| 2.04.151 | Biomarker Testing (Including Liquid Biopsy) for Targeted Treatment and Immunotherapy in Breast Cancer | | |
|---|---|---|---|
| **Original Policy Date:** | February 1, 2021 | **Effective Date:** | February 1, 2021 |
| **Section:** | 2.0 Medicine | **Page:** | Page 1 of 31 |

## Policy Statement

***PIK3CA* Testing**

*PIK3CA* testing may be **medically necessary** to predict treatment response to alpelisib (Piqray) in patients with hormone receptor-positive, HER2 negative advanced or metastatic breast cancer (see Policy Guidelines).

*PIK3CA* testing of tissue is considered **investigational** in all other situations unless included in a panel approved under another policy.

***NTRK* Gene Fusion Testing**

Analysis of *NTRK* gene fusions may be considered **medically necessary** to predict treatment response to entrectinib (Rozlytrek) or larotrectinib (Vitrakvi) in patients with locally advanced or metastatic breast cancer that has progressed following standard treatment and who have no alternative treatment option (see Policy Guidelines).

Analysis of *NTRK* gene fusions is considered **investigational** in all other situations unless included in a panel approved under another policy.

**PD-L1 Testing**

PD-L1 testing may be considered **medically necessary** to predict treatment response to atezolizumab (Tecentriq) in patients with hormone receptor-negative/HER2-negative (triple negative) metastatic or unresectable breast cancer (see Policy Guidelines).

PD-L1 testing may be considered **medically necessary** to predict treatment response to pembrolizumab (Keytruda) in patients with hormone receptor-negative/HER2-negative (triple negative) recurrent or metastatic breast cancer (see Policy Guidelines).

PD-L1 testing is considered **investigational** in all other situations unless included in a panel or separately approved under another policy.

**MSI-H/dMMR Testing**

MSI-H/dMMR testing may be considered **medically necessary** to predict treatment response to pembrolizumab (Keytruda) in patients with unresectable or metastatic breast cancer that has progressed following standard treatment and who have no alternative treatment option (see Policy Guidelines).

MSI-H/dMMR testing is considered **investigational** in all other situations unless included in a panel or separately approved under another policy.

**Tumor Mutational Burden Testing**

Tumor mutational burden testing to predict response to immunotherapy in patients with breast cancer is considered **investigational.**

**Circulating Tumor DNA**

*PIK3CA* testing using FoundationOne Liquid CDx (FDA approved companion test) may be considered **medically necessary** to predict treatment response to alpelisib (Piqray) in patients with hormone receptor-positive, HER2 negative advanced or metastatic breast cancer (see Policy Guidelines) when there is insufficient tissue to be tested and an additional invasive procedure would be required otherwise.

Circulating tumor DNA testing is considered **investigational** in all other situations unless included in a panel approved under another policy, such as use in Non-Small Cell Lung Cancer (NSCLC).

**Circulating Tumor Cells**
Analysis of circulating tumor cells to select treatment in patients with breast cancer is considered **investigational** (see Background section).

**NOTE**: Refer to Appendix A to see the policy statement changes (if any) from the previous version.

## Policy Guidelines

This policy does not address testing of germline variants (see Genetic Testing for BRCA1 or BRCA2 for Hereditary Breast/Ovarian Cancer Syndrome and Other High-Risk Cancers).

**Note:** The use of PARP inhibitors (e.g., Lynparza/olaparib or talazoparib) in HER2-negative metastatic breast cancer with a germline BRCA mutation, is sometimes based on germline rather than somatic mutations in BRCA. Both may be tested as well as HER2 somatic tumor testing. Myriad myChoice (CPT 0172U) may be used for somatic BRCA testing (esp. for ovarian cancer) and BRACAnalysis CDx (Myriad Genetic Laboratories) may be used for germline BRCA testing to help determine eligible patients.

See FDA labels, clinical trials, and NCCN guidelines for specific population descriptions. Descriptions varied slightly across sources.

**Coding**
The following CPT codes may be used for this genomic sequence analysis:

- **0037U**: Targeted genomic sequence analysis, solid organ neoplasm, DNA analysis of 324 genes, interrogation for sequence variants, gene copy number amplifications, gene rearrangements, microsatellite instability and tumor mutational burden (PLA for the FoundationOne CDx™ (F1CDx®) test)
- **0155U:** Oncology (breast cancer), DNA, PIK3CA (phosphatidylinositol-4,5-bisphosphate 3-kinase, catalytic subunit alpha) (e.g., breast cancer) gene analysis (i.e., p.C420R, p.E542K, p.E545A, p.E545D [g.1635G>T only], p.E545G, p.E545K, p.Q546E, p.Q546R, p.H1047L, p.H1047R, p.H1047Y), utilizing formalin-fixed paraffin-embedded breast tumor tissue, reported as PIK3CA gene mutation status (PLA code for the therascreen® PIK3CA RGQ PCR Kit from QIAGEN)
- **0177U:** Oncology (breast cancer), DNA, PIK3CA (phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha) gene analysis of 11 gene variants utilizing plasma, reported as PIK3CA gene mutation status (PLA code for the therascreen® PIK3CA RGQ PCR Kit test from QIAGEN)
- **81309:** PIK3CA (phosphatidylinositol-4, 5-biphosphate 3-kinase, catalytic subunit alpha) (e.g., colorectal and breast cancer) gene analysis, targeted sequence analysis (e.g., exons 7, 9, 20)

***Effective January 1, 2021***, there is a new Molecular Pathology codes to support Neurotrophic receptor tyrosine kinase (NTRK) gene testing:

- **81191:** NTRK1 (neurotrophic receptor tyrosine kinase 1) (e.g., solid tumors) translocation analysis
- **81192:** NTRK2 (neurotrophic receptor tyrosine kinase 2) (e.g., solid tumors) translocation analysis
- **81193:** NTRK3 (neurotrophic receptor tyrosine kinase 3) (e.g., solid tumors) translocation analysis
- **81194:** NTRK (neurotrophic-tropomyosin receptor tyrosine kinase 1, 2, and 3) (e.g., solid tumors) translocation analysis

Testing for variants in the other genes listed above would be reported with the following code:

- **81479**: Unlisted molecular pathology procedure

## Description

Multiple biomarkers are being evaluated to predict response to targeted treatments and immunotherapy for patients with advanced breast cancer. These include tissue-based testing as well as circulating tumor DNA and circulating tumor cell testing (known as liquid biopsy).

The objective of this evidence review is to examine whether biomarker testing for *PIK3CA*, *NTRK gene fusions*, PD-L1, MSI-H/dMMR, TMB, circulating tumor DNA, or circulating tumor cells improves the net health outcome in patients with recurrent, metastatic, or unresectable breast cancer.

## Related Policies

- Genetic Testing for BRCA1 or BRCA2 for Hereditary Breast/Ovarian Cancer Syndrome and Other High-Risk Cancers
- Comprehensive Genomic Profiling for Selecting Targeted Cancer Therapies
- Circulating Tumor DNA and Circulating Tumor Cells for Cancer Management (Liquid Biopsy)
- Assays of Genetic Expression in Tumor Tissue as a Technique to Determine Prognosis in Patients with Breast Cancer

## 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. Laboratories that offer laboratory-developed tests must be licensed by the Clinical Laboratory Improvement Amendments for high-complexity testing. To date, the U.S. Food and Drug Administration has chosen not to require any regulatory review of these tests.

Table 1 summarizes available targeted treatments with FDA approval for recurrent or metastatic breast cancer (including immunotherapy) and the FDA approved companion diagnostic tests associated with each.

**Table 1. Targeted Treatments for Metastatic Breast Cancer and FDA Approved Companion Diagnostic Tests**

| Treatment | Class | Indications in Breast Cancer | Companion Diagnostic |
|---|---|---|---|

| | | | |
|---|---|---|---|
| **ado-trastuzumab emtansine (Kadcyla)** | HER2-targeted antibody and microtubule inhibitor conjugate | As a single agent, for:<br>• Treatment of patients with HER2-positive, metastatic breast cancer who previously received trastuzumab and a taxane, separately or in combination. Patients should have either:<br>o received prior therapy for metastatic disease, or<br>o developed disease recurrence during or within 6 months of completing adjuvant therapy.<br>• Adjuvant treatment of patients with HER2-positive early breast cancer who have residual invasive disease after neoadjuvant taxane and trastuzumab-based treatment. | FoundationOne CDx<br>HER2 FISH pharmDx Kit<br>HercepTest<br>INFORM HER2 Dual ISH DNA Probe Cocktail<br>PATHWAY anti-Her2/neu (4B5) Rabbit Monoclonal Primary Antibody |
| **Alpelisib (Piqray)** | Kinase inhibitor | In combination with fulvestrant for the treatment of postmenopausal women, and men, with hormone receptor (HR) positive, human epidermal growth factor receptor 2 (HER2)-negative, PIK3CA-mutated, advanced or metastatic breast cancer as detected by an FDA approved test following progression on or after an endocrine-based regimen | FoundationOne CDx<br>FoundationOne Liquid CDx<br>therascreen PIK3CA RGQ PCR Kit |
| **Atezolizumab (Tecentri)** | PD-L1 blocking antibody | In combination with paclitaxel protein-bound for the treatment of adult patients with unresectable locally advanced or metastatic TNBC whose tumors express PD-L1, as determined by an FDA approved test. | VENTANA PD-L1(SP142) Assay |
| **Entrectinib (Rozlytrek)** | Kinase inhibitor | Adult and pediatric patients 12 years of age and older with solid tumors that:<br>• have a neurotrophic tyrosine receptor kinase (NTRK) gene fusion without a known acquired resistance mutation,<br>• are metastatic or where surgical resection is likely to result in severe morbidity, and<br>• have progressed following treatment or have no satisfactory alternative therapy | No FDA approved companion diagnostic test |
| **Larotrectinib (Vitrakvi)** | Kinase inhibitor | Adult and pediatric patients 12 years of age and older with solid tumors that:<br>• have a neurotrophic tyrosine receptor kinase (NTRK) gene fusion without a known acquired resistance mutation,<br>• are metastatic or where surgical resection is likely to result in severe morbidity, and<br>• have progressed following treatment or have no satisfactory alternative therapy | FoundationOne CDx |
| **Olaparib (Lynparza)** | PARP inhibitor | Adult patients with deleterious or suspected deleterious germline BRCA mutated, HER2-negative metastatic breast cancer who have been treated with chemotherapy in the neoadjuvant, adjuvant or metastatic setting. Patients with hormone receptor (HR)-positive breast cancer should have been treated with a prior endocrine therapy or be considered inappropriate for endocrine therapy. Select patients for therapy based on an FDA approved companion diagnostic for Lynparza. | BRACAnalysis CDx |
| **Pembrolizumab (Keytruda)** | PD-L1-blocking antibody | • in combination with chemotherapy, for the treatment of patients with locally recurrent unresectable or metastatic TNBC whose tumors express PD-L1 as determined by an FDA approved test | PD-L1 IHC 22C3 pharmDx |
| | | • Adult and pediatric patients with unresectable or metastatic, microsatellite | No FDA approved |

| | | | |
|---|---|---|---|
| | | instability-high (MSI-H) or mismatch repair deficient (dMMR) solid tumors that have progressed following prior treatment and who have no satisfactory alternative treatment options | companion diagnostic test |
| | | • Unresectable or metastatic tumor mutational burden-high (≥10 mutations/megabase) solid tumors, as determined by an FDA approved test, that have progressed following prior treatment and who have no satisfactory alternative treatment options. | FoundationOne CDx (Solid tumors TMB ≥ 10 mutations per megabase) |
| **Pertuzumab (Perjeta)** | HER2/neu receptor antagonist | • Use in combination with trastuzumab and docetaxel for treatment of patients with HER2-positive metastatic breast cancer who have not received prior anti-HER2 therapy or chemotherapy for metastatic disease.<br>• Use in combination with trastuzumab and chemotherapy as<br>o neoadjuvant treatment of patients with HER2-positive, locally advanced, inflammatory, or early stage breast cancer (either greater than 2 cm in diameter or node positive) as part of a complete treatment regimen for early breast cancer.<br>o adjuvant treatment of patients with HER2-positive early breast cancer at high risk of recurrence | HER2 FISH pharmDx Kit<br>HercepTest<br>FoundationOne CDx |
| **Talzenna (Talazoparib)** | PARP inhibitor | Adult patients with deleterious or suspected deleterious germline BRCA-mutated HER2-negative locally advanced or metastatic breast cancer. | BRACAnalysis CDx |
| **Trastuzumab (Herceptin)** | HER2/neu receptor antagonist | The treatment of HER2-overexpressing breast cancer | Bond Oracle HER2 IHC System<br>FoundationOne CDx<br>HER2 CISH pharmDx Kit<br>HER2 FISH pharmDx Kit<br>HercepTest<br>INFORM HER-2/neu<br>INFORM HER2 Dual ISH DNA Probe Cocktail<br>InSite Her-2/neu KITPathVysion HER-2 DNA Probe Kit<br>PATHWAY anti-Her2/neu (4B5) Rabbit Monoclonal Primary Antibody<br>SPOT-LIGHT HER2 CISH KitVENTANA HER2 Dual ISH |

| | |
|---|---|
| | DNA Probe Cocktail |

dMMR: mismatch repair deficient; FDA: U.S. Food & Drug Administration; HER2: human epidermal growth factor receptor 2; MSI-H: microsatellite instability-high; NTRK: neurotrophic-tropomyosin receptor kinase; PD-L1: programmed death-ligand 1 ; PIK3CA: phosphatidylinositol 3-kinase catalytic alpha polypeptide; TNBC: triple-negative breast cancer
Sources: [4,5,]

## Rationale

### Background

#### *PIK3CA* Testing

Alterations in the protein coding gene *PIK3CA* (Phosphatidylinositol-4,5-Bisphosphate 3-Kinase Catalytic Subunit Alpha) occur in approximately 40% of patients with HR-positive, HER2-negative breast cancer.

#### *NTRK* Gene Fusions

Neurotrophic-tropomyosin receptor kinase *(NTRK)* gene fusions encode tropomyosin receptor kinase fusion proteins that act as oncogenic drivers for solid tumors including lung, salivary gland, thyroid, and sarcoma. *NTRK* gene fusion findings might be more highly associated with rare breast cancer subtypes (e.g. secretory carcinoma).[1,].

#### Programmed Cell Death Ligand Protein-1

Programmed death ligand-1 (PD-L1) is a transmembrane protein expressed on the surface of multiple tissue types, including many tumor cells. Blocking the PD-L1 protein may prevent cancer cells from inactivating T cells.

#### Mismatch Repair Deficiency/Microsatellite Instability

Mismatch repair deficiency (dMMR) and high levels of microsatellite instability (MSI-H) describe cells that have alterations in certain genes involved in correcting errors made when DNA is replicated. dMMR tumors are characterized by a high tumor mutational load and potential responsiveness to anti-PD-L1-immunotherapy. MMR deficiency is most common in colorectal cancer, other types of gastrointestinal cancer, and endometrial cancer, but it may also be found in other cancers including breast cancer. Microsatellite instability testing is generally performed using polymerase chain reaction (PCR) for 5 biomarkers, although other biomarker panels and next generation sequencing are sometimes performed. High microsatellite instability is defined as 2 or more of the 5 biomarkers showing instability or more than 30% of the tested biomarkers showing instability depending on what panel is used. Microsatellite instability testing is generally paired with immunohistochemistry (IHC) assessing lack of protein expression from 4 DNA mismatch repair genes thereby reflecting dMMR.[2,]

#### Tumor Mutational Burden

Tumor mutational burden (TMB), a measure of gene mutations within cancer cells, is an emerging biomarker of outcomes with immunotherapy in multiple tumor types. Initially, assessments of TMB involved whole exome sequencing (WES). More recently, targeted next generation sequencing (NGS) panels are being adapted to estimate TMB. Currently FoundationOne CDx is the only U.S. Food and Drug Administration (FDA) approved panel for estimating TMB, but others are in development.[3,]

#### Circulating Tumor DNA

Normal and tumor cells release small fragments of DNA into the blood, which is referred to as cell-free DNA. Cell-free DNA from nonmalignant cells is released by apoptosis. Most cell-free tumor DNA is derived from apoptotic and/or necrotic tumor cells, either from the primary tumor, metastases, or CTCs.1, Unlike apoptosis, necrosis is considered a pathologic process and generates larger DNA fragments due to incomplete and random digestion of genomic DNA. The

length or integrity of the circulating DNA can potentially distinguish between apoptotic and necrotic origin. Circulating tumor DNA can be used for genomic characterization of the tumor.

**Circulating Tumor Cells**
Intact CTCs are released from a primary tumor and/or a metastatic site into the bloodstream. The half-life of a CTC in the bloodstream is short (1-2 hours), and CTCs are cleared through extravasation into secondary organs. Most assays detect CTCs through the use of surface epithelial markers such as EpCAM and cytokeratins. The primary reason for in detecting CTCs is prognostic, through quantification of circulating levels.

**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. Evidence reviews assess the evidence on whether a test is clinically valid and clinically useful. Technical reliability is outside the scope of these reviews, and credible information on technical reliability is available from other sources.

**Biomarker Testing Using Tissue Biopsy to Select Targeted Treatment**
**Clinical Context and Test Purpose**
Breast cancer treatment selection is informed by tumor type, grade, stage, patient performance status and preference, prior treatments, and the molecular characteristics of the tumor such as the presence of driver mutations. One purpose of biomarker testing of patients who have advanced cancer is to inform a decision regarding treatment selection (e.g., whether to select a targeted treatment or standard treatment).

The question addressed in this evidence review is: Does biomarker testing of tumor tissue for *PIK3CA*, *NTRK* gene fusions, PD-L1, MSI-H/dMMR, or TMB improve the net health outcome in individuals with breast cancer?

The following PICO was used to select literature to inform this review.

***Populations***
The relevant population of interest is patients with advanced or metastatic breast cancer for whom the selection of treatment depends on the molecular characterization of the tumor. The setting of interest is oncology care.

***Interventions***
The technologies being considered are testing for *PIK3CA*, *NTRK* gene fusions, PD-L1, MSI-H/dMMR, or TMB using tissue biopsy.

***Comparators***
Decisions about treatment in advanced breast cancer are based on clinical characteristics.

***Outcomes***
The general outcomes of interest in oncology are overall survival, disease-specific survival, quality of life, treatment-related mortality and morbidity.

Beneficial outcomes resulting from a true-positive test result are prolonged survival, reduced toxicity, and improved QOL associated with receiving a more effective targeted therapy. Beneficial outcomes from a true negative result are prolonged survival associated with receiving chemotherapy in those without driver mutations.

Harmful outcomes resulting from a false-negative test result include shorter survival from receiving less effective and more cytotoxic chemotherapy in those with driver mutations; possible harmful outcomes resulting from a false-positive test result are a shorter survival from receiving potentially ineffective targeted treatment and delay in initiation of chemotherapy in those without driver mutations.

The overall response rate (ORR) may be used as a surrogate endpoint reasonably likely to predict clinical benefit in patients with refractory solid tumors. ORR can be measured by the proportion of patients with best overall confirmed response of complete response) or partial response by the Response Evaluation Criteria in Solid Tumors, version 1.1 (RECIST 1.1),[6] or Response Assessment in Neuro-Oncology criteria,[7] as appropriate by a blinded and independent adjudication committee.

There are clearly defined quantitative thresholds for the follow-up of patients in oncology trials. A general rule is a continuation of treatment until disease progression or unacceptable toxicity. Long-term follow-up outside of a study setting is conducted to determine survival status. The duration of follow-up for the outcomes of interest is 6 months and 1 year.

**Study Selection Criteria**
Methodologically credible studies were selected using the following principles:

- To assess efficacy outcomes, comparative controlled prospective trials were sought, with a preference for RCTs;
- In the absence of such trials, comparative observational studies were sought, with a preference for prospective studies.
- To assess long-term outcomes and adverse events, single-arm studies that capture longer periods of follow-up and/or larger populations were sought.
- Studies with duplicative or overlapping populations were excluded.

The evidence is presented below by biomarker (*PIK3CA*, *NTRK*, PD-L1, MIS-H/dMMR, TMB*)* and by recommended therapy.

**PIK3CA**
**Companion Diagnostic Tests**
U.S. Food and Drug Administration (FDA) approved companion diagnostic tests for alpelisib in patients with *PIK3CA*-mutated breast cancer include both tissue-based and liquid biopsy assays (see Table 1). These tests are approved to measure 11 variants in the *PIK3CA* gene.

**Randomized Controlled Trial**
Andre et al (2019) reported results of SOLAR-1 (Clinical Studies of Alpelisib in Breast Cancer 1), a phase 3 trial to evaluate alpelisib plus fulvestrant in patients with HR-positive, HER2-negative advanced breast cancer who had received endocrine therapy previously.[8] Patients were enrolled into 2 cohorts based on tumor-mutation status (PIK3CA-mutated vs not PIK3CA-mutated) and randomly assigned within cohorts to receive oral alpelisib plus fulvestrant or placebo plus fulvestrant. *PIK3CA* status was determined with the use of a tumor-tissue sample, and patients had to have adequate tumor tissue for central analysis of *PIK3CA* mutational status. The primary end point was progression-free survival in the cohort of patients with *PIK3CA*-mutated cancer.

Among patients with *PIK3CA*-positive tumors who received targeted therapy, PFS was 11.0 months (95% CI, 7.5 to 14.5), compared to 5.7 months (95% CI, 3.7 to 7.4) in *PIK3CA*-positive patients who received standard care (HR 0.65; 95% CI, 0.50 to 0.85). In contrast, the hazard ratio

for PFS in the cohort without *PIK3CA*-mutated cancer was not significantly different for the active vs placebo groups.

**Table 2. RCT of Alpelisib in Patients with *PIK3CA*-Mutated Breast Cancer- Characteristics**

| Study | Countries | Sites | Dates | Participants | Interventions | | Endpoints | Median Duration of followup |
|---|---|---|---|---|---|---|---|---|
| | | | | | ***Active*** | ***Comparator*** | | |
| **Andre et al (2019)[8]. SOLAR-1 NCT02437318** | Multiple, US, Asia, Europe (N=34) | 198 | 2015 - 2018 | Men and postmenopausal women with HR-positive, HER2-negative advanced breast cancer, eligible to receive further endocrine therapy after relapse or progression, and receiving or had received aromatase inhibitor treatment in the context of neoadjuvant or adjuvant therapy or for advanced disease. | Alpelisib plus fulvestrant n=169 | Placebo plus fulvestrant n=172 | Primary: PFS in the cohort of patients with *PIK3CA*-mutated cancer. Secondary: OS (not reported in the primary publication), overall response, clinical benefit (complete or partial response or stable disease for >6 months), safety | 20.0 months (10.7 to 33.3) |

N: sample size; PIK3CA: phosphatidylinositol 3-kinase catalytic alpha polypeptide; RCT: randomized controlled trial; SOLAR-1: Study Assessing the Efficacy and Safety of Alpelisib Plus Fulvestrant in Men and Postmenopausal Women With Advanced Breast Cancer Which Progressed on or After Aromatase Inhibitor Treatment

**Table 3. RCT of Alpelisib in Patients with *PIK3CA*-Mutated Breast Cancer- Results**

| Study | PFS (95% CI) | PFS at 12 months (95% CI) | Overall Response | Clinical Benefit | Adverse events- Grade 3 or 4 |
|---|---|---|---|---|---|
| **Andre et al (2019)[8]. SOLAR-1NCT02437318** | | | | | |
| **N analyzed** | 341 | 341 | 341 | 341 | 571 |
| **Targeted therapy** | 11.0 months (95% CI, 7.5 to 14.5) | 46.3% | 45/169<br>26.6% (20.1 to 34.0) | 45/169<br>26.6 (20.1 to 34.0) | Serious AEs: 34.9%<br>Hyperglycemia:36.6%<br>Rash: 9.9%<br>Maculopapular rash: 8.8%<br>Diarrhea: 6.7%<br>Discontinuation due to AEs: 25.0%<br>Death: 2.5% |
| **Standard care** | 5.7 months (95% CI, | 32.9% | 22/172<br>12.8 (8.2 to 18.7) | 22/172<br>12.8 (8.2 to 18.7) | Serious AEs: 16.7%<br>Hyperglycemia: 0.7%<br>Rash: 0.3%<br>Maculopapular rash: |

| | | |
|---|---|---|
| | 3.7 to 7.4) | 0.3%<br>Diarrhea: 0.3%<br>Discontinuation due to AEs:4.2%<br>Death: 4.2% |
| **HR (95% CI)** | 0.65; 95% CI, 0.50 to 0.85 | |
| **p** | <.001 | |

CI: confidence interval; HR: hazard ratio; N: sample size; PFS: progression-free survival; PIK3CA: phosphatidylinositol 3-kinase catalytic alpha polypeptide; RCT: randomized controlled trial; SOLAR-1: Study Assessing the Efficacy and Safety of Alpelisib Plus Fulvestrant in Men and Postmenopausal Women With Advanced Breast Cancer Which Progressed on or After Aromatase Inhibitor Treatment

**Section Summary: *PIK3CA* Testing**
In a randomized, placebo-controlled trial of alpelisib compared to placebo in men and postmenopausal women with advanced breast cancer who had previously received endocrine therapy, PFS was longer among patients with *PIK3CA*-positive tumors who received targeted therapy, PFS was 11.0 months (95% CI, 7.5 to 14.5), compared to 5.7 months (95% CI, 3.7 to 7.4) in *PIK3CA*-positive patients who received standard care. In contrast, the hazard ratio for PFS in the cohort without *PIK3CA*-mutated cancer was not significantly different for the active vs placebo groups. The overall response rate was higher in patients with *PIK3CA*-positive tumors compared to the rate in the standard care group (26.6% [95% CI [20.1 to 34.0] vs 12.8% [8.2-18.7%]), with an acceptable side effect profile.

***NTRK* Gene Fusions**
**Companion Diagnostic Tests**
There is currently no FDA approved companion diagnostic test for entrectinib. FoundationOne CDX is an approved companion diagnostic test for larotrectinib.

**Nonrandomized Trials of Targeted Treatment**
***Entrectinib***
Doebele et al (2020) reported an analysis of data from 3 Phase 1-2 trials of entrectinib in patients with *NTRK*-fusion solid tumors (Table 4).[9] Of 54 patients included in the analysis, 6 had breast cancer (11%). Patients were assessed for eligibility for the 3 trials using either local molecular profiling or central RNA-based next-generation sequencing to test for the presence of *NTRK* fusions. The primary endpoints were objective response and duration of response. PFS and OS were secondary endpoints.

Of the total cohort of 54 patients, 31 had an objective response (57%; 95% CI 43.2–70.8) (Table 5). Four patients (7%) had a complete response and 27 a partial response (50%). Responses were recorded in all tumor types, including 5 (83%; 36–100) of 6 patients with breast cancer. Median PFS for the full cohort was 11 months (95% CI 8.0–1) and median overall survival was 21 months (95% CI 14.9 to not estimable). There were 7 serious treatment-related adverse events (10%), and 3 (4%) patients discontinued due to a treatment-related adverse event.

**Table 4. Entrectinib in *NTRK*-Fusion-Positive Solid Tumors - Study Characteristics**

| Studies | Design | Countries | Sites | Dates | Participants | Intervention | Endpoints |
|---|---|---|---|---|---|---|---|
| **Doebele et al (2020)[9]; STARTRK-1 (NCT02097810); STARTRK-2 (NCT02568267); ALKA-372–001 (EudraCT,** | Phase 1 (STARTRK-1 and ALKA) and Phase 2 (STARTRK-2) | STARTRK-1: US, Spain, Korea STARTRK-2: multiple (N=15) | STARTRK1:10 sites STARTRK-2: 150 sites ALKA: 2 sites | STARTRK-1: August 2014-May 2018 STARTRK-2: November 2015-ongoing | 54 adults with metastatic or locally advanced NTRK fusion-positive solid | Entrectinib | Primary: objective response, duration of response Secondary: PFS, OS, clinical benefit |

| | | | | |
|---|---|---|---|---|
| **2012–000148–88)** | ALKA: Italy | ALKA: October 2012-March 2018 | tumors; included patients with 10 different tumor types and 19 different histologies (11%) had breast tumors | rate. time to CNS progression, and safety. |

CNS: central nervous system; NTRK: neurotrophic-tropomyosin receptor kinase; OS: overall survival; PFS: progression-free survival; STARTRK-1: Study of Oral RXDX-101 in Adult Patients with Locally Advanced or Metastatic Cancer Targeting NTRK1, NTRK2, NTRK3, ROS1, or ALK Molecular Alterations; STARTRK-2: Basket Study of Entrectinib (RXDX-101) for the Treatment of Patients With Solid Tumors Harboring NTRK 1/2/3 (Trk A/B/C), ROS1, or ALK Gene Rearrangements (Fusions)

**Table 5. Entrectinib in *NTRK*-Fusion-Positive Solid Tumors - Study Results**

| Studies | Objective Response | Duration of Response | Median PFS | Median OS | Adverse Events |
|---|---|---|---|---|---|
| **Doebele et al (2020)[9]. STARTRK-1 (NCT02097810); STARTRK-2 (NCT02568267); ALKA-372–001 (EudraCT, 2012–000148–88)** | | | | | |
| **N analyzed** | 54 | 54 | 54 | 54 | 68 |
| | 31/54 (57%; 95% CI 43.2–70.8)<br>• 4 (7%) complete response<br>• 27 (50%) partial response.<br>Responses were recorded in all tumor types, including 5 (83%; 36–100) of 6 patients with breast cancer, | 10 months (95% CI 7.1 to not estimable) | 11 months (95% CI 8.0–1) | 21 months (95% CI 14.9 to not estimable) | 7 serious treatment-related adverse events (10%) 3 (4%) patients discontinued due to a treatment-related adverse events deaths (9%) |

CI: confidence interval; N: sample size; NTRK: neurotrophic-tropomyosin receptor kinase; OS: overall survival; PFS: progression-free survival; STARTRK-1: Study of Oral RXDX-101 in Adult Patients with Locally Advanced or Metastatic Cancer Targeting NTRK1, NTRK2, NTRK3, ROS1, or ALK Molecular Alterations; STARTRK-2: Basket Study of Entrectinib (RXDX-101) for the Treatment of Patients With Solid Tumors Harboring NTRK 1/2/3 (Trk A/B/C), ROS1, or ALK Gene Rearrangements (Fusions)

***Larotrectinib***

Hong et al (2020) reported an analysis of 3 phase 1-2 trials of larotrectinib in patients with *NTRK*-fusion positive solid tumors in adults and children.[10]

*NTRK* fusions were identified by next-generation sequencing, according to the procedures and analytic pipelines established by each laboratory, or by fluorescence in situ hybridization.

The trials included adults and children with 17 different solid tumors. Five patients had breast cancer (3%).

These results were consistent with a preliminary analysis of data from these trials reported by Drilon et al in 2018. [11,]

**Table 6. Larotrectinib in *NTRK*-Fusion-Positive Solid Tumors - Study Characteristics**

| Study | Dates | Participants | Intervention | Endpoints |
|---|---|---|---|---|
| **Hong et al (2020)[10,] NCT02122913, NCT02637687, NCT02576431** | May 2014-Feb 2019 | 159 adults and children with locally advanced or metastatic NTRK-fusion positive solid tumors.5 (3%) with breast cancer | Larotrectinib | Primary: Objective response, as assessed by an independent radiology review committee Secondary: Overall response according to the investigator's assessment, duration of response, PFS, and safety. |

NTRK: neurotrophic-tropomyosin receptor kinase; PFS: progression-free survival

**Table 7. Larotrectinib in *NTRK*-Fusion-Positive Solid Tumors - Study Results**

| Study | Overall response | Duration of response | PFS | OS | Adverse events |
|---|---|---|---|---|---|
| **Hong et al (2020)[10,]NCT02122913, NCT02637687, NCT02576431** | | | | | |
| **N analyzed** | 153 | 108 | 159 | 159 | 260 (expanded safety population) |
| **Targeted therapy** | Overall: 79% (95% CI 72 to 85)Complete: 16% of the patients (24 patients) had a complete response, Partial: 63% (97) had a partial response, Stable disease: 12% (19) had stable disease, Progressive disease: 6% (9) had progressive diseaseNot evaluated due to early withdrawal for clinical deterioration: 3% (4)Patients with breast cancer (n=4): 3 (75%; 19%-99%) | Median 35·2 months (95% CI 22.8–NE). At 12 months 80% (95% CI 71–89) NE for patients with breast cancer | Median: 28·3 months (95% CI 22·1–NE) At 12 months: 67% (95% CI 58–76) | Median: 44·4 months (95% CI 36·5–NE) Proportion surviving at 12 months: 88% (95% CI 83-94) | 23 deaths (14%) at median followup of 13.9 months The most common grade 3 or worse treatment-emergent adverse events (regardless of attribution) were anaemia (25 [10%] of 260 patients) and decreased neutrophil count (14 [5%]; table 4). The most common treatment-emergent serious adverse events were pneumonia (6 [2%] of 260 patients), |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | pyrexia (6 [2%]), abdominal pain (5 [2%]), and diarrhoea (5 [2%]); |

CI: confidence interval; N: sample size; NE: not estimable; NTRK: neurotrophic-tropomyosin receptor kinase; OS: overall survival; PFS: progression-free survival; PFS: progression-free survival

**Section Summary: *NTRK* Gene Fusion Testing**
In an analysis of 159 patients with *NTRK*-fusion positive solid tumors who received larotrectinib, including 5 patients with breast tumors, the overall response rate was 79% (95% CI 72-85). The median PFS was 28.3 months (95% CI 22.1 to not estimable), and 67% of patients were progression-free at 12 months (95% CI 58–76). In an integrated analysis of 3 phase 1-2 trials in 54 patients with *NTRK*-positive solid tumors who received entrectinib, 6 of whom had breast cancer, the overall response rate was 57% (95% CI 43.2–70.8). At data cutoff, 16 (30%) of 54 patients had died, and the estimated median overall survival was 21 months (95% CI 14.9 to not estimable). Responses were observed regardless of tumor type or age of the patient.

**PD-L1 Testing**
**FDA Companion Diagnostic Tests**
VENTANA PD-L1(SP142) Assay is an approved companion diagnostic test to select patients with triple negative breast cancer for treatment with atezolizumab.

PD-L1 IHC 22C3 pharmDx is an approved companion diagnostic test to select patients with triple negative breast cancer for treatment with pembrolizumab.

**Randomized and Nonrandomized Trials of Immunotherapy**
***Atezolizumab***
Schmid et al (2018) reported results of a randomized, placebo-controlled trial of atezolizumab in combination with nab-paclitaxel for patients with metastatic or unresectable triple-negative breast cancer and PD-L1-positive tumors (defined as expression on ≥1% of tumor-infiltrating immune cells).[12] PFS was longer in the group of PD-L1-positive patients who received targeted treatment, compared to those who received placebo (Table 9).

**Table 8. RCT of Atezolizumab plus Nab-Paclitaxel in Patients with PD-L1-Positive Triple Negative Breast Cancer - Characteristics**

| Study | Countries | Sites | Dates | Participants | Interventions | | Endpoints |
|---|---|---|---|---|---|---|---|
| | | | | | Active | Comparator | |
| **Schmid et al 2018[12] IMpassion130NCT02425891** | 41 (Europe US, Canada, Asia, Latin America, Australia) | 246 | Jun 2015-May 2017 | Patients with metastatic or unresectable locally advanced triple-negative breast cancer with PD-L1–positive tumors (expression on | n=185Atezolizumab + Nab-Paclitaxel | n=184Placebo + Nab-Paclitaxel | Primary: Investigator-assessed PFS and OS. Secondary: Rate and duration of objective response, safety |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | | | tumor-infiltrating immune cells ≥1%) | | | | |

Impassion130: A Study of Atezolizumab in Combination with Nab-Paclitaxel Compared with Placebo with Nab-Paclitaxel for Participants with Previously Untreated Metastatic Triple-Negative Breast Cancer; n: sample size; OS: overall survival; PD-L1: programmed death-ligand 1 ; PFS: progression-free survival; RCT: randomized controlled trial

**Table 9. RCT of Atezolizumab plus Nab-Paclitaxel in Patients with PD-L1-Positive Triple Negative Breast Cancer - Results**

| Study | Median PFS (95% CI) | PFS st 12 months | Median OS | 2-Year Rate of OS | Objective response | Median Duration of Response | Adverse events |
|---|---|---|---|---|---|---|---|
| **Schmid et al 2018[12], IMpassion130 NCT02425891** | | | | | | | |
| **Number analyzed** | 369 | 369 | 369 | 369 | 368 | | |
| **Targeted therapy** | 7.5 months (6.7-9.2) | 29.1% | 25.0 months (22.6-NE) | 53.5 (42.3-64.6) | 58.9 (51.5–66.1) | 8.5 months (7.3–9.7) | Deaths: 64/185 (34.6%) |
| **Standard care** | 5.0 months (3.8-5.6) | 16.4% | 15.5 month (13.1-19.4) | 36.6 (26.4-46.7) | 42.6 (35.4–50.1) | 5.5 months (3.7–7.1) | Deaths: 88/184 (47.8%) |
| **HR (95% CI)** | 0.62; (95% CI, 0.49 to 0.78) | | 0.62 (95% CI, 0.45–0.86) | | 1.96 (1.29–2.98) | 0.60 (0.43–0.86) | |

CI: confidence interval; HR: hazard ratio; Impassion130: A Study of Atezolizumab in Combination with Nab-Paclitaxel Compared with Placebo with Nab-Paclitaxel for Participants with Previously Untreated Metastatic Triple-Negative Breast Cancer ; NE: not estimable; OS: overall survival; PD-L1: programmed death-ligand 1; PFS: progression-free survival; RCT: randomized controlled trial

***Pembrolizumab***

Two nonrandomized, single-arm trials reported outcomes in a total of 111 patients with PD-L1 positive TNBC treated with pembrolizumab (Tables 10 and 11).[13,14]

**Table 10. Pembrolizumab in Patients with PD-L1-Positive Triple Negative Breast Cancer - Study Characteristics**

| Study | Design | Participants | Intervention | Endpoints |
|---|---|---|---|---|
| **Adams et al (2019)[13], KEYNOTE-086 NCT02447003** | Nonrandomized, multicohort, phase 2 | 84 patients with metastatic triple-negative breast cancer86.9% received prior (neo)adjuvant therapy; none had prior prior systemic therapy for metastatic disease | Pembrolizumab monotherapy | Primary: Safety Secondary: Objective response, disease control rate, duration of response, PFS, OS |
| **Nanda et al 2016[14], KEYNOTE-012 NCT01848834** | Nonrandomized, multicohort, phase Ib | 27 Patients with recurrent or metastatic PD-L1 positive triple-negative breast cancer. Most were heavily pretreated, having received therapy in both the early and advanced disease settings. | Pembrolizumab monotherapy | Primary: OR : defined as percentage of patients with a best overall response of complete response or partial response Secondary: PFS, duration of response, OS |

OS: overall survival; PD-L1: programmed death-ligand 1; PFS: progression-free survival

**Table 11. Pembrolizumab in Patients with PD-L1-Positive Triple Negative Breast Cancer - Study Results**

| Study | Response | Median PFS | Duration of Response | OS | Adverse Events |
|---|---|---|---|---|---|
| **Adams et al (2019)[13], NCT02447003** | | | | | |
| **N analyzed** | 84 | 84 | | | 84 |
| **Targeted therapy** | Objective response rate: 21.4% (95% CI 13.9–31.4) | Median: 2.1 months (95% CI 2.0–2.2) Rate at 6 months: 27.0% | Median: 10.4 months (range 4.2 to 19.2+) | Median 18.0 months (95% CI 12.9–23.0) 6-month rate 81.0% 12-month rate: 61.7% | 53 (63.1%) patients experienced 1 or more treatment-related AE,8 (9.5%) with 1 or more grade 3 event. No grade 4 events, no AEs that led to death1 (1.2%) discontinued due to AEs. Most common treatment-related AEs were fatigue (26.2%), nausea (13.1%), and diarrhea (11.9%)43 deaths (51.2%) |
| **Nanda et al 2016[14]KEYNOTE-012NCT01848834** | | | | | |
| **N Analyzed** | 27 | 22 | | | |
| **C** | Overall response rate:18.5% (95% CI, 6.3 to 38.1) Complete response: 1 (3.7%) Partial response: 4 (14.8%)PD 13 (48.1%) | Median 1.9 months (95% CI, 1.7 to 5.5), 6 months PFS: 24.4% | Median not yet reached (range 15.0 to ≥47.3 weeks) | Median : 11.2 months (95% CI, 5.3 to [not reached]) 6 month rate: :66.7%12-month OS: 43.1% | 56.3% of patients experienced at least one treatment-related toxicity, including 15.6% who experienced at least one grade 3 to 5 event. One patient died as a result of disseminated intravascular coagulation(DIC) accompanied by grade 4 decreased blood fibrinogen, both of which were considered by the investigator to be treatment related. |

AE: adverse events; CI: confidence interval; OS: overall survival; PD-L1: programmed death-ligand 1; PFS: progression-free survival

**Section Summary: PD-L1 Testing**

In a placebo controlled trial of atezolizumab in combination with nab-paclitaxel for patients with PD-L1 positive TNBC, median PFS (HR 0.62; 95% CI, 0.49 to 0.78) and OS 0.62 (95% CI, 0.45–0.86) were longer among patients who received the targeted immunotherapy. In 2 nonrandomized trials of pembrolizumab for patients with PD-L1 positive TNBC, the objective response rate was 21.4% (95% CI, 13.9 to 31.4) and 18.5% (95% CI, 6.3 to 38.1).

### MSI-H/dMMR Testing

#### FDA Companion Diagnostic Tests

There is no FDA approved test for the detection of MSI-H or dMMR. In clinical trials, the identification of MSI-H or dMMR tumor status for the majority of patients (135/149) was prospectively determined using local laboratory-developed, polymerase chain reaction (PCR) tests for MSI-H status or immunohistochemistry (IHC) tests for dMMR.

#### Nonrandomized Trials of Immunotherapy

***Pembrolizumab***

Marabelle et al.(2020) reported results of a phase 2 trial of pembrolizumab in 233 previously treated patients with MSI-H solid tumors (Tables 12 and 13), 5 of whom had breast cancer.[15.] The overall response rate, the primary outcome, was 34.3% (95% CI, 28.3% to 40.8%). Median PFS was 4.1 months (95% CI, 2.4 to 4.9 months) and median OS was 23.5 months (95% CI, 13.5 months to not reached). Treatment-related adverse events occurred in 151 patients (64.8%). Earlier, Le et al (2015) reported on a small (N = 41) phase 2 trial that compared response to pembrolizumab in patients with solid tumors that did or did not have mismatch repair.[16.] Most of the patients had colorectal cancer, but a cohort of 9 patients with dMMR tumors that were not colorectal was included. In the full cohort, mismatch-repair status predicted clinical benefit of pembrolizumab, and patients with dMMR noncolorectal cancer had responses similar to those of patients with dMMR colorectal cancer.

**Table 12. Pembrolizumab in Patients with MSI-H/dMMR-Positive Solid Tumors - Study Characteristics**

| Study | Countries | Sites | Dates | Design | Participants | Intervention | Endpoints |
|---|---|---|---|---|---|---|---|
| **Marabelle et al (2020) [15.]KEYNOTE-158NCT02628067** | Multiple (N=21) | 81 | Feb 2016 -May 2018 | Nonrandomized, open-label, multisite phase 2 | 233 patients 18 years or older with unresectable and/or metastatic incurable noncolorectal solid tumor with disease progression on or intolerance to prior standard therapy.<br>27 tumor types<br>5 patients had breast cancer (2.1%) | Pembrolizumab | Primary: Overall response rate<br>Secondary: duration of response, PFS, OS, safety |

dMMR: mismatch repair deficient; MSI-H: microsatellite instability-high; N: sample size; OS: overall survival; PFS: progression-free survival

**Table 13. Pembrolizumab in Patients with MSI-H/dMMR-Positive Solid Tumors - Study Results**

| Study | Response | Duration of Response | PFS | OS | Adverse events |
|---|---|---|---|---|---|
| *Marabelle et al (2020)[15.]*<br>*KEYNOTE-158*<br>*NCT02628067* | | | | | |
| **N analyzed** | 233 | | | | 233 |

| | | | | | |
|---|---|---|---|---|---|
| **Targeted therapy** | Overall response rate: 34.3% (95% CI, 28.3% to 40.8%)<br>Complete: 23 (9.9%)<br>Partial: 57 (24.5%) | Median: not reached, range, 2.9 to 31.3+ months<br>Response 12 months or longer: 86.9%<br>24 months or longer: 77.6% | Median: 4.1 months (95% CI, 2.4 to 4.9 months)<br>12 months: 33.9%<br>24-months: 29.3% | Median: 23.5 months (95% CI, 13.5 months to not reached)<br>12 months: 60.7%<br>24-months: 48.9% | Overall, 151 patients (64.8%) had treatment-related adverse events and 34 (14.6%) had grade 3 to 5 treatment-related adverse events, one of which was grade 5 (pneumonia).<br><br>Eighteen patients (7.7%) had serious treatment-related adverse events, and 22 (9.4%) discontinued treatment because of a treatment-related adverse event<br><br>Deaths: 113 (48.5%) |

CI: confidence interval; dMMR: mismatch repair deficient; MSI-H: microsatellite instability-high; N: sample size; OS: overall survival; PFS: progression-free survival

**Section Summary: MSI-H/dMMR Testing**
In a phase 2 trial of pembrolizumab in 233 previously treated patients with MSI-H solid tumors, the overall response rate was 34.3% (95% CI, 28.3% to 40.8%). Median PFS was 4.1 months (95% CI, 2.4 to 4.9 months) and median OS was 23.5 months (95% CI, 13.5 months to not reached). Treatment-related adverse events occurred in 151 patients (64.8%).

**Tumor Mutational Burden**
**FDA Companion Diagnostic Test**
FoundationOne is an FDA approved companion diagnostic test to measure TMB in patients with solid tumors being considered for pembrolizumab treatment.
**Nonrandomized Trials**
Ott et al (2018) reported an exploratory analysis of the association between TMB and response to pembrolizumab. All patients in the study were PD-L1 positive.[17,]
Marabelle et al (2020) reported the association of high TMB to response to pembrolizumab in patients with solid tumors enrolled in a prespecified exploratory analysis of the KEYNOTE-158 study.[18,] High TMB was defined as >10 mutations per megabase according to the FoundationOne CDx panel. The proportion of patients with an objective response in the tTMB-high group was 29%. At a median follow-up of approximately 3 years, the median duration of response was not reached in the tTMB-high group and was 33·1 months in the non-tTMB-high group. Notably, TMB-high status was associated with improved response irrespective of PD-L1. Median PFS and OS did not differ between the high and non-high TMB groups. Objective responses were observed in 24 (35%; 95% CI 24–48) of 68 participants who had both tTMB-high status and PD-L1-positive tumours (ie, PD-L1 combined positive score of ≥1) and in 6 (21%; 8–40) of 29 participants who had tTMB-high status and PD-L1-negative tumors.

**Table 14. Association of TMB to Response to Pembrolizumab in Patients with Solid Tumors Enrolled in the KEYNOTE-158 Study**

| Study | Response | Median Duration of Response | Median PFS | Median OS (95% CI) | Adverse events |
|---|---|---|---|---|---|
| **Marabelle et al (2020)[18,] NCT02628067** | | | | | |

| | | | | | |
|---|---|---|---|---|---|
| **TMB ≥10 per megabase; N=102** | Objective Response: 29% (21-39%) Complete: 4% | Median not yet reached range 2·2+ to 34·8+ months | 2.1 months (95% CI 2·1–4·1) | Median: 11·7 months (95% CI 9·1–19·1) | Deaths: 69/102 (68%) |
| **TMB <10 per megabase; N=688** | Objective Response: 6% (5-8%) Complete: 2% | Median 33·1 months (4·0 to 35·7+) | 2·1 months (2·1–2·2) | 12·8 months (11·1–14·1) | 534/688 (78%) |

CI: confidence interval N: sample size; OS: overall survival; PFS: progression-free survival; TMB: tumor mutational burden

**Section Summary: Tumor Mutational Burden**
In a prespecified subgroup analysis of a nonrandomized trial of pembrolizumab in patients with various solid tumors, **o**bjective responses were observed in 24 (35%; 95% CI 24–48) of 68 participants who had both tTMB-high status and PD-L1-positive tumors and in 6 (21%; 8–40) of 29 participants who had tTMB-high status and PD-L1-negative tumors. In exploratory analyses, retrospective observational studies have reported an association between higher TMB and longer PFS and OS in patients receiving immunotherapy. These results need to be confirmed in additional, well-designed prospective studies.

**Circulating Tumor DNA Testing to Select Targeted Treatment**
**Clinical Context and Test Purpose**
The purpose of circulating tumor DNA testing in patients who have advanced or metastatic breast cancer is to inform a decision about selecting targeted treatment.
The question addressed in this evidence review is: Does biomarker testing using circulating tumor DNA improve the net health outcome in individuals with breast cancer?
The following PICO was used to select literature to inform this review.

***Populations***
The relevant population of interest is individuals with advanced or metastatic breast cancer..

***Interventions***
The test being considered is circulating tumor DNA testing.

***Comparators***
Tissue biopsy is used to make decisions about targeted treatment or immunotherapy for metastatic breast cancer.

***Outcomes***
Liquid biopsies are easier to obtain and less invasive than tissue biopsies. True-positive liquid biopsy test results lead to the initiation of appropriate treatment (e.g., targeted therapy) without a tissue biopsy. False-positive liquid biopsy test results lead to the initiation of inappropriate therapy, which could shorten progression-free survival.

In patients able to undergo a tissue biopsy, negative liquid biopsies reflex to tissue testing. In patients unable to undergo a tissue biopsy, a negative liquid biopsy result would not change empirical treatment. Therefore, health outcomes related to negative test results do not differ between liquid biopsy and tissue biopsy.

**Study Selection Criteria**
For the evaluation of clinical validity of the circulating tumor DNA test, studies that meet the following eligibility criteria were considered:

- Reported on the accuracy of the marketed version of the technology (including any algorithms used to calculate scores)
- Included a suitable reference standard (describe the reference standard)

- Patient/sample clinical characteristics were described
- Patient/sample selection criteria were described.

**Companion Diagnostic Test for Targeted Treatment**
FoundationOne Liquid is FDA approved as a companion diagnostic test for alpelisib (Piqray) for measuring 11 variants in the *PIK3CA* gene.

**Clinical Validity**
Woodhouse 2020 reported the clinical validity of FoundationOne liquid for detection of *PIK3CA* alterations through retrospective testing of plasma samples of patients enrolled in the SOLAR-1 trial.[19.]

All available plasma samples from patients collected at baseline prior to randomization into the SOLAR-1 clinical trial were tested with FoundationOne Liquid CDx, with results compared to tissue genotyping performing using the SOLAR-1 CTA. The positive predictive agreement and negative predictive agreement between FoundationOne Liquid CDx and the tissue-based CTA assay were 71.7% (95% CI 65.4%, 77.5%) and 100% (97.2%, 100%), respectively.

**Table 15. Clinical Validity of FoundationOne Liquid CDx to detect *PIK3CA* Alterations- Results**

| Study | Study Population | Reference Standard | Initial N | Final N | Excluded Samples | Prevalence of Condition | Clinical Validity(95% Confidence Interval) | |
|---|---|---|---|---|---|---|---|---|
| | | | | | | | ***PPA*** | ***NPA*** |
| **Woodhouse et al (2020)[19.]** | Plasma samples from advanced or metastatic HR-positive, HER2-negative breast cancer patients enrolled in the SOLAR-1 trial | Tumor tissue PCR-based clinical trial assay | 432 | 375 | 16 | 230 positive129 negative | 71.7% (95% CI 65.4%, 77.5%) | 100% (97.2%, 100%) |

CI: confidence interval; N: sample size; NPA: negative predictive agreement; PCR: polymerase chain reaction; PIK3CA: phosphatidylinositol 3-kinase catalytic alpha polypeptide; PPA: positive predictive agreement

**Clinical Utility**
In the SOLAR-1 trial (discussed above in the section on *PIK3CA* testing), the clinical efficacy of alpelisib in combination with fulvestrant for the FoundationOne Liquid CDx-positive population was demonstrated with an estimated 54% risk reduction in disease progression or death in the alpelisib plus fulvestrant arm compared to the placebo plus fulvestrant arm (HR = 0.46, 95% CI: 0.30, 0.70).[8.]

**Section Summary: Circulating Tumor DNA Testing**
Clinical validity of the FoundationOne Liquid CDx test was demonstrated through retrospective testing of plasma samples of patients enrolled in the SOLAR-1 trial. The positive predictive agreement and negative predictive agreement between FoundationOne Liquid CDx and the tissue-based assay were 71.7% (95% CI 65.4%, 77.5%) and 100% (97.2%, 100%), respectively. Among the circulating tumor DNA-positive population, there was an estimated 54% risk reduction in disease progression or death in the alpelisib plus fulvestrant arm compared to the placebo plus fulvestrant arm (HR = 0.46, 95% CI: 0.30, 0.70).

## Circulating Tumor Cell Testing to Select Targeted Treatment

### Clinical Context and Test Purpose

The purpose of testing circulating tumor cells in patients who have breast cancer is to inform a decision about selecting targeted treatment.
The question addressed in this evidence review is: Does CTC testing improve the net health outcome in individuals with breast cancer?

The following PICO was used to select literature to inform this review.

***Populations***
The relevant population of interest is individuals with recurrent or metastatic breast cancer

***Interventions***
The test being considered is CTC testing.
The primary reason for CTCs would be to aid in decision-making about alternative treatment. CTC testing has been proposed as a method to guide the choice between chemotherapy and endocrine therapy as first-line treatment, or to change early to an alternative chemotherapy regimen in patients for whom chemotherapy has failed to reduce CTCs.

***Comparators***
Decisions about first-line treatment and alternative treatments in metastatic breast cancer are based on clinical evaluation and biopsy.

***Outcomes***
The general outcomes of interest in oncology are overall survival, disease-specific survival, quality of life, treatment-related mortality and morbidity.

Follow-up at 6-12 months is of interest to monitor outcomes.

### Study Selection Criteria

For the evaluation of clinical validity of CTC test, studies that meet the following eligibility criteria were considered:

- Reported on the accuracy of the marketed version of the technology (including any algorithms used to calculate scores)
- Included a suitable reference standard (describe the reference standard)
- Patient/sample clinical characteristics were described
- Patient/sample selection criteria were described.

### Clinical Validity

Systematic reviews and meta-analyses have described an association between CTCs and poor prognosis in metastatic breast cancer. [20,]

### Clinical Utility

#### Randomized Controlled Trials

Two RCTs have evaluated the clinical utility of using CTC to guide treatment decisions in patients with metastatic breast cancer.

Smerage et al (2014) reported on the results of an RCT of patients with metastatic breast cancer and persistently increased CTC levels to test whether changing chemotherapy after 1 cycle of first-line therapy could improve overall survival.[21,]. Level of CTCs were enumerated using the CellSearch system. Five or more CTCs per 7.5 mL WB was considered an increased level, and it served as the cut point for separation of favorable versus unfavorable prognosis. Patients who did not have increased CTC levels at baseline remained on initial therapy until progression (arm A), patients with initially increased CTC levels that decreased after 21 days of therapy remained on initial therapy (arm B), and patients with persistently increased CTC levels after 21 days of therapy were randomized to continue initial therapy (arm C1) or change to an alternative

chemotherapy (arm C2). There were 595 eligible and evaluable patients, 276 (46%) of whom did not have increased CTC levels (arm A). Of patients with initially increased CTC levels, 31 (10%) were not retested, 165 were assigned to arm B, and 123 were randomized to arms C1 or C2. There was no difference in median OS between arms C1 (10.7 months) and C2 12.5 months; p=0.98). CTC levels were strongly prognostic, with a median OS for arms A, B, and C (C1 and C2 combined) of 35 months, 23 months, and 13 months, respectively (p<0.001). While the trial showed the prognostic significance of CTCs in patients with metastatic breast cancer, changing to an alternative chemotherapeutic regimen did not improve outcomes in patients whose CTCs were not reduced after 1 cycle of first-line chemotherapy.

More recently, Bidard et al (2020) reported on a noninferiority trial comparing CTC-driven vs clinician driven first-line therapy choice in patients with metastatic breast cancer.[22] Median PFS was 15.5 months (95% CI, 12.7-17.3) in the CTC arm and 13.9 months (95% CI, 12.2-16.3) in the standard arm. The primary end point was met, with a hazard ratio of 0.94 (90% CI, 0.81-1.09).

**Table 17. RCTs of CTC-Guided Treatment in Breast Cancer- Characteristics**

| Study | Countries | Sites | Dates | Participants | Interventions | | Endpoints |
|---|---|---|---|---|---|---|---|
| | | | | | **Active** | **Comparator** | |
| **Smerage et al (2014)[21] NCT00382018** | | | Oct 2006-Mar 2012 | Women with histologically confirmed breast cancer and clinical and/or radiographic evidence of metastatic disease Persistent increased CTCs following 1 cycle of chemotherapy. | Changing chemotherapy after 1 cycle of first-line chemotherapy N=59 | Continued initial therapy N=64 | OS, PFS |
| **Bidard et al 2020)[22]** | France | 17 | Feb 2012-Jul 2016 | 778 women with hormone-receptor positive, HER2-negative metastatic breast | CTC-driven treatment choice N=391 | Clinician-driven treatment choice N=387 | PFS, OS, rate of treatment changes, AEs |

AEs: adverse events; CTC: circulating tumor cell; HER2: human epidermal growth factor receptor 2; N: sample size; OS: overall survival; PFS: progression-free survival; RCTs: randomized controlled trials

**Table 18. RCTs of CTC-Guided Treatment in Breast Cancer- Results**

| Study | OS | PFS |
|---|---|---|
| **Smerage et al (2014)[21]** | | |
| **N analyzed** | | |
| **CTC-Directed Treatment** | 12.5 months | 4.6 months |
| **Standard care** | 10.7 months | 3.5 months |
| **HR (95% CI)** | 1.00 (95% CI, 0.69 to 1.47) | 0.92 (95% CI, 0.64 to 1.32) |
| **p** | .98 | .64 |
| **Bidard et al 2020)[22]** | | |
| **N analyzed** | | |

| | |
|---|---|
| **CTC-directed treatment** | 15.5 months (12.7-17.3) |
| **Standard care** | 13.9 months (12.2-16.3) |
| **HR (95% CI)** | 0.94 (0.81 to 1.09) |

CI: confidence interval; CTC: circulating tumor cell; HR: hazard ratio; N: sample size; OS: overall survival; PFS: progression-free survival; RCTs: randomized controlled trials

**Section Summary: Circulating Tumor Cell Testing**
Systematic reviews and meta-analyses have described an association between CTCs and poor prognosis in metastatic breast cancer, but evidence that CTC-driven treatment improves health outcomes is lacking. One RCT found no improvement in OS or PFS with CTC-driven treatment (early switching to a different chemotherapy regimen) compared to continuing initial therapy. A second RCT found that CTC-driven first-line therapy was noninferior to clinician-driven therapy in previously untreated patients with metastatic breast cancer (hazard ratio for PFS 0.94; 95% CI 0.81 to 1.09).

**Summary of Evidence**
For individuals with hormone receptor-positive, HER2 negative advanced or metastatic breast cancer who receive *PIK3CA* gene testing to select targeted treatment, the evidence includes a randomized, placebo-controlled trial of alpelisib compared to placebo in men and postmenopausal women with advanced breast cancer who had previously received endocrine therapy. Relevant outcomes include overall survival, disease-specific survival, test validity, quality of life, and treatment-related morbidity. Among patients with *PIK3C*A-positive tumors who received targeted therapy, PFS was 11.0 months (95% CI, 7.5 to 14.5), compared to 5.7 months (95% CI, 3.7 to 7.4) in *PIK3CA*-positive patients who received standard care. In contrast, the hazard ratio for PFS in the cohort without PIK3CA-mutatied cancer was not significantly different for the active vs placebo groups. The overall response rate was higher in patients with *PIK3CA*-positive tumors compared to the rate in the standard care group (26.6% [95% CI [20.1- 34.0] vs 12.8% [8.2-18.7%]). The evidence is sufficient to determine that the technology results in an improvement in the net health outcomes.

For individuals with locally advanced or metastatic breast cancer being considered for immunotherapy who receive *NTRK* gene fusion testing, the evidence includes integrated analyses of nonrandomized trials of larotrectinib and entrectinib in patients with *NTRK*-fusion positive solid tumors. Relevant outcomes are OS, disease-specific survival, test validity, QOL, and treatment-related morbidity. In an analysis of 159 patients with *NTRK*-fusion positive solid tumors who received larotrectinib, including 5 patients with breast tumors, the overall response rate was 79% (95% CI 72 to 85). The median PFS was 28·3 months (95% CI 22.·1 to not estimable), and 67% of patients were progression-free at 12 months (95% CI 58–76). In an integrated analysis of 3 phase 1-2 trials in 54 patients with *NTRK*-positive solid tumors who received entrectinib, 6 of whom had breast cancer, the overall response rate was 57% (95% CI 43.2–70.8). At data cutoff, 16 (30%) of 54 patients had died, and the estimated median overall survival was 21 months (95% CI 14.9 to not estimable. Responses were observed regardless of tumor type or age of the patient. The evidence is sufficient to determine that the technology results in an improvement in the net health outcomes.

For individuals with recurrent, metastatic, or unresectable hormone receptor-negative, HER2 negative (triple negative) breast cancer being considered for immunotherapy who receive PD-L1 testing, the evidence includes a RCT of atezolizumab and nonrandomized trials of pembrolizumab. Relevant outcomes include overall survival, disease-specific survival, test validity, quality of life, and treatment-related morbidity. In a placebo controlled trial of atezolizumab in combination with nab-paclitaxel for patients with PD-L1 positive TNBC, median PFS (HR 0.62; 95% CI, 0.49 to 0.78) and OS 0.62 (95% CI, 0.45–0.86) were longer among patients who received the targeted immunotherapy. In 2 nonrandomized trials of pembrolizumab for patients with PD-L1 positive TNBC, the objective response rate was 21.4% (95% CI, 13.9 to 31.4) and 18.5% (95% CI, 6.3 to 38.1). The evidence is sufficient to determine that the technology results in an improvement in the net health outcomes.

For individuals with unresectable or metastatic breast cancer who are being considered for immunotherapy who receive MSI-H/dMMR testing, the evidence includes nonrandomized trials of pembrolizumab in patients with solid tumors. Relevant outcomes include overall survival, disease-specific survival, test validity, quality of life, and treatment-related morbidity. In a phase 2 trial of pembrolizumab in 233 previously treated patients with MSI-H solid tumors, the overall response rate was 34.3% (95% CI, 28.3% to 40.8%). Median PFS was 4.1 months (95% CI, 2.4 to 4.9 months) and median OS was 23.5 months (95% CI, 13.5 months to not reached). Treatment-related adverse events occurred in 151 patients (64.8%). The evidence is sufficient to determine that the technology results in an improvement in the net health outcomes.

For individuals with unresectable or metastatic breast cancer who are being considered for immunotherapy who receive tumor mutational burden (TMB) testing, the evidence includes prospective and retrospective subgroup analyses of nonrandomized trials. Relevant outcomes include overall survival, disease-specific survival, test validity, quality of life, and treatment-related morbidity. In a prespecified subgroup analysis of a nonrandomized trial of pembrolizumab in patients with various solid tumors, objective responses were observed in 24 (35%; 95% CI 24–48) of 68 participants who had both tTMB-high status and PD-L1-positive tumors and in 6 (21%; 8–40) of 29 participants who had tTMB-high status and PD-L1-negative tumors. In exploratory analyses, retrospective observational studies have reported an association between higher TMB and longer PFS and OS in patients receiving immunotherapy. These results need to be confirmed in additional, well-designed prospective studies. The evidence is insufficient to determine that the technology results in an improvement in the net health outcomes.

For individuals with hormone receptor-positive, HER2 negative advanced or metastatic breast cancer who receive circulating tumor DNA testing to select targeted treatment, the evidence includes a randomized, placebo-controlled trial of alpelisib compared to placebo in men and postmenopausal women with advanced breast cancer who had previously received endocrine therapy. Relevant outcomes include overall survival, disease-specific survival, test validity, quality of life, and treatment-related morbidity. Clinical validity of the FoundationOne Liquid CDx test was demonstrated through retrospective testing of plasma samples of patients enrolled in the SOLAR-1 trial. The positive predictive agreement and negative predictive agreement between FoundationOne Liquid CDx and the tissue-based assay were 71.7% (95% CI 65.4%, 77.5%) and 100% (97.2%, 100%), respectively. Among the circulating tumor DNA-positive population, there was an estimated 54% risk reduction in disease progression or death in the alpelisib plus fulvestrant arm compared to the placebo plus fulvestrant arm (HR = 0.46, 95% CI: 0.30, 0.70). The evidence is sufficient to determine that the technology results in an improvement in the net health outcomes.

For individuals with metastatic breast cancer who receive CTC testing to guide treatment decisions, the evidence includes randomized controlled trials, observational studies, and systematic reviews. Relevant outcomes include overall survival, disease-specific survival, test validity, quality of life, and treatment-related morbidity. Systematic reviews and meta-analyses have described an association between CTCs and poor prognosis in metastatic breast cancer, but evidence that CTC-driven treatment improves health outcomes is lacking. One RCT found no improvement in OS or PFS with CTC-driven treatment (early switching to a different chemotherapy regimen) compared to continuing initial therapy. A second RCT found that CTC-driven first-line therapy was noninferior to clinician-driven therapy in previously untreated patients with metastatic breast cancer (hazard ratio for PFS 0.94; 95% CI 0.81 to 1.09).The evidence is insufficient to determine that the technology results in an improvement in the net health outcomes.

**Supplemental Information**
**Professional Society Guidelines**

**National Comprehensive Cancer Network**

Table 19 summarizes National Comprehensive Cancer Network guidelines (v.6.2020) on biomarker testing for the biomarkers included in this policy.[1,] The guidelines state that the use of circulating tumor cells or circulating tumor DNA in metastatic breast cancer is not yet included in algorithms for disease assessment and monitoring. For patients being considered for treatment with alpelisib, testing for PIK3CA with either tissue or liquid biopsy is recommended (category of evidence 2A). The guidelines do not address TMB testing.

**Table 19. National Comprehensive Cancer Network Guidelines on Biomarker Testing for Targeted Treatment of Breast Cancer**

| Biomarker | Breast Cancer Subtype | FDA Approved Agents | Testing Recommendation | Targeted Therapy Category of Evidence | Targeted Therapy Category of Preference |
|---|---|---|---|---|---|
| **PIK3CA** | HR-positive/HER2-negative | Alpelisib + fulvestrant | For HR-positive/HER2-negative breast cancer, assess for PIK3CA mutations with tumor or liquid biopsy to identify candidates for alpelisib plus fulvestrant.PIK3CA mutation testing can be done on tumor tissue or ctDNA in peripheral blood (liquid biopsy). If liquid biopsy is negative, tumor tissue testing is recommended. | 1 | Preferred second-line therapy |
| **PD-L1 expression (≥1% on tumor-infiltrating immune cells)** | HR-negative/HER2-negative | Atezolizumab + albumin-bound paclitaxel | For triple-negative breast cancer, assess PD-L1 expression biomarker status on tumor-infiltrating immune cells to identify patients most likely to benefit from candidates for atezolizumab plus albumin-bound paclitaxel | 2A | Preferred |
| **NTRK fusion** | Any | Larotrectinib Entrectinib | No specific testing recommendation. If a patient with recurrent/stage IV breast cancer presents with a tumor with an NTRK fusion, treatment with an NTRK inhibitor is an option if no satisfactory alternative treatments exist or that have progressed following treatment, treatment with an NTRK inhibitor is an option | 2A | Useful in certain circumstances |
| **MSI-H/dMMR** | Any | Pembrolizumab | No specific testing recommendation. If a patient with recurrent/stage IV breast cancer has a tumor with a MSI-H/dMMR mutation, whose disease has progressed following prior treatments and no satisfactory alternative | 2A | Useful in certain circumstances |

| | | |
|---|---|---|
| | treatments exist, treatment with pembrolizumab is an option | |

Source: Adapted from National Comprehensive Cancer Network guidelines on Breast Cancer (v.6.2020)[1].

### Ongoing and Unpublished Clinical Trials

Some currently unpublished trials that might influence this review are listed in Table 20.

**Table 20. Summary of Key Trials**

| NCT No. | Trial Name | Planned Enrollment | Completion Date |
|---|---|---|---|
| *Ongoing* | | | |
| **NCT02889978**[a] | The Circulating Cell-free Genome Atlas Study | 15000 | Mar 2024 |
| **NCT02568267**[a] | An Open-Label, Multicenter, Global Phase 2 Basket Study of Entrectinib for the Treatment of Patients With Locally Advanced or Metastatic Solid Tumors That Harbor NTRK1/2/3, ROS1, or ALK Gene Rearrangements | 500 | Dec 2024 |
| **NCT04098640** | Molecular Profiling Using FoundationOne CDx in Young (<50 Years of Age) Patients With Metastatic Breast Cancer (ML41263) | 200 | Jul 2021 |
| **NCT04591431** | The Rome Trial From Histology to Target: the Road to Personalize Target Therapy and Immunotherapy | 384 | Aug 2024 |

NCT: national clinical trial.
[a] Denotes industry-sponsored or cosponsored trial.

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## Documentation for Clinical Review

**Please provide the following documentation:**

- History and physical and/or consultation notes including:
  - Clinical findings (i.e., pertinent symptoms and duration)
  - Current diagnoses and status (i.e., type of cancer, stage)
  - Family history, if applicable
  - Reason for test when applicable
  - Pertinent past procedural and surgical history (i.e., biopsies, resections, etc.)

- Pertinent past genetic tests (i.e., somatic/tumor or germline test results including but not limited to HER2, PD-L1, MSI, BRCA, etc.)

**Post Service (in addition to the above, please include the following):**

- Results/reports of tests performed
- Procedure report(s)

## 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 codes does not constitute or imply member coverage or provider reimbursement.*

| Type | Code | Description |
|---|---|---|
| **CPT®** | 0037U | Targeted genomic sequence analysis, solid organ neoplasm, DNA analysis of 324 genes, interrogation for sequence variants, gene copy number amplifications, gene rearrangements, microsatellite instability and tumor mutational burden (PLA for the Foundation One CDx™ (F1CDx®) test) |
| | 0048U | Oncology (solid organ neoplasia), DNA, targeted sequencing of protein-coding exons of 468 cancer-associated genes, including interrogation for somatic mutations and microsatellite instability, matched with normal specimens, utilizing formalin-fixed paraffin-embedded tumor tissue, report of clinically significant mutation(s) (PLA code for the MSK–IMPACT™ (Integrated Mutation Profiling of Actionable Cancer Targets), Memorial Sloan Kettering Cancer Center) |
| | 0155U | Oncology (breast cancer), DNA, PIK3CA (phosphatidylinositol-4,5-bisphosphate 3- kinase, catalytic subunit alpha) (e.g., breast cancer) gene analysis (i.e., p.C420R, p.E542K, p.E545A, p.E545D [g.1635G>T only], p.E545G, p.E545K, p.Q546E, p.Q546R, p.H1047L, p.H1047R, p.H1047Y), utilizing formalin-fixed paraffin-embedded breast tumor tissue, reported as PIK3CA gene mutation status (PLA code for the therascreen® PIK3CA RGQ PCR Kit from QIAGEN) |
| | 0177U | Oncology (breast cancer), DNA, PIK3CA (phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha) gene analysis of 11 gene variants utilizing plasma, reported as PIK3CA gene mutation status (PLA code for the therascreen® PIK3CA RGQ PCR Kit test from QIAGEN) ***(Code effective 7/1/2020)*** |
| | 0211U | Oncology (pan-tumor), DNA and RNA by next-generation sequencing, utilizing formalin-fixed paraffin-embedded tissue, interpretative report for single nucleotide variants, copy number alterations, tumor mutational burden, and microsatellite instability, with therapy association (PLA code for the MI Cancer Seek™ – NGS Analysis from Caris MPI d/b/a Caris Life Sciences.) ***(Code effective 10/1/2020)*** |
| | 81191 | NTRK1 (neurotrophic receptor tyrosine kinase 1) (e.g., solid tumors) translocation analysis ***(Code effective 1/1/2021)*** |
| | 81192 | NTRK2 (neurotrophic receptor tyrosine kinase 2) (e.g., solid tumors) translocation analysis ***(Code effective 1/1/2021)*** |
| | 81193 | NTRK3 (neurotrophic receptor tyrosine kinase 3) (e.g., solid tumors) translocation analysis ***(Code effective 1/1/2021)*** |

| Type | Code | Description |
|---|---|---|
| | 81194 | NTRK (neurotrophic-tropomyosin receptor tyrosine kinase 1, 2, and 3) (e.g., solid tumors) translocation analysis ***(Code effective 1/1/2021)*** |
| | 81301 | Microsatellite instability analysis (e.g., hereditary non-polyposis colorectal cancer, Lynch syndrome) of markers for mismatch repair deficiency (e.g., BAT25, BAT26), includes comparison of neoplastic and normal tissue, if performed |
| | 81309 | PIK3CA (phosphatidylinositol-4, 5-biphosphate 3-kinase, catalytic subunit alpha) (e.g., colorectal and breast cancer) gene analysis, targeted sequence analysis (e.g., exons 7, 9, 20) |
| | 81445 | Targeted genomic sequence analysis panel, solid organ neoplasm, DNA analysis, and RNA analysis when performed, 5-50 genes (e.g., ALK, BRAF, CDKN2A, EGFR, ERBB2, KIT, KRAS, NRAS, MET, PDGFRA, PDGFRB, PGR, PIK3CA, PTEN, RET), interrogation for sequence variants and copy number variants or rearrangements, if performed |
| | 81455 | Targeted genomic sequence analysis panel, solid organ or hematolymphoid neoplasm, DNA analysis, and RNA analysis when performed, 51 or greater genes (e.g., ALK, BRAF, CDKN2A, CEBPA, DNMT3A, EGFR, ERBB2, EZH2, FLT3, IDH1, IDH2, JAK2, KIT, KRAS, MLL, NPM1, NRAS, MET, NOTCH1, PDGFRA, PDGFRB, PGR, PIK3CA, PTEN, RET), interrogation for sequence variants and copy number variants or rearrangements, if performed |
| | 86152 | Cell enumeration using immunologic selection and identification in fluid specimen (e.g., circulating tumor cells in blood) |
| | 86153 | Cell enumeration using immunologic selection and identification in fluid specimen (e.g., circulating tumor cells in blood); physician interpretation and report, when required |
| | 88360 | Morphometric analysis, tumor immunohistochemistry (e.g., Her-2/neu, estrogen receptor/progesterone receptor), quantitative or semiquantitative, per specimen, each single antibody stain procedure; manual |
| | 88361 | Morphometric analysis, tumor immunohistochemistry (e.g., Her-2/neu, estrogen receptor/progesterone receptor), quantitative or semiquantitative, per specimen, each single antibody stain procedure; using computer-assisted technology |
| | 81479 | Unlisted molecular pathology procedure |
| **HCPCS** | None | |

## Policy History

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

| Effective Date | Action |
|---|---|
| 02/01/2021 | New policy |

## Definitions of Decision Determinations

**Medically Necessary:** Services that are Medically Necessary include only those which have been established as safe and effective, are furnished under generally accepted professional standards to treat illness, injury or medical condition, and which, as determined by Blue Shield, are: (a) consistent with Blue Shield medical policy; (b) consistent with the symptoms or diagnosis; (c) not furnished primarily for the convenience of the patient, the attending Physician or other provider; (d) furnished at the most appropriate level which can be provided safely and

effectively to the patient; and (e) not more costly than an alternative service or sequence of services at least as likely to produce equivalent therapeutic or diagnostic results as to the diagnosis or treatment of the Member's illness, injury, or disease.

**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 at (800) 541-6652, or the Transplant Case Management Department at (800) 637-2066 ext. 3507708 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.*

## Appendix A

| POLICY STATEMENT | |
|---|---|
| **BEFORE** | **AFTER**<br>**Blue font: Verbiage Changes/Additions** |
| **New Policy**<br><br>**Policy Statement:**<br>**N/A** | **Biomarker Testing (Including Liquid Biopsy) for Targeted Treatment and Immunotherapy in Breast Cancer 2.04.151**<br><br>**Policy Statement:**<br>*PIK3CA* Testing<br>*PIK3CA* testing may be **medically necessary** to predict treatment response to alpelisib (Piqray) in patients with hormone receptor-positive, HER2 negative advanced or metastatic breast cancer (see Policy Guidelines).<br><br>*PIK3CA* testing of tissue is considered **investigational** in all other situations unless included in a panel approved under another policy.<br><br>***NTRK* Gene Fusion Testing**<br>Analysis of *NTRK* gene fusions may be considered **medically necessary** to predict treatment response to entrectinib (Rozlytrek) or larotrectinib (Vitrakvi) in patients with locally advanced or metastatic breast cancer that has progressed following standard treatment and who have no alternative treatment option (see Policy Guidelines).<br><br>Analysis of *NTRK* gene fusions is considered **investigational** in all other situations unless included in a panel approved under another policy.<br><br>**PD-L1 Testing**<br>PD-L1 testing may be considered **medically necessary** to predict treatment response to atezolizumab (Tecentriq) in patients with hormone receptor-negative/HER2-negative (triple negative) metastatic or unresectable breast cancer (see Policy Guidelines).<br><br>PD-L1 testing may be considered **medically necessary** to predict treatment response to pembrolizumab (Keytruda) in patients with hormone receptor-negative/HER2-negative (triple negative) recurrent or metastatic breast cancer (see Policy Guidelines). |

| POLICY STATEMENT | |
|---|---|
| **BEFORE** | **AFTER**<br>**Blue font: Verbiage Changes/Additions** |
| | PD-L1 testing is considered **investigational** in all other situations unless included in a panel or separately approved under another policy.<br><br>**MSI-H/dMMR Testing**<br>MSI-H/dMMR testing may be considered **medically necessary** to predict treatment response to pembrolizumab (Keytruda) in patients with unresectable or metastatic breast cancer that has progressed following standard treatment and who have no alternative treatment option (see Policy Guidelines).<br><br>MSI-H/dMMR testing is considered **investigational** in all other situations unless included in a panel or separately approved under another policy..<br><br>**Tumor Mutational Burden Testing**<br>Tumor mutational burden testing to predict response to immunotherapy in patients with breast cancer is considered **investigational.**<br><br>**Circulating Tumor DNA**<br>*PIK3CA* testing using FoundationOne Liquid CDx (FDA approved companion test) may be considered **medically necessary** to predict treatment response to alpelisib (Piqray) in patients with hormone receptor-positive, HER2 negative advanced or metastatic breast cancer (see Policy Guidelines) when there is insufficient tissue to be tested and an additional invasive procedure would be required otherwise.<br>Circulating tumor DNA testing is considered **investigational** in all other situations unless included in a panel approved under another policy, such as use in Non-Small Cell Lung Cancer (NSCLC).<br><br>**Circulating Tumor Cells**<br>Analysis of circulating tumor cells to select treatment in patients with breast cancer is considered **investigational** (see Background section). |