The literature is utilized. Incorporates segregation analysis in families, co-occurrences with deleterious mutations, and the variant rate within that group is predicted to decline.

Currently, variants of uncertain significance are more common in non-European ancestries. Of particular interest is the large decline in the African American (a decline of 87.0%) and Latin American populations (a decline of 85.1%; Figure 1). The dramatic decline in the overall rate of VUS for Lynch syndrome associated genes (a decline of 86.8%; Table 1) can be attributed to the development and application of improved statistical techniques and targeted methodologies for data gathering through clinical genetic testing in these populations. However, Myriad has been able to further characterize most of these variants and subsequently reduce the VUS rate to only 2.9% for the BRCA genes. Similarly, for the genes associated with Lynch syndrome, there has been a steady decline in the percentage of variants reported as being of uncertain clinical significance. The current VUS rate for MLH1/MSH2/MSH6/EPCAM is 6.6% and for PMS2 is 4.0%.

Interpreting the clinical significance of new variants is a challenge associated with any sequencing-based genetic test. This requires a costly labor and data intensive effort to analyze variants of uncertain significance using various lines of evidence. Of the many and varied types of mutations in the BRCA genes detected when testing first became clinically available in 1996, over 40% (data not shown) of results were originally of unknown significance. Myriad has been able to further characterize most of these variants and subsequently reduce the VUS rate to only 2.9% for the BRCA genes. Similarly, for the genes associated with Lynch syndrome, there has been a steady decline in the percentage of variants reported as being of uncertain clinical significance. The current VUS rate for MLH1/MSH2/MSH6/EPCAM is 6.6% and for PMS2 is 4.0%. Although the overall rate of VUS has decreased, it is important to note that novel variants are still discovered in these genes.

Myriad utilizes multiple lines of evidence to evaluate and reclassify VUS, and new classification methods are continually being assessed by Myriad scientists. Current methodologies include:

- Co-occurrence with a deleterious mutation in-trans for genes with known compound heterozygosity phenotype
- Deleterious mutation co-occurrence with mutations in other genes associated with the same syndrome
- Personal and family history analysis (Phenotype analysis)
- Segregation analysis
- Evolutionary conservation
- Literature evaluation

Myriad has been able to apply methodologies and statistical techniques developed on a large BRCA mutation data set to Lynch syndrome associated genes thereby reducing the rates across all of the associated genes. Clarification of the meaning of a VUS gives more precise information to clinicians managing their patient and assists them in making medical management choices.

### References


### Results

**Variant of uncertain significance rates and ancestry for BRCA genes:** In 2002, 12.8% of all BRCA1/2 tests resulted in a VUS result by 2012, that rate had declined to 2.9% (77.3% decline, Table 1). From 2002 to 2012 there was a decline in the VUS rate across all ancestries. Of particular interest is the large decline in the African American (a decline of 87.0%) and Latin American populations (a decline of 85.1%; Figure 1). The dramatic decline in the VUS rate in these ancestries is due to both improved methods for establishing the clinical significance of variants and increased utilization of testing in these populations, which provides more data for analysis.

**Variant of uncertain significance rates and ancestry for Lynch syndrome associated genes:** Testing for Lynch syndrome has evolved over time. In 2002 testing at Myriad consisted of sequencing of MLH1 and MSH2 genes. As of 2012, testing includes evaluation of the mismatch repair genes associated with Lynch syndrome (MLH1, MSH2, MSH6, and PMS2) and EPCAM mutations in EPCAM can effect expression of MSH2). In order to compare variant rates in Lynch syndrome testing over time we compared MLH1/MSH2 variant rate from 2005 to the 2012 overall variant rate of MLH1/MSH2/MSH6/EPCAM. The variant rates for 2012 are reported separately which reflects the way testing is conducted at Myriad at this time. While PMS2 is conducted concurrently and included in Myriad's standard Lynch syndrome testing along with MLH1/MSH2/MSH6/EPCAM these tests are run separately. Similar to BRCA1/2 the overall rate of VUS for Lynch syndrome associated genes has decreased over time with the current rate for MLH1/MSH2/MSH6/EPCAM being 6.6% (34.7% decline; Table 2). This decline was noted across multiple ancestries with the exception of the Latin American population (Table 2). The overall rate for VUS as of 2012 for MLH1/MSH2/MSH6/EPCAM genes is 6.6% and for PMS2 is 4%.

### Discussion

Interpreting the clinical significance of new variants is a challenge associated with any sequencing-based genetic test. This requires a costly labor and data intensive effort to analyze variants of uncertain significance using various lines of evidence. Of the many and varied types of mutations in the BRCA genes detected when testing first became clinically available in 1996, over 40% (data not shown) of results were originally of unknown significance. Myriad has been able to further characterize most of these variants and subsequently reduce the VUS rate to only 2.9% for the BRCA genes. Similarly, for the genes associated with Lynch syndrome, there has been a steady decline in the percentage of variants reported as being of uncertain clinical significance. The current VUS rate for MLH1/MSH2/MSH6/EPCAM is 6.6% and for PMS2 is 4.0%. Although the overall rate of VUS has decreased, it is important to note that novel variants are still discovered in these genes.

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### References