**Policy Statement**

HIV tropism testing (see Policy Guidelines section for testing methods) may be considered **medically necessary** for selecting patients for treatment with HIV coreceptor antagonists, such as maraviroc, when there is an immediate plan to prescribe a coreceptor antagonist.

HIV tropism testing without immediate plans to prescribe HIV coreceptor antagonists such as maraviroc is considered **not medically necessary**.

Repeat HIV tropism testing during coreceptor antagonist treatment or after failure with coreceptor antagonists is considered **investigational**.

HIV tropism testing to predict disease progression (irrespective of coreceptor antagonist treatment) is considered **investigational**.

**Policy Guidelines**

**Coding**

There are no specific CPT codes for human immunodeficiency virus (HIV) tropism testing. In coding advice disseminated by NHIC Corp., a local Medicare carrier, it was suggested that the following CPT code be used for this test along with “Trofile test for maraviroc” in the claim comments field:

- **87999**: Unlisted microbiology procedure

**Testing**

Testing should be conducted immediately before intended prescribed use of maraviroc to obtain the most accurate prediction of tropism at the start of treatment.

Either phenotypic or V3 population genotypic testing may be used to determine HIV tropism; both are not necessary.

V3 population genotypic testing may be conducted by either standard V3 sequencing via Sanger methods (amplification and population sequence analysis of patient-derived V3 region) or V3 deep sequencing methods (synonyms: ultra-deep sequencing; pyrosequencing; next-generation sequencing). In the United States, the only currently commercially available plasma HIV DNA coreceptor genotypic test (requires HIV viral load of ≥1000 copies/mL) includes step-wise testing, with an initial standard sequencing with reflex to V3 deep sequencing if standard sequencing detects only CCR5-tropic virus.

The FDA has not regulated the Trofile test because it is a laboratory-developed test (LDT) conducted only at Monogram Biosciences’ Clinical Laboratory Improvement Amendments (CLIA)-licensed laboratory, and it does not meet the definition of an in vitro diagnostic multivariate index assay, the only type of LDT that the FDA is currently regulating. Laboratories performing LDTs not regulated by the FDA must only be certified for high-complexity testing under the CLIA of 1988.

**Genetic Counseling**

Genetic counseling is primarily aimed at patients who are at risk for inherited disorders, and experts recommend formal genetic counseling in most cases when genetic testing for an inherited condition is considered. The interpretation of the results of genetic tests and the understanding of risk factors can be very difficult and complex.
Therefore, genetic counseling will assist individuals in understanding the possible benefits and harms of genetic testing, including the possible impact of the information on the individual’s family. Genetic counseling may alter the utilization of genetic testing substantially and may reduce inappropriate testing. Genetic counseling should be performed by an individual with experience and expertise in genetic medicine and genetic testing methods.

### Description

HIV tropism testing can determine the predominant coreceptor protein used by HIV to infect target cells. Tropism testing can help select patients for treatment with HIV coreceptor antagonists (e.g., maraviroc), which block specific coreceptor proteins.

### Related Policies

- N/A

### Benefit Application

Benefit determinations should be based in all cases on the applicable contract language. To the extent there are any conflicts between these guidelines and the contract language, the contract language will control. Please refer to the member’s contract benefits in effect at the time of service to determine coverage or non-coverage of these services as it applies to an individual member.

Some state or federal mandates [e.g., Federal Employee Program (FEP)] prohibits plans from denying Food and Drug Administration (FDA)-approved technologies as investigational. In these instances, plans may have to consider the coverage eligibility of FDA-approved technologies on the basis of medical necessity alone.

### Regulatory Status

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. HIV tropism tests are available under the auspices of the Clinical Laboratory Improvement Amendments. Laboratories that offer laboratory-developed tests must be licensed by the Clinical Laboratory Improvement Amendments for high-complexity testing. To date, the FDA has chosen not to require any regulatory review of this test.

The FDA-approved full prescribing information for maraviroc (Selzentry™, Pfizer) states that: “Tropism testing must be conducted with a highly sensitive and specific tropism assay that has demonstrated the ability to identify patients appropriate for [maraviroc] use.”

### Rationale

#### Background

**HIV**

HIV-1, which causes AIDS, uses coreceptor proteins (either CCR5 or CXCR4) on the surface of target cells to enter and infect the cells. The most commonly transmitted strains of HIV-1 bind to CCR5 and are said to have “tropism” for CCR5-expressing cells. Dual or mixed (D/M) tropic viruses can bind to either receptor type. It is estimated that around 85% of treatment-naive patients harbor CCR5-tropic virus only, around 15% harbor D/M virus, and less than 1% are infected with CXCR4-tropic virus alone. CXCR4-tropic virus is associated with immunosuppression and later stages of disease. Coreceptor antagonists have been designed to interfere with the interaction between HIV-1 and its coreceptors.
HIV Coreceptor Antagonists

Maraviroc (Selzentry) was the first coreceptor antagonist to be approved by the U.S. Food and Drug Administration (FDA). Maraviroc is a selective, slowly reversible, small-molecule antagonist of the interaction between human cell surface CCR5 and HIV-1 gp120, also necessary for HIV-1 cell infection. Blocking this interaction prevents CCR5-tropic HIV-1 entry into cells. However, CXCR4-tropic HIV-1 entry is not prevented. According to the drug’s original label, maraviroc, in combination with other antiretroviral agents, is indicated for adults who are infected with only CCR5-tropic detectable HIV-1, who have evidence of viral replication and HIV-1 strains resistant to multiple antiretroviral agents.1

The currently approved maraviroc label indicates that maraviroc is indicated for combination antiretroviral treatment for adults infected with only CCR5-tropic HIV-1, without discussion of the presence of viral replication.2 The FDA-approved full prescribing information for the drug states: “Tropism testing must be conducted on a current sample with a highly sensitive tropism assay that has demonstrated the ability to identify patients appropriate for use of SELZENTRY.” This is because efficacy was not demonstrated in a phase 2 study of maraviroc in patients with D/M or CXCR4-tropic HIV-1. Due to potential adverse events (hepatic and cardiac toxicity), maraviroc should only be used in indicated patients.

Other HIV coreceptor antagonists are in the drug development pipeline. Cenicriviroc (Tobira Therapeutics) is a small-molecule antagonist of both CCR5 and CCR2, a receptor involved in a number of inflammatory diseases, that is currently being investigated for treatment of CCR5-tropic HIV.3 In January 2015, cenicriviroc was granted fast track designation by the FDA for the treatment of nonalcoholic steatohepatitis in patients with liver fibrosis, but the drug does not yet have the FDA approval.

HIV Tropism Testing

HIV tropism testing is available by either phenotypic or genotypic methods. Tropism testing with a phenotypic assay, a cellular-based assay that functionally determines tropism, is available with the enhanced sensitivity Trofile® assay (ESTA; Monogram Biosciences, South San Francisco, CA). This phenotypic assay uses virus stocks pseudotyped with envelope sequences derived from patient plasma to infect cell lines engineered to express CCR5 or CXCR4 HIV-2 coreceptors. Genotypic tropism testing is based on sequencing the third variable (V3) loop of the HIV glycoprotein 120 gene; this is because the V3 loop interacts with the HIV co-receptor, and variants in V3 are associated with measurable changes in HIV tropism. Tropism assignment is derived from the sequence data using a bioinformatic algorithm such as geno2pheno. In the United States, Quest Diagnostics (Madison, NJ) offers the only commercially available genotypic HIV coreceptor tropism assay, which uses triplicate population sequencing with reflex to ultra-deep sequencing if only CCR5-tropic virus is detected. Quest Diagnostics also offers a proviral DNA tropism test (Trofile® DNA), which sequences the tropism of HIV-1 DNA that has integrated into the host genome of infected T lymphocytes via triplicate population sequencing, without the use of ultra-deep sequencing.

Literature Review

Evidence reviews assess whether a medical test is clinically useful. A useful test provides information to make a clinical management decision that improves the net health outcome. That is, the balance of benefits and harms is better when the test is used to manage the condition than when another test or no test is used to manage the condition.

The first step in assessing a medical test is to formulate the clinical context and purpose of the test. The test must be technically reliable, clinically valid, and clinically useful for that purpose. The following is a summary of the key literature.
HIV Tropism Testing to Identify Candidates for HIV Coreceptor Antagonist Therapy

Clinical Context and Test Purpose
The purpose of HIV tropism testing in patients who have HIV infection is to inform a decision whether the patient might be a candidate for treatment with HIV coreceptor antagonist therapy.

The question addressed in this evidence review is: Does assessment of HIV tropism, to identify HIV-infected patients who are candidates for HIV coreceptor therapy, result in an improved health outcome compared with HIV coreceptor therapy without HIV tropism testing?

The following PICOTS were used to select literature to inform this review.

**Patients**
The relevant populations of interest are treatment-naive and treatment-experienced HIV-infected patients.

**Interventions**
The interventions of interest are HIV tropism testing using the Trofile assay, the enhanced sensitivity Trofile assay (ESTA), V3 sequencing, or V3 deep sequencing.

**Comparators**
The comparator of interest is no HIV tropism testing.

**Outcomes**
The potential beneficial outcomes of primary interest would be identification of HIV-infected patients who might benefit from treatment with HIV coreceptor antagonist therapy.

The potential harmful outcomes are those resulting from a false test result. False-positive or false-negative test results can lead to the initiation of unnecessary treatment and adverse events from that treatment or undertreatment.

**Timing**
HIV tropism testing is conducted before starting HIV coreceptor antagonist therapy.

**Setting**
Ordering and interpreting HIV tropism testing should be done by physicians specializing in infectious diseases. Most patients are likely to be tested in an outpatient setting. Referral for genetic counseling is important for the explanation of genetic disease, heritability, genetic risk, test performance, and possible outcomes.

**Technically Reliable**
The technical reliability of different HIV tropism testing and comparison in performance of these testing techniques are discussed in this section.

**Tropism Testing Using the Trofile Assay or Enhanced Sensitivity Trofile Assay**
For the clinical studies of patients with treatment failure, Whitcomb et al (2007) determined tropism at enrollment and again at baseline using the original phenotypic Trofile assay for 2560 potential enrollees; 56% of the enrollees were CCR5-tropic only and were eligible for the clinical trials. Most other patients had dual or mixed (D/M) HIV infection; CXCR4-infection alone is rare.

Of patients enrolled, 90% had CCR5-tropic virus at baseline, 4% had D/M-tropic virus, and 5% had nontypable virus infection. The original phenotypic Trofile assay had a turnaround time of 14 to 18 days, failed to work in 3% to 7% of patients, and required at least 1000 copies/mL of HIV RNA. The assay was 100% effective in detecting model CXCR4-tropic or D/M HIV present in a 10% mixture, and 83% effective at a 5% mixture. Validation studies also indicated 100% accuracy of results for 38 samples with known tropism, and 100% reproducibility including repeat assays.
using multiple operators, instrumentation, reagent lots, and conducted over a 14-day period. No false-positive results were obtained on samples that were HIV-negative but positive for either hepatitis B or C virus.

An enhanced sensitivity Trofile assay (ESTA) has replaced the original Trofile. The ESTA can detect CXC4-tropic virus present at levels less than 0.3% of the total virus population, and at that level of virus or higher, the assay is stated to be 100% sensitive. Total viral concentration of at least 1000 copies/mL is required. However, ESTA remains limited by long turnaround time and the relatively high minimum level of viremia required, making it not useful in patients in virologic failure with low viremia. Additionally, a small proportion of samples cannot be successfully phenotyped with either generation of the Trofile assay.

The Maraviroc versus Efavirenz Regimens as Initial Therapy trial (MERIT) study (2010) of treatment-naive patients was retrospectively reanalyzed using ESTA; approximately 15% of the subjects originally identified as CCR5-tropic had D/M- or CXC4-tropic virus by ESTA.

Wilkin et al (2011) used ESTA to reanalyze samples from 4 large cohort studies that had originally been evaluated for HIV tropism with the original Trofile assay. Moreover, 9% to 26% of patients with CCR5-tropic virus by the original Trofile assay had CXC4-using virus by ESTA.

V3 Population Genotyping to Determine Tropism

The Trofile assay is a cell-based, functional (phenotypic) assay. Genotypic assays are based on the sequencing of the patient-derived HIV-1 gp120 V3 domain, which determines the protein amino acid sequence for the major determinant of coreceptor binding. This sequencing method results in a V3 sequence that represents the average or dominant viral population sequence for each patient. The patient-derived HIV V3 sequence is used to predict HIV-1 tropism using web-based bioinformatic interpretation tools developed from prior data. These are most often the support vector machine-based geno2pheno (G2P) coreceptor and position-specific scoring matrices. Newer genotypic assays have incorporated additional components of the HIV envelope genotype (e.g., gp41) and/or components of the gp120 gene other than the V3 domain.

Genotyping can be conducted on either viral RNA samples (plasma) or on proviral DNA (peripheral blood mononuclear cells), the latter allowing tropism determination in the context of undetectable viremia. Other potential advantages of genotypic assays are reduced cost, shorter turnaround time, and fewer sample failures.

Early genotyping studies that made comparisons with original Trofile assay results reached contradictory conclusions on the adequacy of genotyping for predicting CXC4 coreceptor usage. Some of the variability in genotype-phenotype assay correlation might have been due to the lower sensitivity of the original Trofile assay, and some variability might have accrued from inclusion of samples containing HIV subtypes other than B (the dominant form in Europe, the Americas, Japan, Thailand, and Australia). Ultimately, the best indication against which tropism assay results should be compared is the virologic outcome of patients who receive CCR5-antagonist medication. Comparison of different tropism assay techniques with reference to virologic outcome of patients is discussed in the Clinical Validity section below.

Newer bioinformatics algorithms continue to be developed, some of which incorporate clinical variables such as HIV-1 viral load and nadir CD4-positive count, into their prediction modeling. Some studies, such as that reported by Ceresola et al (2015) in a cohort of 67 subjects with HIV, have suggested that the G2P algorithm may be more likely to overestimate the frequency of CXC4-tropic viruses compared with other methods.

Table 1 summarizes studies that have evaluated the results of V3 sequencing using ESTA as the reference standard; treatment outcomes were not considered in these analyses. All studies sequenced HIV V3 RNA from plasma (standard assay); two additionally sequenced HIV V3 DNA
from whole blood, which targets proviral DNA (useful for patients with low plasma levels of virus). In general, the sensitivity results indicate that V3 genotyping detects somewhat fewer CXCR4-tropic viral samples than does ESTA; the specificity results indicated that the false-positive rate is not high (i.e., few CCR5-tropic samples were identified as CXCR4-tropic). Assay concordance was relatively high. Where reported, genotyping results for proviral DNA appeared very similar to those for RNA in paired samples from the same patient population (see also Tropism Testing in Patients with Undetectable Viral Load section).

Overall and based largely on the studies of tropism assays with reference to maraviroc treatment outcome (see Clinical Validity section), the evidence has suggested that HIV V3 genotyping classifies patients as well as Trofile assays. Genotyping has additional advantages of shorter turnaround time, ability to generate results for patients who cannot be assayed by Trofile, and more access to assay providers.

### Table 1. Performance of HIV V3 Genotyping With Reference to ESTA

<table>
<thead>
<tr>
<th>Study</th>
<th>N Patients</th>
<th>RT-PCR Replicates</th>
<th>V3 Genotyping Algorithm</th>
<th>V3 Genotyping vs ESTA, %</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>RNA=55</td>
<td>RNA=96</td>
</tr>
<tr>
<td>Prosperi et al (2010)</td>
<td>55</td>
<td>Patients failing antiretroviral treatment</td>
<td>1× G2P clonal, FPR=5.75%</td>
<td>G2P clonal, FPR=10%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Svicher et al (2010)</td>
<td>365</td>
<td>63% treatment-experienced patients</td>
<td>1× G2P clonal, FPR=5%</td>
<td>G2P clonal, FPR=10%</td>
</tr>
<tr>
<td>Sanchez et al (2010)</td>
<td>119</td>
<td>Naive and treatment-experienced patients</td>
<td>1× (?) G2P clonal, FPR=5%</td>
<td>G2P clonal, FPR=10%</td>
</tr>
<tr>
<td>Pou et al (2009)</td>
<td>79</td>
<td>Banked samples, pre-ART</td>
<td>3× G2P</td>
<td>RNA=40</td>
</tr>
</tbody>
</table>

ART: Antiretroviral Therapy; ESTA: Enhanced Sensitivity Trofile Assay; FPR: False-Positive Rate (Used as Cutoff Value); G2P: Geno2pheno Coreceptor System; NR: Not Reported; RT-PCR: Reverse-Transcriptase Polymerase Chain Reaction.

a Abstract.

### Tropism Testing by Deep Sequencing

Because of concern that standard V3 sequencing methods used for tropism testing might miss clinically significant minor HIV variants, so-called “deep sequencing” (i.e., V3 sequencing using next-generation sequencing methods) has been investigated for tropism testing. While standard sequencing essentially determines a population average V3 loop sequence, deep sequencing allows simultaneous sequencing and quantifying of thousands of individual V3 variants within a viral population. From this, the proportion of non-R5 variants in a given sample can be calculated using bioinformatic interpretation tools similar to those for standard V3 genotyping. Similar to the standard V3 sequencing methods, the false-positive rate for tropism prediction must be prespecified. Retrospective analyses have used G2P and a false-positive rate of 3.5% or less. The proportion of the viral population that can be detected as non-CCR5 for maraviroc treatment to remain effective has been established as 2% or less. Other studies have also
reported high concordance between deep sequencing and current tropism assays\textsuperscript{17,24} and between different sequencing platforms.\textsuperscript{25} Gibson et al (2014) reported high concordance (84\%, $\kappa=0.37$) between tropism prediction for samples sequenced with deep sequencing and those sequenced with population-based sequencing.\textsuperscript{26}

**Tropism Testing in Patients with Undetectable Viral Load**

The original studies of genotypic tropism tests, such as those shown in Table 1, were conducted on RNA samples from viremic patients. However, there has been interest in the use of maraviroc as part of a simplification strategy in patients already on antiretroviral therapy with undetectable plasma HIV RNA levels. Another potential indication is as intensification strategy in patients with prolonged suppression of HIV levels but with impaired CD4 gains. A 2012 study by Svicher et al demonstrated the feasibility of determining viral tropism using sequencing of proviral DNA with prediction of tropism with the G2P algorithm in peripheral blood mononuclear cells from 53 subjects with HIV, most of whom had undetectable (94.3\%) or low (3.7\%) viral loads.\textsuperscript{27} Additional studies, outlined in Table 2, have demonstrated high rates of concordance between tropism predicted by proviral DNA or RNA sequencing.
### Table 2. Performance of HIV Proviral DNA Genotyping

<table>
<thead>
<tr>
<th>Study</th>
<th>Study Population</th>
<th>DNA Sequence Success Rate</th>
<th>V3 Genotyping Algorithm</th>
<th>Comparison</th>
<th>Concordance</th>
<th>Sens</th>
<th>Spec</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prosperi et al (2010)</td>
<td>55 patients failing antiretroviral treatment</td>
<td>NR</td>
<td></td>
<td>Proviral DNA vs RNA (ref) (n=29)</td>
<td>87.5% (κ=0.74; 95% CI 0.53 to 0.95; p&lt;0.001)</td>
<td>NR</td>
<td>NR</td>
</tr>
<tr>
<td>Svicher et al (2014)</td>
<td>253 patients with plasma HIV-1 RNA &lt;50 copies/mL</td>
<td>97.6%</td>
<td>G2P clonal, FPR=5.75%</td>
<td>Proviral DNA (whole blood or PBMCs) vs RNA (ref) (n=143)</td>
<td>96.5%</td>
<td>NR</td>
<td>NR</td>
</tr>
<tr>
<td>Brown et al (2014)</td>
<td>42 patients with plasma HIV-1 RNA ≥1000 copies/mL</td>
<td>97.6%</td>
<td>G2P clonal, FPR=10%</td>
<td>Proviral DNA (whole blood) vs RNA (ref)</td>
<td>93% (κ=0.85)</td>
<td>100%</td>
<td>89%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Proviral DNA (PBMCs) vs RNA (ref)</td>
<td>95% (κ=0.90)</td>
<td>100%</td>
<td>93%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Proviral DNA (whole blood or PBMCs) vs RNA (ref)</td>
<td>98% (κ=0.95)</td>
<td>100%</td>
<td>96%</td>
</tr>
</tbody>
</table>

CI: confidence interval; ESTA: enhanced sensitivity Trofile assay; FPR: false-positive rate; G2P: geno2pheno; NR: not reported; PBMC: peripheral blood mononuclear cell; ref: reference group; Sens: sensitivity (defined as concordant x4 results between test and reference methods/reference method CCX4); Spec: specificity (defined as concordant CCR5 results between test and reference methods/reference method CCR5); VL: viral load.
Section Summary: Technically Reliable
The evidence comparing HIV V3 population genotyping with original Trofile and ESTA using maraviroc response as the reference for all assays, strongly suggests that genotyping is equivalent to the Trofile assays in selecting patients likely to respond to maraviroc, the outcomes of interest. Studies evaluating genotyping and using paired ESTA results for reference suggest that genotyping might be somewhat less sensitive for detecting CXCR4-tropic samples; however, these studies were smaller, and most did not test in triplicate. V3 ultra-deep sequencing methods appear to have greater sensitivity in identifying CXCR4-tropic viruses, and therefore are likely to identify additional patients with HIV tropism who are negative on standard sequencing. Based largely on the maraviroc response results, HIV V3 population genotyping is considered medically necessary for patients considering immediate maraviroc treatment.

Clinically Valid
HIV Coreceptor Antagonist Therapy in Treatment-Experienced Patients
The Maraviroc versus Optimized Therapy in Viremic Antiretroviral Treatment-Experienced Patients (MOTIVATE) 1 and 2 trials assessed the efficacy of maraviroc in patients previously treated or resistant to 3 antiretroviral drug classes and with HIV-1 RNA levels exceeding 5000 copies/mL.30 MOTIVATE-1 was conducted in Canada and the United States, and MOTIVATE-2 in Australia, Europe, and the United States, using identical study designs. A total of 1075 patients were randomized to 3 trial arms, and 1049 received at least 1 dose of study drug: placebo (n=209), maraviroc once daily (n=414), or maraviroc twice daily (n=426). Selected subjects had only CCR5-tropic HIV-1 infections, as determined by the original Trofile assay for HIV tropism. At 48-week follow-up in an intention-to-treat analysis, 16% in the placebo group and 45% in both maraviroc-treated groups had HIV-1 RNA levels less than 50 copies/mL. The mean increase in CD4 count from baseline was 60 in the placebo group compared with 120 in the maraviroc groups. Based on the early trial results and review by the Food and Drug Administration Antiviral Drugs Advisory Committee, Food and Drug Administration concluded that, compared with placebo, maraviroc significantly reduced HIV RNA copy number, and significantly increased CD4 cells count, both validated markers of improved health outcomes.31 At nearly 2 years of follow-up (96 weeks), 81% to 87% of maraviroc-treated patients maintained these responses with no new or unexpected events impacting safety.32 At 5-year follow-up, 46 deaths were reported, with ongoing low rates of hepatic failure, malignancy, and myocardial infarction.33

In contrast, in a 2009 trial of 167 patients infected with dual- or mixed-tropic HIV-1, randomized to receive optimal therapy plus maraviroc or placebo, there was no difference in outcomes between treatment groups, indicating maraviroc treatment failure in patients harboring assay-detectable CXCR4-tropic HIV-1 populations.34

HIV Coreceptor Antagonist Therapy in Treatment-Naive Patients
The MERIT study (discussed above) was a randomized, double-blind, multicenter study evaluating subjects infected with CCR5-tropic HIV-1 according to the original Trofile assay.8 In this study, patients had plasma HIV-1 RNA levels of at least 2000 copies/mL; moreover, patients did not have: (1) prior antiretroviral therapy for longer than 14 days, (2) an active or recent opportunistic infection or primary HIV-1 infection, or (3) resistance to zidovudine, lamivudine, or efavirenz. Subjects were randomized to 2 doses of either maraviroc or efavirenz, each in combination with zidovudine/lamivudine. In a preplanned interim analysis, the lower dose of maraviroc failed to meet prespecified efficacy criteria and was discontinued. Patients were stratified by screening HIV-1 RNA levels and by geographic region. The median CD4 cell counts and mean HIV-1 RNA levels at baseline were similar for both treatment groups.

At 96 weeks, after reanalysis using results from the ESTA test (see Tropism Testing section next), virologic response rates in both treatment arms were approximately equal, and there were fewer discontinuations due to adverse events in the maraviroc arm.

Although most newly infected patients harbor CCR5-tropic HIV virus alone, a study of 150 individuals from 2 seroconverter cohorts documented 4% infection with detectable CXCR4-
tropic virus (either mixed or, rarely, CXCR4-only), indicating that tropism analysis is necessary, even for the recently infected.35

Comparison of HIV Tropism Testing Methods to Identify Candidates for HIV Coreceptor Antagonist Therapy

Table 3 summarizes the results of studies comparing V3 genotyping results with virologic outcomes after maraviroc treatment. Because most studies use G2P for interpretation, only these results are presented. Where reported, results of original Trofile and ESTA results are also shown.

Only the study reported by Gonzalez-Serna et al (2012) was prospective; for the others, V3 genotyping was conducted retrospectively on banked samples.36 McGovern et al (2010)37 likely included data reported by Harrigan et al (2009).38 Results varied by the false-positive rate cutoff chosen for the G2P algorithm. If the result provided by G2P for a specific V3 sequence was higher than the chosen cutoff, the prediction of HIV-1 coreceptor tropism was CXCR4. Because the G2P distributions for CCR5- and CXCR4-tropic viruses overlapped, no cutoff value permitted perfect classification. Using a higher cutoff value was considered a conservative choice because predictions of CXCR4-tropism were more likely to be true predictions; the trade-off was that some true CXCR4-tropic HIV infections would be falsely identified as CCR5-tropic. For example, a cutoff value of 5.75% was optimized retrospectively for the MOTIVATE trial data (2009).39 but, for routine clinical practice, the 2011 European guidelines on HIV-1 tropism testing recommended a cutoff of 10% for sequencing of samples in triplicate, or a cutoff of 20% when only a single sequence is generated.40

The results in Table 3 indicate that, depending on the G2P cutoff value chosen, V3 sequencing results can be generated that are very similar in their ability to predict response to maraviroc to both the original Trofile assay and the ESTA test. The Gonzalez-Sema study reported somewhat different results, with lower sensitivity and higher specificity for maraviroc response using similar G2P cutoff values.36 This study prospectively enrolled patients attending the infectious disease service of a university hospital, as opposed to the other retrospective studies of carefully selected clinical trial participants, but was also much smaller. Sequencing in this study was not done in triplicate (as it was in the other studies).

Table 3. Performance of HIV V3 Genotyping, Trofile, and ESTA Assays With Reference to Maraviroc Treatment Outcomes

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample size</td>
<td>705</td>
<td>623</td>
<td>73</td>
<td>1164</td>
</tr>
<tr>
<td>Patients</td>
<td>Drug-naive patients from MERIT trial</td>
<td>Treatment-experienced patients from MOTIVATE and 1029 studies</td>
<td>Patients with persistent viral load and on treatment hiatus</td>
<td>Treatment-experienced patients from MOTIVATE and 1029 studies</td>
</tr>
<tr>
<td>RT-PCR replicates</td>
<td>3×</td>
<td>3×</td>
<td>1×</td>
<td>3×</td>
</tr>
<tr>
<td>VR definition</td>
<td>&lt;50 copies/mL at week 48</td>
<td>• &lt;50 copies/mL or reduction</td>
<td>• &lt;50 copies/mL or reduction</td>
<td>• &lt;50 copies/mL or reduction</td>
</tr>
<tr>
<td></td>
<td></td>
<td>• ≥2 log at week 8</td>
<td>• ≥1 log on day 8</td>
<td>• ≥2 log at week 8</td>
</tr>
<tr>
<td>V3 genotyping algorithm</td>
<td>G2P, FPR=5.75%</td>
<td>G2P, FPR=5%</td>
<td>G2P clonal, FPR=10%</td>
<td>G2P, FPR=5%</td>
</tr>
<tr>
<td>V3 genotyping vs VR to MVC</td>
<td>Sens=94% Spec=13%</td>
<td>Sens=85% Spec=36%</td>
<td>Sens=89% Spec=24%</td>
<td>Sens=92% Spec=20%</td>
</tr>
<tr>
<td>Original Trofile vs VR to MVC</td>
<td>NR</td>
<td>Sens=90% Spec=31%</td>
<td>NR</td>
<td>Sens=92% Spec=20%</td>
</tr>
</tbody>
</table>

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The studies in Table 4 suggest that deep sequencing has similar performance characteristics as ESTA and the original Trofile assay in predicting response to maraviroc treatment. Moreover, as noted by Swenson et al (2011), the group of patients with 2% to 20% non-CCR5 virus, according to deep sequencing, had minority non-CCR5 variants that were not reliably detected by the original Trofile assay; however, this particular group of patients had poor response to maraviroc, with 27% of the patients achieving virologic suppression at week 48—this is similar to the non-CCR5 group as a whole (26%) and to patients with greater than 20% non-R5 virus (25%). Kagan et al (2012) reanalyzed samples from the MOTIVATE and A4001029 studies to compare ultra-deep sequencing either alone or as a reflex test following standard triplicate V3 sequencing with the ESTA test. Both ultra-deep sequencing methods demonstrated improved sensitivity in identifying maraviroc responders compared with standard sequencing. These results would suggest that detection of minority non-CCR5 variants by deep sequencing may be important for predicting response.

Table 4. Performance of HIV V3 Deep Sequencing, Trofile, and ESTA Assays With Reference to Maraviroc Treatment Outcomes

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample size</td>
<td>27</td>
<td>859</td>
<td>851</td>
<td>327</td>
</tr>
<tr>
<td>Patients</td>
<td>Patients with persistent viral load and on treatment hiatus</td>
<td>Drug-naive patients from MERIT trial</td>
<td>Treatment-experienced patients from MOTIVATE and A4001029 studies</td>
<td>Treatment-experienced patients from MOTIVATE and A4001029 studies who received MVC</td>
</tr>
<tr>
<td>RT-PCR replicates</td>
<td>3x</td>
<td>3x</td>
<td>3x</td>
<td>3x</td>
</tr>
<tr>
<td>VR definition</td>
<td>• &lt;50 copies/mL or reduction</td>
<td>&lt;50 copies/mL at week 48</td>
<td>&lt;50 copies/mL at week 48</td>
<td>&lt;50 copies/mL or &gt;2 log decline at week 8</td>
</tr>
<tr>
<td>V3 genotyping algorithm</td>
<td>G2P clonal, FPR ≤3.5%</td>
<td>G2P clonal, FPR ≤3.5%</td>
<td>• PSSM x4/R5, FPR ≥ -4.75</td>
<td>• G2P, FPR ≤5.75%</td>
</tr>
<tr>
<td>V3 genotyping vs VR to MVC</td>
<td>Sens=83% Spec=22%</td>
<td>Sens=93% Spec=15%</td>
<td>Sens=83% Spec=36%</td>
<td>PPV=65% NPV&lt;sub&gt;a&lt;/sub&gt;=61%</td>
</tr>
<tr>
<td>Original Trofile vs VR to MVC</td>
<td>NR</td>
<td>NR</td>
<td>Sens=93% Spec=17%</td>
<td>PPV=66% NPV=59%</td>
</tr>
<tr>
<td>ESTA vs VR to MVC</td>
<td>NR</td>
<td>NR</td>
<td>Sens=90% Spec=21%</td>
<td>PPV=66% NPV=59%</td>
</tr>
</tbody>
</table>

conc: concordance; ESTA: enhanced sensitivity Trofile assay; FPR: false-positive rate (used as cutoff value); G2P: Geno2pheno coreceptor system; MERIT: Maraviroc versus Efavirenz Regimens as Initial Therapy trial; MOTIVATE: Maraviroc Plus Optimized Therapy in Viremic Antiretroviral Treatment-Experienced Patients trials; MVC: maraviroc; NPV: negative predictive value; NR: not reported; PPV: positive predictive value; PSSM: position-specific scoring matrix; RT-PCR: reverse-transcriptase polymerase chain reaction; Sens: sensitivity; Spec: specificity; VR: virologic response.

<sub>a</sub> PPV refers to the proportion of CCR5 subjects who achieved virologic response at 8 wk. NPV refers to the proportion of non-CCR5 subjects who failed to have a virologic response at 8 wk.
A 2014 prospective, phase 3 trial by Heera et al, which randomized treatment-naive patients with HIV to genotypic or phenotypic (Trofile) testing, showed no significant differences in treatment response. Previously presented results of European cohort studies have shown maraviroc virologic extended response rates of 69% to 82% in those patients in which HIV variants were genotypically classified CCR5-tropic.

Nozza et al (2016) conducted a multicenter, randomized, open-label, noninferiority trial among treatment-experienced subjects with HIV-1 RNA of 500 or more copies per milliliter. One hundred fifty-five participating patients were randomized (1:1) to undergo coreceptor tropism testing by the G2P algorithm (false-positive rate >10%) or the Trofile assay before starting a new antiretroviral regimen. Only patients with an R5 tropic virus were enrolled and received treatment with maraviroc plus optimized background therapy. The primary end point was the 48-week proportion of patients with treatment success (defined as HIV RNA <50 copies/mL). In the Trofile arm, 87% of patients achieved treatment success at 48 weeks, and in the G2P arm, 89% achieved treatment success at 48 weeks; these results suggest noninferiority.

Garcia et al (2014) reported in abstract form the results of the PROTEST study, which evaluated the initiation of maraviroc plus 2 nucleoside reverse-transcriptase inhibitors in aviremic subjects based on genotypic tropism testing of proviral DNA, rather than viral RNA. The study included 74 maraviroc-naive HIV-1 patients with viral load less than 50 c/mL on stable antiretroviral therapy, requiring medication change due to toxicity, and CCR5-tropic HIV by proviral DNA genotypic tropism testing. Of the included subjects, 62 (84%) maintained a viral load less than 50 c/mL through 48 weeks of therapy. The remaining 12 (16%) discontinued treatment: 2 (3%) withdrew informed consent; 2 (3%) died of non-study-related causes; 5 (7%) developed protocol-defined virologic failure; and 1 each (1% each) had a shift to CCX4 between the screening and baseline visits or was lost to follow-up, or developed an antiretroviral therapy-related adverse event.

**Clinically Useful**

Among patients who are undergoing HIV tropism testing to determine if they are suitable for maraviroc treatment, there is no direct evidence that HIV tropism testing results in improved health outcome in terms of overall or disease-specific survival. However, there is evidence that selection of candidates for HIV coreceptor antagonist therapy using HIV tropism tests results in a high rate of treatment success, demonstrated as increased virologic suppression. Plasma viral load is the single best predictor of progression to AIDS and death. Successful virologic suppression leads to longer overall survival and disease-specific survival among HIV-infected patients.

**Section Summary: HIV Tropism Testing to Identify Candidates for HIV Coreceptor Antagonist Therapy**

Evidence from randomized controlled trials (RCTs) and observational studies has suggested high sensitivity of the Trofile assay, the ESTA test, V3 sequencing, and V3 deep sequencing in identifying treatment-naive and treatment-experienced HIV-infected candidates for HIV coreceptor antagonist therapy, with treatment outcome as the reference. Studies have also suggested a moderate (>70%) level of concordance between different HIV tropism testing techniques.

**HIV Tropism Testing for Treatment Monitoring and Therapy Failure**

**Clinical Context and Test Purpose**

The purpose of HIV tropism testing in patients with HIV infection receiving treatment with HIV coreceptor antagonist or who have failed coreceptor antagonist therapy is to monitor or detect possible tropism switching.

The question addressed in this evidence review is: Does assessment of HIV tropism among HIV-infected patients undergoing maraviroc therapy, or patients who have experienced virologic
failure while on maraviroc therapy, result in an improved health outcome compared with no testing to identify HIV tropism switching?

The following PICOTS were used to select literature to inform this review.

**Patients**
The relevant populations of interest are 1 of 2 patient populations: (1) HIV-infected patients undergoing treatment with HIV coreceptor antagonists; or (2) patients who have failed coreceptor antagonist therapy.

**Interventions**
The interventions of interest are HIV tropism testing using the Trofile assay, ESTA, V3 sequencing, or V3 deep sequencing.

**Comparators**
The comparator of interest is no HIV tropism testing.

**Outcomes**
The potential beneficial outcomes of primary interest would be identification of HIV-infected patients who might benefit from changes in antiretroviral therapy regimen. The potential harmful outcomes are those resulting from a false test result. False-positive or false-negative test results can lead to the initiation of unnecessary treatment and adverse events from that treatment.

**Timing**
HIV tropism testing should be conducted before starting HIV coreceptor antagonist therapy.

**Setting**
Ordering and interpreting of HIV tropism testing should be done by physicians specializing in infectious diseases. Most patients are likely to be tested in an outpatient setting. Referral for genetic counseling is important for the explanation of genetic disease, heritability, genetic risk, test performance, and possible outcomes.

**Technically Reliable**
Evidence on the technical reliability of different HIV tropism testing techniques has been discussed in the section on identifying candidates for HIV coreceptor antagonist therapy.

**Clinically Valid**
Viral strains transmitted in vivo are usually CCR5-tropic. Over time, and more often after antiretroviral treatment, detectable CXCR4-tropic virus emerges in about half of patients, and this virus is associated with rapid CD4 cell depletion and clinical disease progression. However, patients whose infection remains predominately CCR5-tropic can also experience disease progression. HIV-1 viral load is a strong prognostic indicator of HIV disease progression, and suppression of viral load is a critical goal of antiretroviral therapy. Viral rebound (virologic failure) is typically followed by a reduction in CD4 cell count (immunologic failure), and if not adequately addressed by changes in treatment, by HIV-related events (clinical progression). Thus, the success of any antiretroviral treatment regimen is monitored by measuring HIV-1 RNA level and CD4 cell count; significant changes direct patient management.

The prominent reason for individual treatment failure in the clinical studies was an outgrowth of a minor CXCR4-tropic virus population not detected at screening. However, treatment failure with CCR5-tropic virus alone also occurred, indicating that resistance to CCR5 antagonists occurs independently of tropism. In vitro studies have provided extensive information on resistance; mechanisms may involve the ability of HIV to bind the CCR5 inhibitor-receptor complex. Resistance to CCR5 antagonists has been associated with an increased affinity for CCR5, changes in the gp120 V3 loop, and with other gp120 (or other envelope) changes.
A concern about treatment with CCR5 coreceptor antagonists is that small, undetectable populations of CXCR4-tropic virus would be enriched and would accelerate disease progression. However, in a randomized, placebo-controlled phase 2 study of maraviroc treatment of patients with D/M-tropic infections, there was no evidence that this was the case. The association between CXCR4 tropism (defined with the original Trofile assay) and clinical progression has been shown to be independent of CD4 cell count and HIV-1 RNA level (adjusted hazard ratio, 3.82; 95% confidence interval, 1.69 to 8.60; p=0.001, vs patients with CCR5-tropic infection only).

Fatkenheuer et al (2008) performed a post hoc analysis of the virologic response according to HIV tropism at baseline and at treatment failure using pooled data from the MOTIVATE 1 and 2 trials. Virologic failure occurred in 53% of placebo-treated patients and in 22% to 23% in the maraviroc treatment arms. However, of the 133 treatment failures in the maraviroc groups, 76 (57%) had CXCR4 or D/M tropism, as compared with only 6 (6%) of 95 in the placebo group; this finding raises concerns that maraviroc treatment could lead to the emergence of CXCR4-tropic subpopulations and, ultimately, more rapid development of clinical progression. However, this was not the case because the CXCR4 maraviroc treatment failures were not associated with declines in CD4 cell counts or with disease progression.

Raymond et al (2015) conducted a multicenter study to characterize virologic failure in patients treated with maraviroc (n=27). Patients had been screened for HIV tropism using population-based V3 genotyping before maraviroc initiation. Authors determined HIV tropism and resistance of R5 viruses to maraviroc at baseline and at virologic failure retrospectively using an ultra-sensitive recombinant virus assay. Among the 27 patients experiencing virologic failure, 12 harbored CXCR4-using viruses, and 15 had R5 viruses at failure. Four of the 12 harboring CXCR4 viruses were infected with D/M-tropic viruses, according to the recombinant virus assay before maraviroc initiation.

The most common mechanism of maraviroc treatment failure is the emergence of a CXCR4-tropic viral population. However, this does not necessarily correlate with rapid clinical progression.

**Clinically Useful**

For HIV-infected patients who are receiving maraviroc treatment, there is no direct evidence that HIV tropism testing—both during treatment monitoring and at virologic failure—results in improved health outcomes. The lack of evidence that HIV tropism testing might predict treatment failure among patients who are on maraviroc therapy, therefore, suggests that HIV tropism testing in this population might not result in improved health outcomes. Treatment failure is detected by increased viral load and decreased CD4 cell count, indicating that maraviroc treatment can be discontinued.

**Section Summary: HIV Tropism Testing for Treatment Monitoring and Therapy Failure**

The evidence for the use of HIV tropism testing for treatment monitoring and virologic failure in patients receiving maraviroc treatment includes post hoc analysis of data from RCTs and observational studies. While the emergence of the CXCR4-tropic viral population is the most common mechanism of maraviroc treatment failure, treatment failure is also common among patients with CCR5-tropic viruses. There is no evidence that tropism testing for treatment monitoring might predict treatment failure.

**HIV Tropism Testing for HIV Prognosis**

**Clinical Context and Test Purpose**

The purpose of HIV tropism testing in patients who have HIV infection is to identify patients who might experience rapid disease progression, such as the short-term risk of AIDS and death.
The question addressed in this evidence review is: Does assessment of HIV tropism to predict disease progression among HIV-infected patients result in an improved health outcome compared with CD4 count or viral load testing?

The following PICOTS were used to select literature to inform this review.

**Patients**
The relevant population of interest is HIV-infected patients.

**Interventions**
The interventions of interest are HIV tropism testing using the Trofile assay, ESTA, V3 sequencing, or V3 deep sequencing.

**Comparators**
The comparator of interest is no HIV tropism testing.

**Outcomes**
The potential beneficial outcomes of primary interest would be identification of HIV-infected patients who might benefit from changes in antiretroviral therapy regimen. The potential harmful outcomes are those resulting from a false test result. False-positive or false-negative test results can lead to the initiation of unnecessary treatment and adverse events from that treatment or undertreatment.

**Timing**
HIV tropism testing should be conducted before starting HIV coreceptor antagonist therapy.

**Setting**
Ordering and interpreting of HIV tropism testing should be done by physicians specializing in infectious diseases. Most patients are likely to be tested in an outpatient setting. Referral for genetic counseling is important for the explanation of genetic disease, heritability, genetic risk, test performance, and possible outcomes.

**Technically Reliable**
Evidence on the technical reliability of different HIV tropism testing techniques has been discussed in the section on identifying candidates for HIV coreceptor antagonist therapy.

**Clinically Valid**
Aside from the specific situation of maraviroc treatment failure, CXCR4-tropic virus infection has been associated with more rapid disease progression, compared with CCR5 infection, in several studies (e.g., Wilkin et al [2011], Almeida et al [2014], Visseaux et al [2014]). However, other studies have demonstrated no independent association between the HIV tropism and HIV-related outcomes, including short-term risk of AIDS and death and hepatic fibrosis in HIV/hepatitis C virus-coinfected patients.

Casadella et al (2017) conducted a nested case-control study within the EuroSIDA cohort to investigate whether plasma HIV-1 tropism testing could identify subjects at higher risk for clinical progression and death in routine clinical management. Cases (N=100) were subjects with AIDS or who had died from any cause, with a plasma sample of HIV-1 RNA greater than 1000 copies/mL available for tropism testing 3 to 12 months prior to the event. At least 1 matched (for age, HIV-1 RNA, and HCV status) control per case was selected (N=166). Baseline tropism was not associated with the risk of clinical progression or death (OR=0.66; 95% CI, 0.33 to 1.33). Female gender (OR=2.13; 95% CI, 1.04 to 4.36), being on antiretroviral therapy (OR=2.12; 95% CI, 1.15 to 4.41), baseline CD4 count (OR=0.90; 95% CI, 0.80 to 1.00), per 100 cells/mm³ higher and calendar year of sample (OR=0.84; 95% CI, 0.77 to 0.91) per more recent year were independently associated with disease progression.
Castagna et al (2016) conducted a longitudinal cohort study of HIV-1–treated adults to determine the rate of HIV tropism switch among subjects using antiretroviral therapy both in presence of persistently detectable (PD) or undetectable (PU) viral load and to evaluate the association between tropism switch and disease progression. Over a median follow-up period of 22.6 months (range, 19.8-28.1 months), 124 PD and 71 PU patients showed similar rates of switch to a non-R5 virus (PD=6.9/100 person-years; 95% CI, 3.7 to 11.2/100 person-years; PU=8.0/100 person-years; 95% CI, 3.4 to 14.5/100 person-years). Switch to non-R5 virus was predicted by nadir CD4-positive count before the start of the follow-up period. Twenty-two (18%) PD and 4 (6%) PU subjects experienced disease progression (p=0.02). The risk of disease progression was independently associated with disease progression (adjusted hazard ratio, 4.06; 95% CI, 1.20 to 13.80).

Clinically Useful
Currently, there is no direct evidence that HIV tropism testing for assessment of disease progression among HIV-infected patients results in improvement of health outcomes. More studies are required comparing HIV tropism testing with other tests (CD4, viral load) for predicting disease progression.

Section Summary: HIV Tropism Testing for HIV Prognosis
The evidence for the use of tropism testing for HIV prognosis includes nested case-control and cohort studies. While some studies demonstrated an association between the HIV tropism and HIV-related outcomes, the findings have been inconsistent. Viral load and CD4 count remain independently associated with disease progression among HIV-infected patients across studies.

Summary of Evidence
For individuals who have HIV infection who are being considered for HIV coreceptor antagonist therapy who receive HIV tropism testing, the evidence includes RCTs. Relevant outcomes are overall survival, disease-specific survival, morbid events, quality of life, hospitalizations, medication use, and treatment-related morbidity. RCTs on treatment-naive and treatment-experienced HIV-infected patients have provided evidence that selection of candidates for HIV coreceptor antagonist therapy using HIV tropism testing results in higher rates of treatment success compared with HIV coreceptor antagonist therapy without HIV tropism testing. The evidence is sufficient to determine that the technology results in a meaningful improvement in the net health outcome.

For individuals with HIV infection receiving HIV coreceptor antagonist therapy or who have failed coreceptor antagonist therapy who receive HIV tropism testing, the evidence includes post hoc analysis of RCTs and observational studies. Relevant outcomes are overall survival, disease-specific survival, morbid events, quality of life, hospitalizations, medication use, and treatment-related mortality and morbidity. Current evidence does not indicate improved outcomes with additional tropism monitoring during treatment. The evidence is insufficient to determine the effects of the technology on health outcomes.

For individuals with HIV infection who are undergoing tests to predict disease progression who receive HIV tropism testing, the evidence includes observational studies. Relevant outcomes are overall survival, disease-specific survival, morbid events, quality of life, hospitalizations, and medication use. Current evidence is inconsistent as relates to whether HIV tropism testing independently predicts disease progression among HIV-infected patients. The evidence is insufficient to determine the effects of the technology on health outcomes.

Supplemental Information
Practice Guidelines and Position Statements

HIV Medicine Association of the Infectious Disease Society of North America
The HIV Medicine Association of the Infectious Disease Society of North America updated its guidelines on the on the management of persons infected with HIV in 2013. These guidelines...
stated that tropism testing should be performed if the use of a CCR5 antagonist is being considered (strong recommendation, high quality evidence). The guidelines also stated that “routine tropism testing is not recommended prior to initiation of other regimens because of cost and lack of demonstrated benefit.” The guidelines did not specify the preferred method of tropism testing.

**European Consensus Group**
The European Consensus Group recommendations on clinical management of tropism testing stated that tropism testing is indicated for patients who fail treatment or have unacceptable toxicity and a CCR5 inhibitor is being considered. In the absence of evidence, the group provided no guidance regarding tropism testing for newly diagnosed patients whose immediate treatment plan does not include a CCR5 inhibitor. In the absence of adequate data, the group provided no guidance on the question of testing treatment-naïve patients prior to the start of a regimen not including a CCR5 inhibitor, in anticipation of the need to switch quickly to a CCR5 inhibitor due to the toxicity of the initial treatment regimen. For patients with a plasma HIV RNA load greater than 1000 copies/mL, tropism testing was recommended with the enhanced sensitivity Trofile assay or by population genotypic analysis of the V3 loop, based on a moderate level of evidence from on well-designed, nonrandomized trials or cohort studies with long-term clinical outcomes. For patients with a plasma HIV RNA load less than 1000 copies/mL, genotyping was the preferred method.

**Department of Health and Human Services**
The Department of Health and Human Services Panel on Antiretroviral Guidelines for Adults and Adolescents published federally approved HIV and AIDS medical practice guidelines in 2014, which made the following recommendations on coreceptor tropism assays:
- Recommendations with “A” (strong) rating:
  - A coreceptor tropism assay should be performed whenever the use of a CCR5 coreceptor antagonist is being considered (level of evidence: I [data from randomized controlled trials]).
  - A phenotypic tropism assay is preferred to determine HIV-1 coreceptor usage (level of evidence: I).
- Recommendations with “B” (moderate) rating:
  - Coreceptor tropism testing is also recommended for patients who exhibit virologic failure on a CCR5 antagonist (level of evidence: III [expert opinion]).
  - A genotypic tropism assay should be considered as an alternative to predict HIV-1 coreceptor usage (level of evidence: II [data from well-designed nonrandomized trials or observational studies with long-term clinical outcomes]).

**U.S. Preventive Services Task Force Recommendations**
Not applicable.

**Medicare National Coverage**
There is no national coverage determination. In the absence of a national coverage determination, coverage decisions are left to the discretion of local Medicare carriers.

**Ongoing and Unpublished Clinical Trials**
A search of ClinicalTrials.gov in November 2017 did not identify any ongoing or unpublished trials that would likely influence this review.

### Appendix

#### Appendix Table 1. Categories of Genetic Testing Addressed in 2.04.49

<table>
<thead>
<tr>
<th>Category</th>
<th>Addressed</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Testing of an affected individual’s germline to benefit the individual</td>
<td>X</td>
</tr>
<tr>
<td>1a. Diagnostic</td>
<td>X</td>
</tr>
<tr>
<td>1b. Prognostic</td>
<td></td>
</tr>
<tr>
<td>1c. Therapeutic</td>
<td></td>
</tr>
</tbody>
</table>
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Laboratory Testing for HIV Tropism

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2. Testing cancer cells from an affected individual to benefit the individual
   2a. Diagnostic
   2b. Prognostic
   2c. Therapeutic
3. Testing an asymptomatic individual to determine future risk of disease
4. Testing of an affected individual's germline to benefit family members
5. Reproductive testing
   5a. Carrier testing: preconception
   5b. Carrier testing: prenatal
   5c. In utero testing: aneuploidy
   5d. In utero testing: familial variants
   5e. In utero testing: other
   5f. Preimplantation testing with in vitro fertilization

References


**Documentation for Clinical Review**

*Please provide the following documentation (if/when requested):*

- History and physical and/or consultation notes including:
  - Description of the lab test being ordered from the prescribing physician
  - Previous treatment plan and response

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Post Service

- Results/reports of tests performed

Coding

This Policy relates only to the services or supplies described herein. Benefits may vary according to product design; therefore, contract language should be reviewed before applying the terms of the Policy. Inclusion or exclusion of a procedure, diagnosis or device code(s) does not constitute or imply member coverage or provider reimbursement.

MN/IE

The following services may be considered medically necessary in certain instances and investigational in others. Services may be considered medically necessary when policy criteria are met. Services may be considered investigational when the policy criteria are not met or when the code describes application of a product in the position statement that is investigational.

<table>
<thead>
<tr>
<th>Type</th>
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<tr>
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<td>ICD-10 Procedure</td>
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Policy History

This section provides a chronological history of the activities, updates and changes that have occurred with this Medical Policy.

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<th>Effective Date</th>
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<tbody>
<tr>
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<td>BCBSA Medical Policy adoption</td>
<td>Medical Policy Committee</td>
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<tr>
<td>06/30/2015</td>
<td>Policy revision without position change</td>
<td>Medical Policy Committee</td>
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<tr>
<td>03/01/2017</td>
<td>Policy revision without position change</td>
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</tr>
<tr>
<td>02/01/2018</td>
<td>Policy revision without position change</td>
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Definitions of Decision Determinations

Medically Necessary: A treatment, procedure, or drug is medically necessary only when it has been established as safe and effective for the particular symptoms or diagnosis, is not investigational or experimental, is not being provided primarily for the convenience of the patient or the provider, and is provided at the most appropriate level to treat the condition.

Investigational/Experimental: A treatment, procedure, or drug is investigational when it has not been recognized as safe and effective for use in treating the particular condition in accordance with generally accepted professional medical standards. This includes services where approval by the federal or state governmental is required prior to use, but has not yet been granted.

Split Evaluation: Blue Shield of California/Blue Shield of California Life & Health Insurance Company (Blue Shield) policy review can result in a split evaluation, where a treatment, procedure, or drug will be considered to be investigational for certain indications or conditions, but will be deemed safe and effective for other indications or conditions, and therefore potentially medically necessary in those instances.
Prior Authorization Requirements (as applicable to your plan)

Within five days before the actual date of service, the provider must confirm with Blue Shield that the member's health plan coverage is still in effect. Blue Shield reserves the right to revoke an authorization prior to services being rendered based on cancellation of the member's eligibility. Final determination of benefits will be made after review of the claim for limitations or exclusions.

Questions regarding the applicability of this policy should be directed to the Prior Authorization Department. Please call (800) 541-6652 or visit the provider portal at www.blueshieldca.com/provider.

Disclaimer: This medical policy is a guide in evaluating the medical necessity of a particular service or treatment. Blue Shield of California may consider published peer-reviewed scientific literature, national guidelines, and local standards of practice in developing its medical policy. Federal and state law, as well as contract language, including definitions and specific contract provisions/exclusions, take precedence over medical policy and must be considered first in determining covered services. Member contracts may differ in their benefits. Blue Shield reserves the right to review and update policies as appropriate.