf webpage](#) was conducted. The following additional publications were cited:

Rationale for exclusion	Reference
Unclear as to what this this is.	Primary research study by CLINICevAL Solutions, LLC, 2019
This was the analytical validation of TruGraf and included in the list above.	First MR, Pierry D, McNulty M, Kurian SM, Rose S, Whisenant T, et al. Analytical performance validation of a molecular diagnostic signature in kidney transplant recipients. J Transplant Technol Res 2017;7:176

Rationale for exclusion	Reference
A Local Coverage Determination.	Local Coverage Determination MoIDX: TruGraf Blood Gene Expression Test (DL38039). Effective Date of Reimbursement: 25, November 2019 from cms.gov.

OmniGraf

An additional search of Eurofins' and Immucor's websites was conducted. No additional evidence was found that assessed the use of OmniGraf to detect allograft rejection.

QSant

An additional search of [NephroSant's website](#) was conducted. The following publications were cited:

Rationale for exclusion	Reference
The aim of this study was to identify biomarkers to develop the Kidney Injury Test (KIT) in patients who presented with CKD. It did not assess the clinical validity or utility of QSant.	Watson D, Yang JYC, Sarwal RD, et al. A Novel Multi-Biomarker Assay for Non-Invasive Quantitative Monitoring of Kidney Injury. J Clin Med. 2019 Apr 12;8(4):499.
The aim of this study was to investigate the clinical utility of the KIT assay in detection of IgAN and predicting progression of renal damage over time. It did not assess the clinical validity or utility of QSant.	Yang JYC, Sarwal RD, Fervenza FC, et al. Noninvasive Urinary Monitoring of Progression in IgA Nephropathy. Int J Mol Sci. 2019 Sep 10;20(18):4463.

Appendix B

Organ Liver

Viracor TRAC

An additional search of the [Eurofins Viracor TRAC Liver dd-cfDNA webpage](#) was conducted. The following publications were cited:

Rationale for exclusion	Reference
<p>This study evaluated AlloSure (not Viracor TRAC) in kidney (not liver) transplant recipients.</p>	<p>Bromberg JS, Brennan DC, et. al. Biological Variation of Donor-Derived Cell-Free DNA in Renal Transplant Recipients: Clinical Implications. Journal of Applied Laboratory Medicine (2017, September); 2:02, 1-13.</p>
<p>This study is a narrative review. It is labeled as a “Minireview” and discusses dd cf DNA measurement methods and several observational studies.</p>	<p>Gielis EM, Ledeganck KJ, De Winter BY, et. al. Cell-Free DNA: An Upcoming Biomarker in Transplantation. American Journal of Transplantation (2015); 15: 2541-2551.</p>
<p>This study evaluated lung (not liver) transplant recipients and measured dd cf DNA by shotgun sequencing. Eurofins states that Viracor TRAC Liver “...is designed to be a noninvasive liquid biopsy to monitor the percent of dd-cfDNA by next generation sequencing (NGS) in the transplant recipient’s plasma post- transplant.”, which is a different measurement method.</p>	<p>De Vlaminck I, Martin L, Kertesz M, et. al. Noninvasive monitoring of infection and rejection after lung transplantation. Proceedings of the National Academy of Sciences (2015, October 27); 112:43, 13336-13341.</p>
<p>This was an analytical validation study for the detection of rejection in heart and kidney (not liver) recipients and conducted using AlloSure rather than Viracor TRAC Liver.</p>	<p>Grskovic M, Hiller DJ, Eubank LA, et. al. Validation of a Clinical-Grade Assay to Measure Donor-Derived Cell-Free DNA in Solid Organ Transplant Recipients. The Journal of Molecular Diagnostics (2016, November); 18:6, 890-902.</p>