Connective Tissue Disorders  
( Marfan, Loeys-Dietz, Thoracic Aortic Aneurysmal Dissections [TAAD], Ehlers-Danlos [EDS], Stickler, Cutis Laxa, Marfan-like and related disorders )

Indication-Specific Gene Panel
Including: sequencing and high resolution deletion/duplication analysis

PANEL DESCRIPTION:
Individuals with inherited connective tissue disorders frequently have an aberration in a gene involved in the structure or function of connective tissue. Such disorders commonly include issues with the joints, eyes, skin, and cardiovascular system, though other body systems are often affected as well. This indication-specific panel includes 50 genes associated with connective tissue disorders and is designed to detect both sequence-based mutations and small-scale deletions or duplications in these genes of interest.

PANEL DETAILS:
- This panel includes both sequencing and high resolution deletion/duplication analysis of the genes specified.
  - **Sequencing** is performed using a customized next generation sequencing library. Analysis includes the coding exons of all genes in the panel plus ten bases into the introns and untranslated regions (5' and 3'). Sanger sequencing is performed to confirm variants suspected or confirmed to be pathogenic.
  - **Deletion/duplication analysis** is performed using a high resolution, custom microarray platform designed to target the genes of interest at the exon level.
- Detection rates are limited to the genes specified; this test does not provide whole genome analysis.
- Gene panels are a more cost-effective approach than single gene testing to confirm or establish a diagnosis. However, if single gene testing is desired for the patient or family members of an individual with a known mutation, that must be ordered separately.

RECOMMENDED TESTING STRATEGY:
Tests below can be ordered individually, however our laboratory’s recommended Comprehensive Testing for connective tissue disorders includes the following two tests:

**Next Generation Sequencing**
- Inherited connective tissue disorders are frequently caused by an aberration in a gene involved in the structure or function of connective tissue.

**Targeted Deletion/Duplication Analysis**
- If no pathogenic aberrations are detected by NGS, deletion/duplication analysis is performed to identify partial or whole gene deletions and duplications in the associated genes.

DISORDERS INCLUDED IN THIS PANEL:
- Aortic valve disease  [NOTCH1]
- Arterial Tortuosity  [SLC2A10]
- Brittle cornea syndrome  [ZNF469, PRDM5]
- Dextrocardia  [SMAD2]
- Ehlers-Danlos syndrome (EDS)
  - Arthrochalasia type  [COL1A1, COL1A2]
  - Cardiac valvular form  [COL1A2]
- Dermatosparaxis \([ADAMTS2]\)
- EDS with progressive kyphoscoliosis, myopathy, and hearing loss \([FKBP14]\)
- Hypermobility (type III) \([TNXB]\) – GeneReviews* [http://www.ncbi.nlm.nih.gov/books/NBK1279/]
- Kyphoscoliotic (type VI) \([PLOD1]\) – GeneReviews* [http://www.ncbi.nlm.nih.gov/books/NBK1462/]
- Musculocontractural (type I) \([CHST14]\)
- Occipital horn (type IX) \([ATP7A]\)
- Periventricular heterotopia variant \([PVNH4]\) \([FLNA]\)

**ELN-related conditions**
- Autosomal dominant supravalvular aortic stenosis
- Cuts laxa \([ALDH18A1, ATP6VOA2, EFEMP2, FBLN5, LTBP4, PYCR1]\)

**FBN1-related disorders**
- Geleophysic dysplasia \([ADAMTSL2, FBN1]\)
- Isolated ectopia lentis \([FBNI]\)
- MASS phenotype \([FBNI]\)

**FLNA-related cardiac valvular disease**

**Familial thoracic aneurysms and aortic dissections (TADD)** \([ACTA2, FBN1, MYH11, MYLK, PRKG1, SMAD3, TGFB1, TGFB2]\) – GeneReviews* [http://www.ncbi.nlm.nih.gov/books/NBK1120/]


**Myhre** \([SMAD4]\)

**Polycystic kidney disease** \([PKD2]\)

**Pseudoxanthoma Elasticum** \([ABCC6]\)

**Sphrintzen-Goldberg** \([SKI]\)

**Stickler syndrome** – GeneReviews* [http://www.ncbi.nlm.nih.gov/books/NBK1302/]
- Type I \([COL2A1]\)
- Type II \([COL11A1]\)
- Type III \([COL11A2]\)
- Type IV \([COL9A1]\)
- Type V \([COL9A2]\)
- Type VI \([COL9A3]\)

**INDICATIONS FOR TESTING:**
- Joint issues: hypermobility, dislocation, chronic pain
- Skin findings: cuts laxa, abnormal or atrophic scars, poor wound healing, spontaneous bruising, thin, translucent, highly elastic, velvety
- Tissue fragility: inguinal and umbilical hernia, hiatal and incisional hernia, rectal prolapse in early childhood
- Auditory: hearing loss, hypermobile tympanic membranes
- Ocular findings: ectopia lentis, myopia, retinal detachment, scleral fragility and rupture of the ocular globe, characteristic vitreous changes or retinal abnormalities
- Facial features: malar hypoplasia, broad or flat nasal bridge, micro/retrognathia, cleft palate
- Pneumothorax, hemopneumothorax
- Cardiac findings: congenital heart defect, mitral valve prolapse, aortic root enlargement, thoracic aneurysm, aortic dissection, other aneurysms/dissections, extra-aortic vascular events, arterial tortuosity
- Skeletal variants: tall or short stature, pectus excavatum, pectus carinatum, arachnodactyly, brachydactyly, pes planus, long wingspan, femoral head failure, radiographically demonstrated osteoarthritis before age 40, scoliosis, spondylolisthesis or Scheurermann-like kyphotic deformity, congenital clubfoot, Marfanoid appearance
- Personal or family history of: sudden cardiac death, aneurysm/dissection, rupture of internal organ, rectal or uterine prolapse
- Craniofacial anomalies or dysmorphic features
SPECIMEN COLLECTION & TRANSPORT:
Complimentary test kits are available upon request, but are not required.

SAMPLE TYPE and REQUIREMENTS:
- **blood, > 3 months of age:** 3-5 ml whole blood in an EDTA tube (purple top)
- **blood, newborn:** 1-3 ml whole blood in an EDTA tube (purple top)
- **buccal swab:** 5 swabs
- **extracted DNA:** 5 µg in a DNA microcentrifuge tube

SHIPPING:
- Maintain and ship samples at room temperature.
- Coordinate transport for sample to be received in our laboratory within 24-48 hours of collection.
  - **LOCAL:** Call 402-559-5070 (option 1)
  - **OUT OF AREA:** Prior to shipment, please fax the completed test request form to 402-559-7248, including the FedEx® airbill tracking number.
    - Saturday delivery MUST be checked when sending FedEx® on Friday.
    - Please include Internal Billing Reference # 3155070600 on the FedEx® airbill.
    - **Ship To:** Human Genetics Laboratory – Zip 5440
      UNMC Shipping & Receiving Dock
      601 S. Saddle Creek Road
      Omaha, NE 68106

REQUIRED FORM: The following form can be downloaded via our website.
- Postnatal Test Request Form

OPTIONAL FORM:
- Informed Consent for Genetic Testing

POTENTIAL TEST RESULTS:
Once a variant is confirmed, our laboratory team interprets this information in conjunction with the patient’s clinical findings and the scientific literature in order to classify a finding. There are three possible results:
- **A normal** result indicates that sequencing or deletion/duplication analysis of the genes analyzed did not find any pathogenic mutations or variants of uncertain clinical significance (or no clinically-significant chromosome anomalies were identified by microarray analysis).
- An **abnormal (or pathogenic)** result indicates that a pathogenic mutation was identified (or microarray analysis identified a genomic dosage anomaly [deletion or duplication] or ROH that likely provides an explanation for the individual’s clinical findings). Any available information regarding the phenotype associated with that mutation will accompany the technical details on the report.
- In some cases, the clinical significance of an identified sequence variant (or chromosomal anomaly detected by microarray) may not be well understood. These variants (anomalies) will be reported as **variants of uncertain clinical significance (UCS)**. Any available information about the molecular characteristics of the genetic change and the relationship of the genetic change to phenotype will be included on the report. Over time, as more patients are reported, a variant of uncertain clinical significance may be revised to an informative result, and a revised report will be generated.
- Parental testing may be recommended in order to classify the result as de novo or familial for the purpose of recurrence risk calculation.

TURN-AROUND-TIMES: For all sample types, results are typically available in 2-6 weeks.

BILLING: Our laboratory offers patient/self-pay, insurance (including Medicare/Medicaid), and client/institution billing options. Verifying coverage requirements or obtaining preauthorization PRIOR TO OR AT THE TIME OF SPECIMEN COLLECTION is often necessary. We provide preauthorization services upon request by calling 402-559-5070 (option 3); the following form is helpful for obtaining the information required by insurance providers and can be downloaded via our website.
- Request for Insurance Preauthorization
In some circumstances, a test may be warranted even though insurance coverage is denied or not guaranteed. For these situations, we request the following form be signed by the patient and submitted with the sample. This helps inform patients of their potential financial responsibility, should the costs of genetic testing not be paid by their insurance provider.

- Advanced Beneficiary Notice of Noncoverage (ABN) – required when billing Medicare

**CPT CODES:**

50 Gene Panel

- Next Generation Sequencing: 81407(x2)
- Targeted Deletion/Duplication Analysis: 81228

**Follow-up Gene Panel**

Performed after negative results from Cutis Laxa, EDS, or Stickler panel

- Next Generation Sequencing: 81408
- Targeted Deletion/Duplication Analysis: no charge

**PRICING:** For current costs contact the laboratory billing staff at 402-559-5070 (option 3).

**GENE LIST:** (expanded panel, version 3_50)

This 50 gene panel is applied to all testing performed on and after April 1, 2016.

<table>
<thead>
<tr>
<th>ABCC6</th>
<th>COL3A1</th>
<th>FKBP14</th>
<th>SLC2A10</th>
</tr>
</thead>
<tbody>
<tr>
<td>ACTA2</td>
<td>COL5A1</td>
<td>FLNA</td>
<td>SLC39A13</td>
</tr>
<tr>
<td>ADAMTS2</td>
<td>COL5A2</td>
<td>LTBP4</td>
<td>SMAD2</td>
</tr>
<tr>
<td>ADAMTS10</td>
<td>COL9A1</td>
<td>MED12</td>
<td>SMAD3</td>
</tr>
<tr>
<td>ADAMTS1L2</td>
<td>COL9A2</td>
<td>MYH11</td>
<td>SMAD4</td>
</tr>
<tr>
<td>ALDH18A1</td>
<td>COL9A3</td>
<td>MYLK</td>
<td>TGFB2</td>
</tr>
<tr>
<td>ATP6V0A2</td>
<td>COL11A1</td>
<td>NOTCH1</td>
<td>TGFB3</td>
</tr>
<tr>
<td>ATP7A</td>
<td>COL11A2</td>
<td>PKD2</td>
<td>TGFB1</td>
</tr>
<tr>
<td>CBS</td>
<td>EFEMP2 (FBLN4)</td>
<td>PLOD1</td>
<td>TGFB2</td>
</tr>
<tr>
<td>CHST14</td>
<td>ELN</td>
<td>PRDM5</td>
<td>TNXB</td>
</tr>
<tr>
<td>COL1A1</td>
<td>FBLN5</td>
<td>PRKG1</td>
<td>ZNF469</td>
</tr>
<tr>
<td>COL1A2</td>
<td>FBN1</td>
<td>PYCR1</td>
<td></td>
</tr>
<tr>
<td>COL2A1</td>
<td>FBN2</td>
<td>S1</td>
<td></td>
</tr>
</tbody>
</table>

updated 4/1/2016