



Next Generation Sequencing Panels for Disorders of Sex Development

Disorders of Sex Development – Overview

Disorders of sex development (DSDs) occur when sex development does not follow the course of typical male or female patterning. Types of DSDs include congenital development of ambiguous genitalia, disjunction between the internal and external sex anatomy, incomplete development of the sex anatomy, and abnormalities of the development of gonads (such as ovotestes or streak ovaries) (1). Sex chromosome anomalies including Turner syndrome and Klinefelter syndrome as well as sex chromosome mosaicism are also considered to be DSDs.

DSDs can be caused by a wide range of genetic abnormalities (2). Determining the etiology of a patient’s DSD can assist in deciding gender assignment, provide recurrence risk information for future pregnancies, and can identify potential health problems such as adrenal crisis or gonadoblastoma (1, 3).

Sex chromosome aneuploidy and copy number variation are common genetic causes of DSDs. For this reason, chromosome analysis and/or microarray analysis typically should be the first genetic analysis in the case of a patient with ambiguous genitalia or other suspected disorder of sex development. Identifying whether a patient has a 46,XY or 46,XX karyotype can also be helpful in determining appropriate additional genetic testing.

Abnormal/Ambiguous Genitalia Sequencing Panel

Our Abnormal/Ambiguous Genitalia Sequencing Panel includes sequence analysis of 72 genes associated with both syndromic and non-syndromic DSDs. This comprehensive panel evaluates a broad range of genetic causes of ambiguous or abnormal genitalia, including conditions in which abnormal genitalia are the primary physical finding as well as syndromic conditions that involve abnormal genitalia in addition to other congenital anomalies.

Our Abnormal/Ambiguous Genitalia Sequencing Panel includes all 72 genes listed below.

Table with 8 columns and 15 rows listing genes: AKR1C2, CREBBP, DYNC2H1, HCCS, LMNA, PTPN11, SRD5A2, WT1, AR, CYB5A, ESCO2, HOXA13, MAP3K1, RIPK4, SRY, ZFPM2, ARX, CYP11A1, FAM58A, HSD17B3, MKKS, ROR2, STAR, ATRX, CYP11B1, FAT4, HSD3B2, MKS1, RSPO1, TBX15, B3GALTL, CYP17A1, FEZF1, ICK, NEK1, SALL1, TCTN3, BCOR, CYP19A1, FIG4, IL17RD, NR0B1, SCARF2, TSPYL1, BMP4, DHCR24, FRAS1, IRF6, NR5A1, SEMA3A, UBR1, CDKN1C, DHCR7, FREM2, KAL1, NSMF, SETBP1, WDR60, CEP41, DHH, GATA4, KISS1R, OPHN1, SOX9, WNT4, CHD7, DNMT3B, GRIP1, LHCGR, POR, SPECC1L, WNT7A

46,XY Disorders of Sex Development/Complete Gonadal Dysgenesis Sequencing Panel

The 46,XY Disorders of Sex Development/Complete Gonadal Dysgenesis (46,XY DSD/CGD) Sequencing Panel includes sequence analysis of 26 genes associated with disorders of sex development (DSD) or complete gonadal dysgenesis (CGD) in patients with a 46,XY karyotype. Individuals with 46,XY CGD (also known as 46,XY sex reversal) have a 46,XY karyotype in conjunction with normal female external genitalia, “streak” gonads, absent sperm production, and presence of a uterus and fallopian tubes. 46,XY CGD has been associated with mutations or copy number variations in several genes, including SRY, DHH, NR5A1, and SOX9 (with campomelic dysplasia) (2).

46,XY DSDs are characterized by one or more of the following in an individual with a 46,XY karyotype: ambiguous genitalia with mild to severe penoscrotal hypospadias, dysgenetic gonads, reduced/absent sperm production, and Müllerian structures that range from absent to presence of a uterus and fallopian tubes (4). Example of genes associated with 46,XY DSDs include *AKR1C2*, *CYB5A*, *GATA4*, and *SRD5A2*. In addition, a 46,XY DSD phenotype may be syndromic, as in Mowatt-Wilson syndrome (5), X-linked lissencephaly-2 (6), ATR-X syndrome (7), and Smith-Lemli-Opitz syndrome (8).

Our 46,XY DSD/CGD Sequencing Panel includes 26 genes listed below.

Our 46,XY DSD/CGD Del/Dup Panel includes 26 genes listed below.

46,XY DSD/CGD Sequencing Panel (26 genes)			
AKR1C2	CYB5A	HCCS	SOX9
AMH	CYP11A1	HSD17B3	SRD5A2
AMHR2	CYP17A1	LHCGR	SRY
AR	DHCR7	MAMLD1	WT1
ARX	DHH	MAP3K1	ZFPM2
ATRX	DYNC2H1	NR5A1	
B3GALT1	GATA4	OPHN1	

46,XX Disorders of Sex Development/Complete Gonadal Dysgenesis Sequencing Panel

The 46,XX Disorders of Sex Development/Complete Gonadal Dysgenesis (46,XX DSD/CGD) Sequencing Panel includes 9 genes associated with disorders of sex development (DSD) or complete gonadal dysgenesis (CGD) in patients with a 46,XX karyotype. A range of phenotypes may be observed in patients with 46,XX DSD/CGD, from müllerian aplasia and hyperandrogenism (9) or ovarian dysgenesis (10), to adrenal hyperplasia with overvirilization. There are syndromic forms of 46,XX DSDs, including Peters-Plus syndrome (11) and WAGR syndrome (12). 46,XX complete gonadal dysgenesis (also referred to as 46,XX sex reversal, 46,XX true hermaphroditism, or ovotesticular DSD) may be observed in patients with presence of the *SRY* gene (13), in patients with copy number variations in *SOX3* (14), or due to mutations in other genes, such as *RSPO1* (15). Please note that due to the presence of a pseudogene, the *CYP21A2* gene is not included on this panel.

Our 46,XX DSD/CGD Sequencing Panel includes 9 genes listed below.

Our 46,XX DSD/CGD Del/Dup Panel includes 9 genes listed below.

46,XX DSD/CGD Sequencing Panel (9 genes)		
B3GALT1	HCCS	RSPO1
CYP11B1	NR5A1	WNT4
CYP19A1	PSMC3IP	WT1

Test methods:

Comprehensive sequence coverage of the coding regions and splice junctions of all genes in the 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 pathogenic and likely 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.

Deletion/duplication analysis of the panel genes 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.

Abnormal or Ambiguous Genitalia Sequencing Panel (72 genes sequencing)

Sample specifications: 3 to10 cc of blood in a purple top (EDTA) tube
Cost: \$4000
CPT codes: 81407
Turn-around time: 8 weeks

Note: We cannot bill insurance for the above test

46,XY DSD/CGD Sequencing Panel (26 genes sequencing)

Sample specifications: 3 to10 cc of blood in a purple top (EDTA) tube
Cost: \$3000
CPT codes: 81407
Turn-around time: 8 weeks

Note: We cannot bill insurance for the above test.

46,XY DSD/CGD Del/Dup Panel (26 genes deletion/duplication analysis)

Sample specifications: 3 to10 cc of blood in a purple top (EDTA) tube
Cost: \$2500
CPT codes: 81407
Turn-around time: 6 weeks

46,XX DSD Sequencing Panel (9 genes sequencing)

Sample specifications: 3 to10 cc of blood in a purple top (EDTA) tube
Cost: \$2000
CPT codes: 81407
Turn-around time: 8 weeks

Note: We cannot bill insurance for the above test.

46,XX DSD/CGD Del/Dup Panel (9 genes deletion/duplication analysis)

Sample specifications: 3 to10 cc of blood in a purple top (EDTA) tube
Cost: \$1545
CPT codes: 81407
Turn-around time: 6 weeks

Results:

Results, along with an interpretive report, will be faxed to the referring physician. Additional reports will be provided as requested. All abnormal results will be reported by telephone.

For more information about our testing options, please visit our website at dnatesting.uchicago.edu or contact us at 773-834-0555.

References:

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13. Nieto K, Peña R, Palma I et al. 45,X/47,XXX/47,XX, del(Y)(p?)/46,XX mosaicism causing true hermaphroditism. *Am J Med Genet A* 2004; 130A: 311-314.
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