Prenatal Testing for Noonan Syndrome in Fetuses with Abnormal Ultrasound Findings, including Cystic Hygroma

Mendelian Inheritance in Man Number: 163950 (Noonan syndrome), 176876 (PTPN11), 182530 (SOS1), 164760 (RAF1), 190070 (KRAS), 190020 (HRAS), 164757 (BRAF), 176872 (MAP2K1), 601263 (MAP2K2), 602775 (SHOC2)

Clinical features:
Individuals with Noonan syndrome (NS) have dysmorphic facial features, such as hypertelorism, downward slanting eyes, epicanthal folds, and low-set and posteriorly rotated ears. Other features include short stature, pterygium colli, short, webbed neck, deafness, motor delay, and bleeding diathesis. Structural cardiac defects (A-V canal defects, pulmonic stenosis, and coartation of the aorta) may be suspected prenatally; however, hypertrophic cardiomyopathy, secundum ASD and patent ductus arteriosus are usually identified after delivery. Most of the features of Noonan syndrome are not identified in the first or second trimester of pregnancy, although transient first trimester cystic hygroma has been associated with a clinical diagnosis of Noonan syndrome in 1-4% of cases with normal karyotype. In addition to Noonan syndrome, increased nuchal translucency has been seen in association with fetal chromosome abnormalities, fetal demise, heart defects, infection, and a number of other genetic conditions. Third trimester ultrasound findings of abnormal facies, lymphedema, macrosomia, cardiac defects, and the obstetric complication of polyhydramnios have been reported in Noonan syndrome.

Inheritance pattern:
Noonan syndrome is a genetically heterogeneous, autosomal dominant disorder. Many cases are sporadic and are likely due to new mutation.

Reasons for referral:
1. Prenatal ultrasound findings suggestive of Noonan syndrome, including cystic hygroma
2. Fetus with ultrasound anomalies & parent with clinical diagnosis of Noonan syndrome

Test method:
Using genomic DNA obtained from chorionic villi, cultured villi, or cultured amniocytes, a comprehensive prenatal Noonan syndrome panel is performed on a custom-designed oligonucleotide Affymetrix resequencing chip. The resequencing array contains 8 genes in the RAS/MAPK pathway involved in Noonan syndrome and related disorders (PTPN11, SOS1, KRAS, RAF1, BRAF, MAP2K1, MAP2K2, HRAS). This panel also includes mutation specific analysis for the Ser2Gly SHOC2 gene mutation using bi-directional dideoxy-based DNA sequencing. In addition, for each test we will perform genotype analysis of maternal and fetal DNA for several polymorphic markers to test for maternal cell contamination. Therefore, in all cases a maternal sample (either blood in EDTA or buccal swabs) should accompany the fetal sample.

Test sensitivity:
In fetuses, transient first trimester cystic hygroma has been associated with a clinical diagnosis of Noonan syndrome in 1-4% of cases with normal karyotype. In a recent retrospective study of 134 fetuses with sonographic findings suggestive of Noonan syndrome, including data from GeneDx and Mount Sinai School of Medicine, 9% (12 fetuses) were found to have a heterozygous missense mutation in PTPN11 (Lee et al. 2009). The prevalence of PTPN11 mutations was higher in fetuses with cystic hygroma associated with additional abnormalities (24%), in particular with congenital heart defects (37%). The mutation detection rate for BRAF, HRAS, KRAS, MAP2K1, MAP2K2, RAF1, SHOC2, and SOS1 has not yet been established. However, a study of 14 patients positive for a RAF1 mutation with postnatal diagnosed Noonan syndrome and available prenatal ultrasound data reports that 6 patients had fetal macrosomia, 5 had polyhydramnios, and 1 had increased nuchal translucency (Pandit et al. 2007). All of these RAF1 mutations were located in exons 7, 14 and 17, which are included in our comprehensive prenatal Noonan syndrome panel.
Mutation spectrum:
All mutations identified in affected fetuses to date (n=12) have been missense changes in PTPN11 (Lee et al. 2009). No data are currently available for any of the other genes associated with Noonan syndrome or related disorders (BRAF, HRAS, KRAS, MAP2K1, MAP2K2, RAF1, SHOC2, and SOS1).

Specimen Requirements and Shipping/Handling:
- **Prenatal Specimen – Based on Abnormal Ultrasound/Other Findings (Test # 357)**: 20 mg villi preferred (minimum 15 mg) or 20 mL amniotic fluid (will be cultured at GeneDx prior to testing) or 2 T25 flasks of cultured CV or cultured amniocytes. Ship overnight at ambient temperature, using a cool pack in hot weather.
- **Maternal cell contamination studies (required for all prenatal testing)**: 1-4 mL of maternal blood in a lavender-top EDTA tube. Ship overnight at ambient temperature, using a cool pack in hot weather. Specimens may be refrigerated for 7 days prior to shipping. Alternatively, buccal brushes (GeneDx buccal kit only) or DNA can be used. The maternal blood sample should accompany the prenatal specimen or should be shipped to arrive prior to or concurrent with the prenatal specimen. (Call to discuss requirements for parental blood.)

*If more than one prenatal test is ordered, 30 mL amniotic fluid, 30mg villi or 3 T-25 flasks of cultured cells are requested*

Required Forms:
- Sample Submission Form – complete all pages
- Payment Options Form or Institutional Billing Instructions (last page of submission form)

CPT Codes and Turn-Around-Times:

<table>
<thead>
<tr>
<th>Test#</th>
<th>Description</th>
<th>CPT codes</th>
<th>Price**</th>
<th>Turnaround time</th>
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</thead>
<tbody>
<tr>
<td>357</td>
<td>Prenatal diagnosis of Noonan syndrome based on abnormal ultrasound findings. Comprehensive Prenatal Noonan Syndrome Resequencing Array: 8 genes sequenced simultaneously, including targeted analysis for S2G mutation in SHOC2 and maternal cell contamination studies (MCC)</td>
<td>83891 x 1, 83892 x 1, 83894 x 1, 83898 x 2, 83900 x 1, 83901 x 15, 88271 x 41, 83909 x 16, 83912 x 1</td>
<td>$2500</td>
<td>2-3 weeks (if submitting cultured AF or CV or direct CV. Direct amniotic fluid will need to be cultured, which increases turnaround time)</td>
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** For contracted institutions

Possible ICD9 Codes: Congenital malformation syndrome = 759.89

References: