Clinical Features:
Primordial Dwarfism is characterized by growth that is profoundly restricted from early in development which continues postnatally. In line with other parts of the body, most individuals with primordial dwarfism also have a reduction in head size in proportion to, or smaller than, their body size (1). Microcephalic Primordial Dwarfism is a heterogeneous group of disorders that include Seckel Syndrome, Microcephalic Osteodysplastic Primordial Dwarfism and Meier-Gorlin syndrome. Core features of these groups include severe intrauterine and postnatal growth deficiency, severe postnatal short stature, primary microcephaly, characteristic facial features and variable intellectual disability.

Seckel syndrome (OMIM#210600) is characterized by severe proportionally short stature with severe microcephaly (mean postnatal growth retardation is -7SD with a range from -5 to -13SD; mean OFC is -9SD with a range from -4 to -14SD), a ‘bird like’ profile include a receding forehead, large eyes, beak-like protusion of the nose, narrow face, receding lower jaw and micrognathia, and intellectual disability (2, 3).

Microcephalic Osteodysplastic Primordial Dwarfism type II (MOPD II, OMIM#210720) is differentiated from Seckel syndrome by more severe growth retardation, radiological abnormalities, and absent or mild mental retardation. The radiological abnormalities in MOPD II are short limbs with preferential distal involvement, coxa vara, epiphysiolysis and metaphyseal flaring with V-shaped distal femora metaphyses (2, 3). Typically, intellect is well preserved.

Microcephalic Osteodysplastic Primordial Dwarfism type I (MOPD I, OMIM#210710) overlaps clinically with other types of Primordial Dwarfism, however it is differentiated by the presence of brain malformations and early lethality (4). There is considerable phenotypic variability, with regard to the type and severity of brain malformations, the degree of developmental delay, and total lifespan, even within the same family (4).

Meier-Gorlin syndrome is defined by absent/hypoplastic patellae and markedly small ears. Many cases have normal intellect with proportionate microcephaly. Growth failure is variable and can be mild (1).

There is substantial clinical heterogeneity and overlap between these and other groups.

Our Seckel Syndrome Sequencing Panel includes all eight genes listed below.
Our Meier-Gorlin Syndrome Sequencing Panel includes all five genes listed below
Our Comprehensive Primordial Dwarfism Sequencing Panel includes all fifteen genes listed below, with the exception of LIG4 and RNU4ATAC.

<table>
<thead>
<tr>
<th>Gene</th>
<th>Molecular Pathology</th>
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<tbody>
<tr>
<td>ATR [OMIM#601215]</td>
<td>Several genes, including ATR, CEP63, CEP152, CENPJ, RBBP8 and NIN have been reported to cause Seckel syndrome in a small number of families</td>
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<tr>
<td>CEP63 [OMIM#614728]</td>
<td>A homozygous synonymous mutation in ATR [OMIM#601215] has been identified in two consanguineous Pakistani families with Seckel syndrome (5). ATR is a central player in a signaling response to DNA damage.</td>
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<td>CENPJ [OMIM#609279]</td>
<td>A homozygous nonsense mutation was identified in CEP63 in a consanguineous family of Pakistani descent with three members with primary microcephaly and</td>
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<tr>
<td>Gene/OMIM</td>
<td>Description</td>
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<tr>
<td>RBBP8 [OMIM#604124]</td>
<td>Mutations in RBBP8 have been identified in multiple families with MOPD I, including one large Amish cohort with a founder mutation [9]. RNU4ATAC encodes U4atac, which is a small nuclear RNA that is a crucial component of the minor spliceosome, and is required for proper excision of the U12-dependent class of introns, which are found in many essential genes [19].</td>
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<tr>
<td>NIN [OMIM#608684]</td>
<td>Mutations in NIN have been identified in 70% of patients reported to date (13). Ninein is a centrosomal protein required for the centrosome to function as a microtubule-organizing center. CENPJ and CEP152 are also genes implicated in autosomal recessive primary microcephaly (MCPH). RBBP8 mutations have also been implicated in Jawad syndrome [OMIM#251255], which has clinical overlap with Seckel syndrome.</td>
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<tr>
<td>PCNT [OMIM#605925]</td>
<td>Mutations in PCNT have been identified in patients with MOPD II/Seckel syndrome. PCNT is located at 21q22.3 and studies have shown that the absence of PCNT results in disorganized mitotic spindles and missegregation of chromosomes. Rauch et al. (2008) identified 29 different homozygous or compound heterozygous mutations in the PCNT gene in 25 patients with MOPD II (12). Williams et al. (2010) identified 12 homozygous and 1 heterozygous mutation in the PCNT gene in 8/8 patients with MOPDII and 5/16 patients diagnosed with Seckel syndrome (3). Clinical analysis of Seckel cases with PCNT mutations showed that they all presented with minor skeletal changes and clinical features compatible with a MOPDII diagnosis.</td>
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<tr>
<td>LIG4 [OMIM#601837]</td>
<td>Biallelic truncating LIG4 mutations have been identified in a series of 11 patients with microcephalic primordial dwarfism (13). The position of truncating mutations has been observed to correlate with phenotypic severity (13). LIG4 mutations have been recurrently reported in Ligase IV syndrome (OMIM#606593), originally identified in a typically-developing 14-year-old with acute lymphoblastic leukemia (14). Bone marrow failure has occurred in 70% of patients reported to date (13). Other patients with LIG4 syndrome are described as having immunodeficiency, developmental delay, Seckel or “bird-like” facies, microcephaly, and growth restriction (15).</td>
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<tr>
<td>ORC1 [OMIM#601902]</td>
<td>Mutations in ORC1 have been identified in 4/33 individuals with Meier-Gorlin syndrome (MGS) (16). Mutation analysis of other genes of this pre-replication complex showed mutations in ORC4, ORC6, CDT1 and CDC6 in 14 individuals from nine families with MGS (17). Guernsey et al. identified mutations in ORC1, ORC4 and CDT1 in 8 individuals from five families with MGS (17). While most affected individuals described had typical features of MGS, a considerable wide phenotypic variation was observed and no clear genotype-phenotype correlation has been elucidated. Mutations in genes from the pre-replication complex are expected to disturb the process of DNA replication (18).</td>
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<tr>
<td>ORC4 [OMIM#603056]</td>
<td>Mutations in RNU4ATAC have been identified in multiple families with MOPD I, including one large Amish cohort with a founder mutation [9]. RNU4ATAC encodes U4atac, which is a small nuclear RNA that is a crucial component of the minor spliceosome, and is required for proper excision of the U12-dependent class of introns, which are found in many essential genes (19).</td>
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<tr>
<td>ORC6 [OMIM#607213]</td>
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<td>CDT1 [OMIM#605525]</td>
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<tr>
<td>CDC6 [OMIM#602627]</td>
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Alazami et al., 2012 identified a homozygous 7bp duplication in LARP7 in a consanguineous Saudi family with facial dysmorphism, intellectual disability and primordial dwarfism (20). All patients had growth parameters 3.5 SD below mean. Consistent dysmorphic features included malar hypoplasia, deep-set eyes, broad nose, short philtrum and macrostomia.

POC1A[OMIM#614783] A homozygous POC1A nonsense mutation has been identified in 5 children from 3 consanguineous Saudi families with primordial dwarfism and distinctive facial features (SOFT syndrome, OMIM#614813) (21). SOFT syndrome is characterized by severely short long bones, characteristic facies with paucity of hair and nail anomalies. Facial dysmorphism include a long triangular face with prominent nose and small ears, and affected individuals have an unusual high-pitched voice.

Inheritance:
Seckel syndrome, MOPD I, MOPD II and Meier-Gorlin syndrome are inherited in an autosomal recessive pattern. Parents of an affected child are most likely obligate carriers. Recurrence risk for carrier parents is 25%.

Test methods:
Comprehensive sequence coverage of the coding regions and splice junctions of all genes in this panel is performed. Targets of interests are enriched and prepared for sequencing using the Agilent SureSelect system. Sequencing is performed using Illumina technology and reads are aligned to the reference sequence. Variants are identified and evaluated using a custom collection of bioinformatic tools and comprehensively interpreted by our team of directors and genetic counselors. All novel and/or potentially pathogenic variants are confirmed by Sanger sequencing. The technical sensitivity of this test is estimated to be >99% for single nucleotide changes and insertions and deletions of less than 20 bp.

Note: only the following known pathogenic variants are analyzed in the non-coding RNU4ATAC gene: g.30G>A, g.50G>C, g.51G>C, g.53C>G, g.55G>A, g.66G>A, g.111G>A, g.124G>A. This testing does not rule-out other sequence changes in the RNU4ATAC gene.

Deletion/duplication analysis is available for all the genes listed above, with the exception of RNU4ATAC, and is performed by oligonucleotide array-CGH. Partial exonic copy number changes and rearrangements of less than 400 bp may not be detected by array-CGH. Array-CGH will not detect low-level mosaicism, balanced translocations, inversions, or point mutations that may be responsible for the clinical phenotype. The sensitivity of this assay may be reduced when DNA is extracted by an outside laboratory.

Sequencing and deletion/duplication testing of the PCNT gene is also offered separately.

Comprehensive Primordial Dwarfism Sequencing Panel (16 genes: ATR, CDT1, CDC6, CENPJ, CEP63, CEP152, LIG4, NIN, ORC1, ORC4, ORC6, PCNT, LARP7, POC1A, RBBP8, and RNU4ATAC)
Sample specifications: 3 to 10 cc of blood in a purple top (EDTA) tube
Cost: $4500
CPT codes: 81407
Turn-around time: 8 weeks
Note: We cannot bill insurance for this test.

Comprehensive Primordial Dwarfism Deletion/Duplication Panel (14 genes: ATR, CDT1, CDC6, CENPJ, CEP63, CEP152, LARP7, NIN, ORC1, ORC4, ORC6, PCNT, POC1A, and RBBP8)
Sample specifications: 3 to 10 cc of blood in a purple top (EDTA) tube
Cost: $2500
CPT codes: 81407
Turn-around time: 6 weeks

Seckel Syndrome Sequencing Panel (PCNT, ATR, CENPJ, CEP152, LIG4, NIN, RBBP8 and CEP63)
Sample specifications: 3 to 10 cc of blood in a purple top (EDTA) tube
Cost: $3500
CPT codes: 81407
Turn-around time: 8 weeks
Note: We cannot bill insurance for this test.
Seckel Syndrome Deletion/Duplication Panel (PCNT, ATR, CENPJ, CEP152, NIN, RBBP8, and CEP63)

Sample specifications: 3 to 10 cc of blood in a purple top (EDTA) tube
Cost: $1545
CPT codes: 81407
Turn-around time: 6 weeks

Meier-Gorlin Syndrome Sequencing Panel (ORC1, ORC4, ORC6, CDT1, CDC6)

Sample specifications: 3 to 10 cc of blood in a purple top (EDTA) tube
Cost: $3500
CPT codes: 81407
Turn-around time: 8 weeks

Note: We cannot bill insurance for this test.

PCNT sequencing analysis

Sample specifications: 3 to 10 cc of blood in a purple top (EDTA) tube
Cost: $2650
CPT codes: 81407
Turn-around time: 4 weeks

PCNT deletion/duplication analysis

Sample specifications: 3 to 10 cc of blood in a purple top (EDTA) tube
Cost: $1000
CPT codes: 81406
Turn-around time: 4 weeks

Results:
Results, along with an interpretive report, are faxed to the referring physician as soon as they are completed. Additional reports are available as requested. All abnormal results are reported by telephone.

References:


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