Genetic Testing for Lactase Insufficiency

Description

Genetic testing of adults with suspected lactase insufficiency is proposed as an alternative to current diagnostic practices. Studies have demonstrated a tight correlation between a single nucleotide polymorphism (SNP) -13910 C>T upstream of the gene coding for the enzyme lactase and lactase insufficiency in persons of European ancestry. Currently, two indirect tests of lactose digestion, the hydrogen breath test (HBT) and lactose tolerance blood test (LTT), are the most preferred diagnostic tests for confirmation of lactase insufficiency.

Background

The predominant carbohydrate in milk is the disaccharide lactose consisting of the simple sugars glucose and galactose. The brush-border enzyme lactase hydrolyzes lactose into its monosaccharide components that are absorbable by the intestinal mucosa. Except for rare instances of congenital hypolactasia, most infants are able to produce lactase with enzyme levels highest at birth. Sometime after weaning in the majority of children there is a decrease in lactase production through a multifactorial process that is regulated at the gene transcription level. (1)

The decrease in lactase level varies significantly by ethnic group both in terms of the lowest level of lactase and time from weaning necessary to reach the nadir of lactase activity. (2) By 2 to 12 years of age two groups emerge: a group with insufficient levels of lactase activity (primary hypolactasia or lactase non-persistence) and a group that retains the infant level of lactase activity through adulthood (lactase-persistence). (3) The ethnic groups with the highest rates of lactase insufficiency are Asian, Native American and Blacks with the lowest rates in people of northern European origin. (Table 1)

<table>
<thead>
<tr>
<th>Population</th>
<th>Lactase Insufficiency*%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Northern Europeans</td>
<td>2 to 15%</td>
</tr>
<tr>
<td>American Whites</td>
<td>6 to 22%</td>
</tr>
<tr>
<td>Central Europeans</td>
<td>9 to 23%</td>
</tr>
<tr>
<td>Northern Indians</td>
<td>20 to 30%</td>
</tr>
<tr>
<td>Southern Indians</td>
<td>60 to 70%</td>
</tr>
<tr>
<td>Hispanics</td>
<td>50 to 80%</td>
</tr>
<tr>
<td>Ashkenazi Jews</td>
<td>60 to 80%</td>
</tr>
<tr>
<td>Blacks</td>
<td>60 to 80%</td>
</tr>
</tbody>
</table>
Problems with the absorption of lactose can be described in several terms:

- **Lactase insufficiency** (lactase non-persistence or primary hypolactasia) – indicates that lactase activity is a fraction of the original infantile level. Direct measurement of lactase activity is tested biochemically through duodenal biopsy. (5) Lactase insufficiency is highly correlated with the C/C genotype at -13910 in the lactase promoter region. In adults with a homozygous lactase persistence genotype (T/T) lactase levels are approximately 10-times higher than for the lactase insufficient genotype (C/C) with heterozygous individuals (C/T) showing intermediate levels. (6) These heterozygous individuals may experience symptoms of lactose intolerance when ingesting quantities of lactose greater than their intermediate level of lactase can digest.

- **Lactose malabsorption** – indicates that a sizable fraction of lactose is not able to be absorbed in the small bowel and is delivered to the colon. Malabsorption is tested by HBT or LTT. (5)

- **Lactose intolerance** – indicates that lactose malabsorption causes gastrointestinal symptoms. There is no genetic test for lactose intolerance and demonstration of lactose intolerance requires patients to self-report symptoms after lactose ingestion (Table 2). Diagnosis of lactose intolerance is highly susceptible to the placebo effect and studies should appropriately conduct a blinded lactose challenge with an indistinguishable placebo. (3) A meta-analysis by Jellema and colleagues indicated that no specific patient complaint could predict lactose malabsorption with sensitivity and specificity ranging from 0-90% and 18-96% for the most common lactose intolerance symptoms. (7) Similarly, patient self-reported milk tolerance was also not found to be accurate in predicting lactose malabsorption with sensitivity and specificity ranging from 30-70% and 25-87% respectively. (7)

### Table 2 Symptoms of Lactose Intolerance (2)

<table>
<thead>
<tr>
<th>Gut-related symptoms</th>
<th>% of total patients who experience symptom</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abdominal pain</td>
<td>100</td>
</tr>
<tr>
<td>Gut distention</td>
<td>100</td>
</tr>
<tr>
<td>Borborygmi</td>
<td>100</td>
</tr>
<tr>
<td>Flatulence</td>
<td>100</td>
</tr>
<tr>
<td>Diarrhea</td>
<td>70</td>
</tr>
<tr>
<td>Constipation</td>
<td>30</td>
</tr>
<tr>
<td>Nausea</td>
<td>78</td>
</tr>
<tr>
<td>Vomiting</td>
<td>78</td>
</tr>
<tr>
<td><strong>Systemic symptoms</strong></td>
<td></td>
</tr>
<tr>
<td>Headache and light headedness</td>
<td>86</td>
</tr>
<tr>
<td>Loss of concentration and poor short-term memory</td>
<td>82</td>
</tr>
<tr>
<td>Long-term severe tiredness</td>
<td>63</td>
</tr>
<tr>
<td>Muscle pain</td>
<td>71</td>
</tr>
<tr>
<td>Joint pain and/or swelling</td>
<td>71</td>
</tr>
<tr>
<td>Allergy (eczema, pruritus, rhinitis, sinusitis, asthma)</td>
<td>40</td>
</tr>
<tr>
<td>Heart arrhythmia</td>
<td>24</td>
</tr>
<tr>
<td>Mouth ulcers</td>
<td>30</td>
</tr>
</tbody>
</table>
Lactase insufficiency is a common condition which occurs in approximately (70%) of persons after weaning. (8) An insufficiency of lactase results in the malabsorption of lactose, which may lead to symptoms of lactose intolerance such as abdominal pain, bloating, diarrhea and increased flatulence, caused by bacterial fermentation of undigested lactose in the colon. (9) However, the demonstration of lactose malabsorption does not necessarily indicate that an individual will be symptomatic. Many variables determine if a person who malabsorbs lactose develops symptoms, including: the dose of lactose ingested, residual intestinal lactase activity, ingestion of food along with lactose, the ability of the colonic flora to ferment lactose and the individual sensitivity to the products of lactose fermentation. Because of these factors, the number of persons reporting symptoms of lactose intolerance is likely only a fraction of those who are lactase insufficient. In addition, lactose malabsorption may be secondary (secondary hypolactasia) to an acquired condition such as: small bowel bacterial overgrowth, infectious enteritis, mucosal damage from celiac disease, inflammatory bowel disease, antibiotics, gastrointestinal surgery, short bowel syndrome, radiation enteritis or other conditions which may lead to reduction of lactase expression in the small intestine. (6)

Clinical Diagnosis of Lactase Insufficiency

Mucosal biopsy of the duodenum followed by biochemical lactase assay to directly measure lactase activity is the reference standard for diagnosis of lactase insufficiency. This approach may also exclude other causes of secondary lactose malabsorption through endoscopy. However, this approach is limited in utility due to the invasiveness of the procedure and the patchy expression of lactase in the duodenum.

Two common alternatives to this direct method of measuring lactase level are the hydrogen breath test (HBT) and lactose tolerance blood test (LTT) which measure lactose malabsorption. Because lactose malabsorption is nearly always attributable to lactase insufficiency, this can typically be imputed from measurements of lactose malabsorption. (3)

The HBT measures the amount of hydrogen exhaled by gas chromatography for up to 3 hours after ingesting 25-50 g of lactose. Persons undergoing HBT are required to fast overnight and refrain from activities that may elevate breath hydrogen during testing. A rise in breath hydrogen of 0.31–2.5 mL/min is indicative of bacterial fermentation from the malabsorbed lactose. A negative HBT can exclude lactose malabsorption as the cause of symptoms, and a positive result indicates that the symptoms may be attributable to ingestion of lactose. (3) The following factors are associated with a rise in breath hydrogen and may cause false-positive results if present at time of testing:

- Diabetes
- Small bowel disease (e.g., celiac, giardiasis)
- Bacterial overgrowth
- Altered colon pH
- Antibiotic usage
- Probiotic usage
The LTT measures blood glucose increase over time with blood drawn at 15, 30, 60, and 90 minutes after ingesting a 25-50 g dose of lactose. A glucose increase of less than 20 mg/dL above an 8-hour fasting level indicates an abnormal test. The following factors are associated with a rise in blood sugar when undergoing a lactose tolerance test and may cause false-positive results:

- Diabetes
- Small-bowel disease (e.g., celiac, giardiasis)
- Thyroid disorders
- Motility disorders (stomach, small bowel)
- Bacterial overgrowth

Molecular Diagnosis of Lactase Insufficiency

In 2002, Enattah et al identified the first DNA variant to control transcription of lactase in 2002. (10) This polymorphism, −13910 C>T, is located in a noncoding region of the MCM6 gene that is upstream of the lactase gene (LCT). The less common T allele has been associated with lactase persistence and has demonstrated an autosomal dominant pattern of inheritance. This polymorphism is thought to be related to the domestication of animals during the last 10,000-12,000 years, and persons with the C/C genotype have been shown to be strongly associated with a lactase insufficiency phenotype in Caucasians. Other polymorphisms in the same MCM6 regulatory region are associated with other ethnic groups (such as Africans and Arabs), but prevalence of these vary geographically (6, 11) and to date no commercially available testing kits have incorporated these polymorphisms.

Prometheus’s LactoTYPE® is a commercially available polymerase chain reaction–based test that assesses the most common lactase nonpersistence variant, MCM6 -13910 C>T, in patients with suspected lactose intolerance. Fulgent Clinical Diagnostics Lab also offers MCM6 sequencing and deletion/duplication analysis using next-generation sequencing. Demonstration of the C/C genotype can be used as indirect evidence of lactase insufficiency and lactose malabsorption.

Treatment of Lactase Insufficiency

The goal of treatment should be to ensure adequate nutrients important for skeletal health. (1) Dietary adjustment to restrict the consumption of foods containing lactose is the principal form of therapy for patients with lactase insufficiency. However, even lactose maldigesters can usually tolerate small amounts of lactose (12 g/day) with no or minimal symptoms. Lactase enzyme preparations are available for symptom relief but may not be effective in all patients.
Regulatory Status

No U.S. Food and Drug Administration (FDA)-cleared genotyping tests were found. Thus, genotyping is offered as a laboratory-developed test. Clinical laboratories may develop and validate tests in-house (“home-brew”) and market them as a laboratory service; such tests must meet the general regulatory standards of the Clinical Laboratory Improvement Act (CLIA). The laboratory offering the service must be licensed by CLIA for high-complexity testing.

Related Policies

None

Policy

*This policy statement applies to clinical review performed for pre-service (Prior Approval, Precertification, Advanced Benefit Determination, etc.) and/or post-service claims.

The use of targeted mutation analysis of -13910 C>T for the prediction of lactase insufficiency is considered investigational.

Benefit Application

The BCBS FEP contract affects coverage for genetic screening and testing. Refer to the current FEP Service Benefit Plan brochure for additional information and guidance.

Rationale

Literature Review

Literature that describes the analytic validity, clinical validity, and clinical utility of genetic testing for lactase insufficiency was sought.

Analytic Validity

Analytic validity is the technical accuracy of the test in detecting a mutation that is present or in excluding a mutation that is absent. According to the Genetic Testing Registry, analytical sensitivity of next-generation sequencing and deletion/duplication analysis of MCM6 exceeds 98%. Analytical specificity and accuracy are 96% and 97%, respectively. (12)

Clinical Validity

Clinical validity is the diagnostic performance of the test (sensitivity, specificity, positive and negative predictive values) in detecting clinical disease.

Many reports on the diagnosis of lactase insufficiency by polymerase chain reaction (PCR) mutation analysis of -13910 C>T have been published, and those that assess the agreement between genotyping and hydrogen breath test (HBT), lactose tolerance blood test (LTT), or biopsy are presented in Table 3. Nineteen studies compared genotyping of single nucleotide polymorphism (SNP) -13910 C>T to HBT and found sensitivities and specificities ranging from 71% to 100% and 46% to 100%,
respectively. Five studies compared genotyping with LTT with sensitivities and specificities ranging from 85% to 100% and 87% to 95%, respectively. One study compared genotyping to a hydrogen/methane breath test, which may be more sensitive than HBT, and reported Cohen's kappa statistic (κ) of 0.44, indicating moderate agreement. (13) Heterogeneity in study populations, dose of lactose given in HBT/LTT, and age of participants contributed to the wide range of observed sensitivities and specificities. Direct comparison of these tests is not possible because no identified studies compared both genotyping and HBT/LTT with the criterion standard of duodenal mucosal biopsy. Indirect comparison is not possible because of the small number of studies comparing genotyping, HBT, or LTT with biopsy.

Incomplete agreement between genotyping for lactase insufficiency and indirect tests of lactose malabsorption is expected because these tests do not measure the same parameters. LTT and HBT are intended to diagnosis lactose malabsorption, which can be caused by factors other than lactase insufficiency. Additionally, because lactase activity persists for years after weaning, the inclusion of children can affect the concordance between HBT/LTT and genotyping. Di Stefano et al (2009) found that the overall κ value for agreement of HBT and genotyping was 0.74, but for those younger than and older than 30 years of age, κ values were 0.56 and 1.0, respectively (p<0.005 for both comparisons). (14)

In addition, the SNP -13910 C>T is not the only MCM6 polymorphism implicated in regulating transcription of the LCT gene. A study by Eadala et al (2011) recruited patients with inflammatory bowel disease along with healthy control patients and found that although the C/C genotype was strongly associated with experiencing symptoms of lactose intolerance after HBT, there was a high proportion of lactose sensitivity in C/T and T/T genotype patients as well. (15) A 2012 Colombian study by Mendoza-Torres et al found low specificity (46%) when comparing HBT with genotyping.(16) The authors attributed this to the genetic heterogeneity of the Colombian and Caribbean population studied and recommended against using genotyping to assess lactase insufficiency in this population. Similarly, Santonocito et al (2015) found a similar proportion (~80%) of homozygous genotypes for lactase nonpersistence among 1426 patients with gastrointestinal symptoms and 1000 healthy volunteers in south central Italy. (17) These results suggest that unmeasured genetic variation may more fully explain lactase insufficiency.

Table 3 Sensitivity and Specificity of Analysis of the Genotyping Compared with HBT, LTT, and Intestinal Biopsy

<table>
<thead>
<tr>
<th>Author, Year, Country</th>
<th>N</th>
<th>Sensitivity (95% CI)</th>
<th>Specificity (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Targeted mutation analysis of SNP -13910 C&gt;T results compared with hydrogen breath test (HBT)</td>
<td></td>
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</tr>
<tr>
<td>Gugatschka, 2005, Austria (18)</td>
<td>51</td>
<td>90 (73-98)</td>
<td>95 (76-100)</td>
</tr>
<tr>
<td>Buning, 2005, Germany (19)</td>
<td>166</td>
<td>98 (93-100)</td>
<td>83 (71-91)</td>
</tr>
<tr>
<td>Hogenauer, 2005, Austria (9)</td>
<td>123</td>
<td>97 (86-100)</td>
<td>86 (77-93)</td>
</tr>
<tr>
<td>Bulhues, 2007, Brazil (20)</td>
<td>20</td>
<td>90 (55-100)</td>
<td>100 (69-100)</td>
</tr>
<tr>
<td>Schirru, 2007, Italy (21)</td>
<td>84</td>
<td>84 (72-93)</td>
<td>96 (81-100)</td>
</tr>
<tr>
<td>Bernardes, 2007, Brazil (22)</td>
<td>147</td>
<td>76 (59-89)</td>
<td>100 (40-100)</td>
</tr>
<tr>
<td>Szilagy, 2007, Canada (23)</td>
<td>30</td>
<td>93 (68-100)</td>
<td>80 (52-96)</td>
</tr>
<tr>
<td>Kerber, 2007, Austria (24)</td>
<td>120</td>
<td>97 (86-100)</td>
<td>72 (61-95)</td>
</tr>
<tr>
<td>Mattar, 2008, Brazil (25)</td>
<td>50</td>
<td>96 (82-100)</td>
<td>100 (85-100)</td>
</tr>
<tr>
<td>Krawcyk, 2008, Germany (26)</td>
<td>58</td>
<td>100 (78-100)</td>
<td>95 (84-99)</td>
</tr>
</tbody>
</table>
A 2012 meta-analysis by Marton et al. assessed the diagnostic accuracy of the LTT and HBT tests compared to genotyping for the polymorphism -13910 C>T for prediction of lactase insufficiency phenotype. (39) Seventeen studies evaluated HBT and 5 evaluated LTT. The overall sensitivity and specificity of the HBT was 88% (95% confidence interval [CI]: 85-90%) and 85% (95% CI: 82-87%), respectively. Both sensitivity and specificity showed high heterogeneity (I² 78% and 87%) and the authors detected a potential for publication bias within their included studies. LTT overall sensitivity was 94% (95% CI: 90-97%) with a specificity of 90% (95% CI: 84 – 95%). No significant heterogeneity was observed for the sensitivity and specificity of the LTT. Three studies also assessed the sensitivity of the -2018 G>A genotype, which has also been described in Europeans, and found a less accurate overall sensitivity and specificity of 87% (95% CI: 79-93%) and 76% (95% CI: 67-83%) than the -13910 C>T polymorphism.
Clinical Utility

Clinical utility is how the results of the diagnostic test will be used to change management of the patient and whether these changes in management lead to clinically important improvements in health outcomes.

No studies were identified that attempted to demonstrate improved patient outcomes or changes to patient management because of genetic testing for lactase insufficiency.

Lactase insufficiency is the normal phenotype for most adults, and a confirmatory diagnosis with HBT, LTT, or genotyping is generally not necessary. Empiric diagnosis by dietary restriction is adequate in most circumstances as this is the primary treatment for lactase insufficient patients. Patients who achieve satisfactory symptom control following dietary modifications do not require further diagnostic testing. For the majority of patients who do not achieve symptom control following dietary modifications, testing is indicated for the presence of other conditions that can cause symptoms similar to lactase deficiency.

The proposed clinical utility of genotyping for lactase insufficiency is that the test offers a more comfortable assessment for patients when compared to the HBT, LTT or biopsy. Traditional testing methods may be associated with discomfort caused by the ingestion of a large volume of lactose and there is dietary preparation and fasting before testing. Additionally, factors that influence the interpretation of the indirect HBT and LTT will not lead to false-positive test results from genotyping. Arroyo and colleagues suggest that genetic testing, when used in conjunction with HBT, can help in the diagnosis of secondary hypolactasia when there is a positive HBT and the patient is not C/C genotype. (40)

Clinical Trials

A search of online site ClinicalTrials.gov identified no ongoing studies evaluating molecular diagnostic tests for lactase deficiency.

Practice Guidelines and Position Statements

No guidelines or position statements were identified with recommendations on testing for lactase deficiency.

U.S. Preventive Services Task Force

Not applicable

Summary of Evidence

Genetic testing of adults with suspected lactase insufficiency is proposed as an alternative to current diagnostic practices, which include hydrogen breath test (HBT), lactose tolerance blood test (LTT) and intestinal biopsy. Studies have demonstrated a tight correlation between a single nucleotide polymorphism (SNP) -13910 C>T upstream of the gene coding for the enzyme lactase and lactase insufficiency in persons of European ancestry, and studies in Caucasian populations report a high
degree of agreement for the diagnosis of lactase insufficiency between genotyping and both the HBT and LTT.

Genetic testing has the potential advantage of sparing patients the discomfort of fasting and experiencing symptoms of lactose intolerance during the administration of HBT or LTT. Genotyping may also have additional utility in the diagnosis of secondary hypolactasia.

However, because there is no current treatment for lactase insufficiency, and management involves dietary restriction and palliation of lactose intolerance symptoms alone, an empiric diagnosis of lactose intolerance in the absence of confirmation with HBT, LTT or genotyping followed by treatment with dietary restriction of lactose is suitable. There is currently insufficient evidence that the assessment of the genetic etiology of lactose intolerance would affect patient management or improve clinical outcomes, and therefore the use of targeted mutation analysis of -13910 C>T for the prediction of lactase insufficiency is considered investigational.

Medicare National Coverage

No national coverage determination was identified.

References


### Policy History

<table>
<thead>
<tr>
<th>Date</th>
<th>Action</th>
<th>Reason</th>
</tr>
</thead>
<tbody>
<tr>
<td>September 2013</td>
<td>New Policy</td>
<td>Policy created with literature search.</td>
</tr>
<tr>
<td>September 2014</td>
<td>Update Policy</td>
<td>Policy updated with literature search adding references 11 and 35. No changes to the policy statement.</td>
</tr>
<tr>
<td>September 2015</td>
<td>Update Policy</td>
<td>Policy updated with literature review through March 10, 2015; references 12-13 and 17 added. Policy statements unchanged.</td>
</tr>
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</table>

### Keywords

Hypolactasia, Genetic Testing  
Lactose Intolerance, Genetic Testing

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This policy was approved by the FEP® Pharmacy and Medical Policy Committee on September 18, 2015 and is effective October 15, 2015.

Signature on file

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