Clinic *for* Special Children

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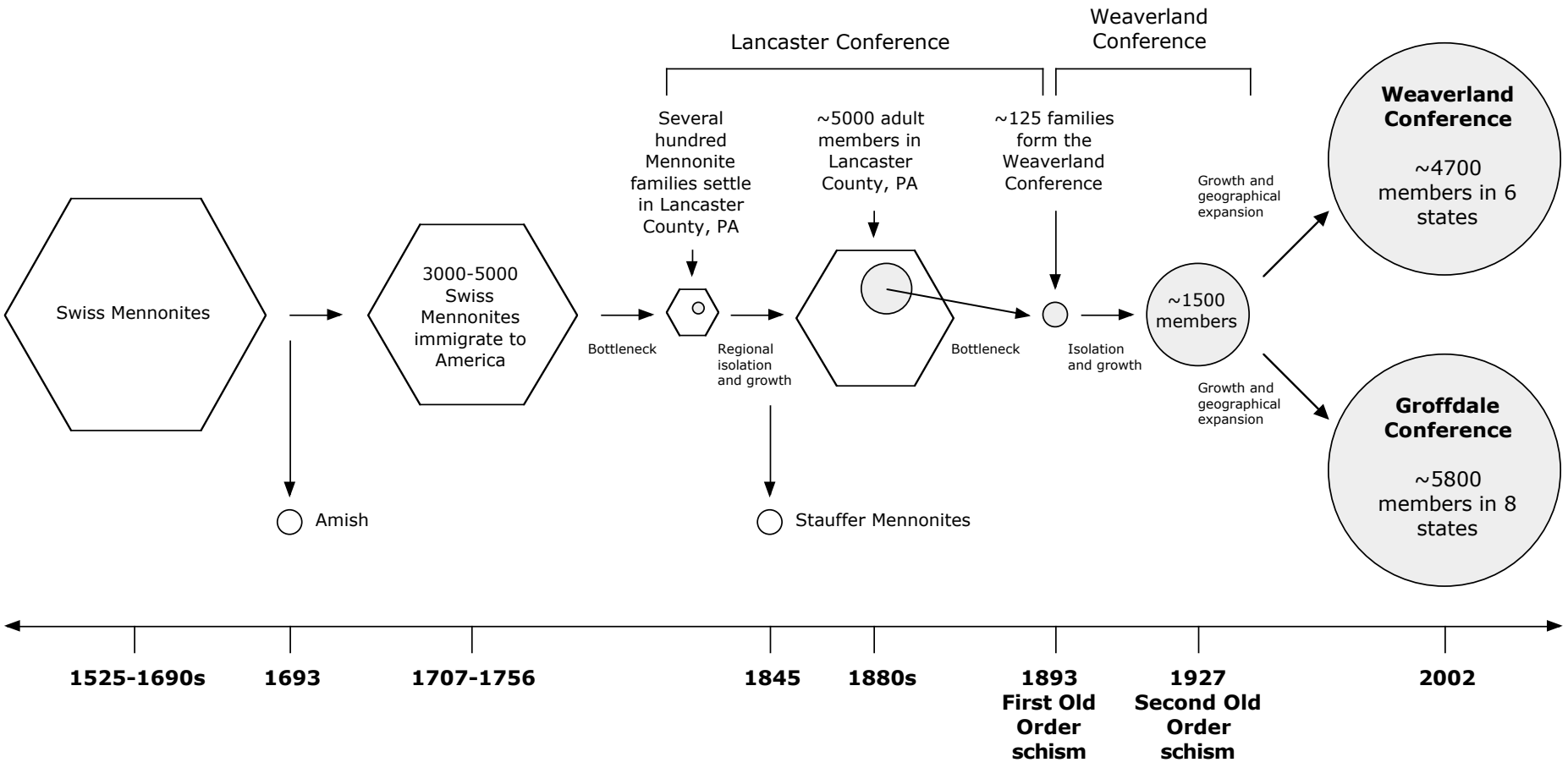
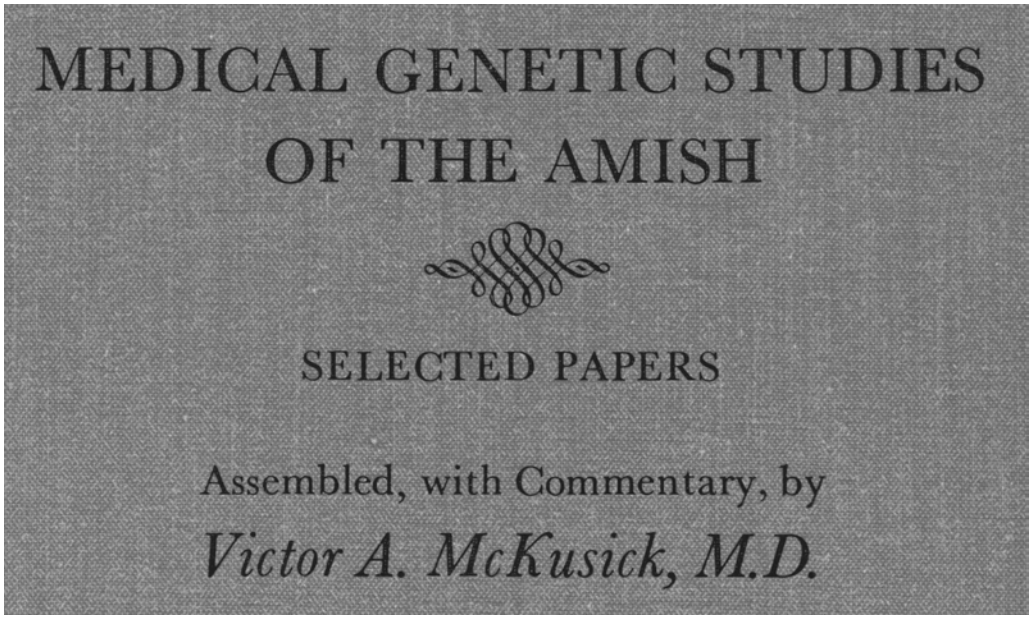
Social History of the Plain Populations



Clinical Genetic Research in Plain Populations



Published in 1978, this was a compilation of medical genetic research articles spanning nearly two decades regarding genetic diseases in the Old Order Amish.

Molecular Genetic Research in Plain Populations

With the advent of disease gene mapping technology in the early 1980s and polymerase chain reaction (PCR) in the late 1980s, numerous studies were initiated to map and identify disease genes in Plain populations. Below is an abridged list of the more notable studies where Plain populations from Pennsylvania were utilized to map and/or identify disease genes.

Disease	Reference	Population
Amish lethal microcephaly	Rosenberg et al., 2002	Amish
Byler disease	Bull et al., 1998	Amish
Ellis-van Creveld syndrome	Ruiz-Perez et al., 2000	Amish
Hirschsprung disease	Puffenberger et al., 1994	Mennonite
Maple syrup urine disease	Zhang et al., 1989	Mennonite
McKusick-Kauffman syndrome	Stone et al., 2000	Amish
Nemaline rod myopathy	Johnston et al., 2000	Amish
Sitosterolemia	Berge et al., 2000	Amish
Sudden infant death with dysgenesis of the testes	Puffenberger et al., 2004	Amish
3-methylcrotonylglycinuria	Baumgartner et al., 2001	Amish and Mennonite

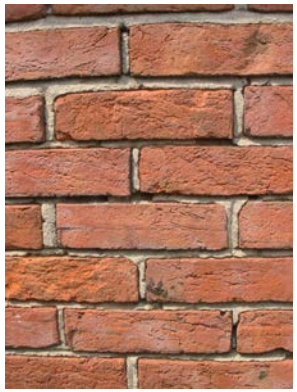
Bringing the Lab to the Clinic

Graduate/Medical School

Biological Sciences

Medical Physiology

LABORATORY



CLINIC

Laboratory Functions

Clinical testing
(amino acids, organic acids, genetic tests)

Basic research
(gene discovery, diagnostic dilemmas, population genetics)

Data analysis
(exomes, copy number variation, genotypes, mapping)

New test development
(LightScanner assays, PGM sequencing, ELISA tests)

Core Laboratory

- CLIA-certified lab
- Fee-based services
- Screening, diagnosis and management
- Routine labs sent to LGH and other reference labs

Why a Core Laboratory?

- In-house lab for clinical problem solving
- Flexible
- Tests depend on the patient population
- Rapid, low-cost testing that is either *unavailable* or *very expensive* elsewhere

Core Laboratory Service

	Commercial		CSC Lab				
TEST	Cost (dollars)	Turnaround	Cost (dollars)	Turnaround	Number per year	Dollars Saved	Time Saved (Days)
Amino acid analysis	700	5 days	75	45 minutes	1310	$517,450.00	5856
Organic acid analysis	247	6 days	85	4 hours	175	$26,862.50	1024
Targeted detection of known mutation	590	14 days	50	1-2 days	405	$144,787.50	6480
Complete gene sequencing(cost per exon)	147	11 days	35	5 days	15	$1,687.50	218
Cytogenetic microarray (DNA copy number)	1,654	21 days	600	4 days	¥145	$222,290.00	2973
TOTALS						$913,077.50	16,549

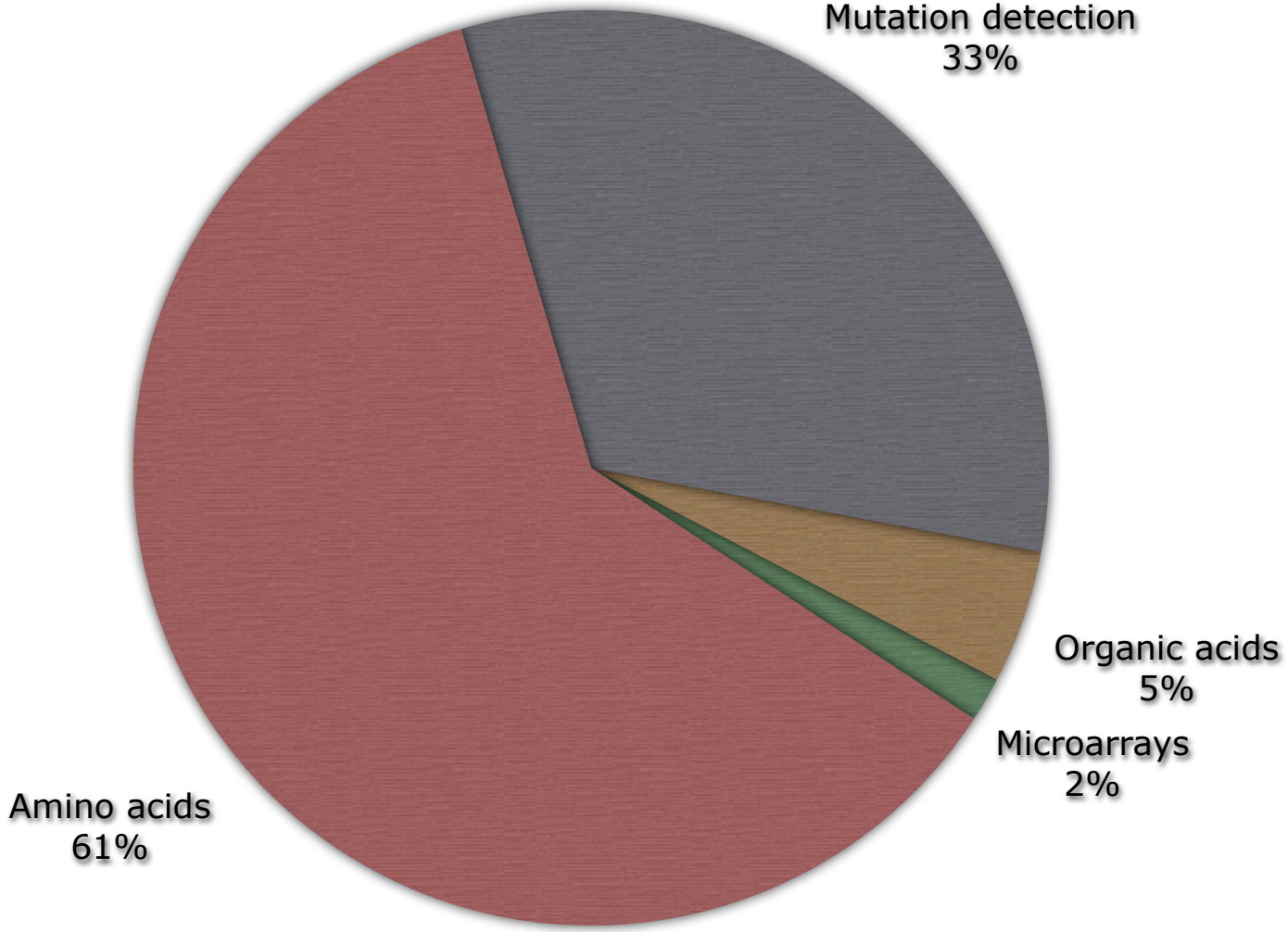
Laboratory Testing, FY 2012

Test	Number	Price
Amino acid quantitation	1467	$75
Targeted mutation detection	780	$50
Organic acid analysis	113	$85
Affymetrix microarray	38	$495
Comprehensive mutation identification	7	Varies

Total in-house tests = 2405

Total <u>outside</u> laboratory testing (LGH, etc.): 2135 tests

Clinic *for* Special Children



Clinic *for* Special Children

Laboratory Expenses, FY 2012

Laboratory Expenses*	$136,639.05
CSC Expenses*	$707,018.87
Proportion	**19.3%**

*excludes payroll

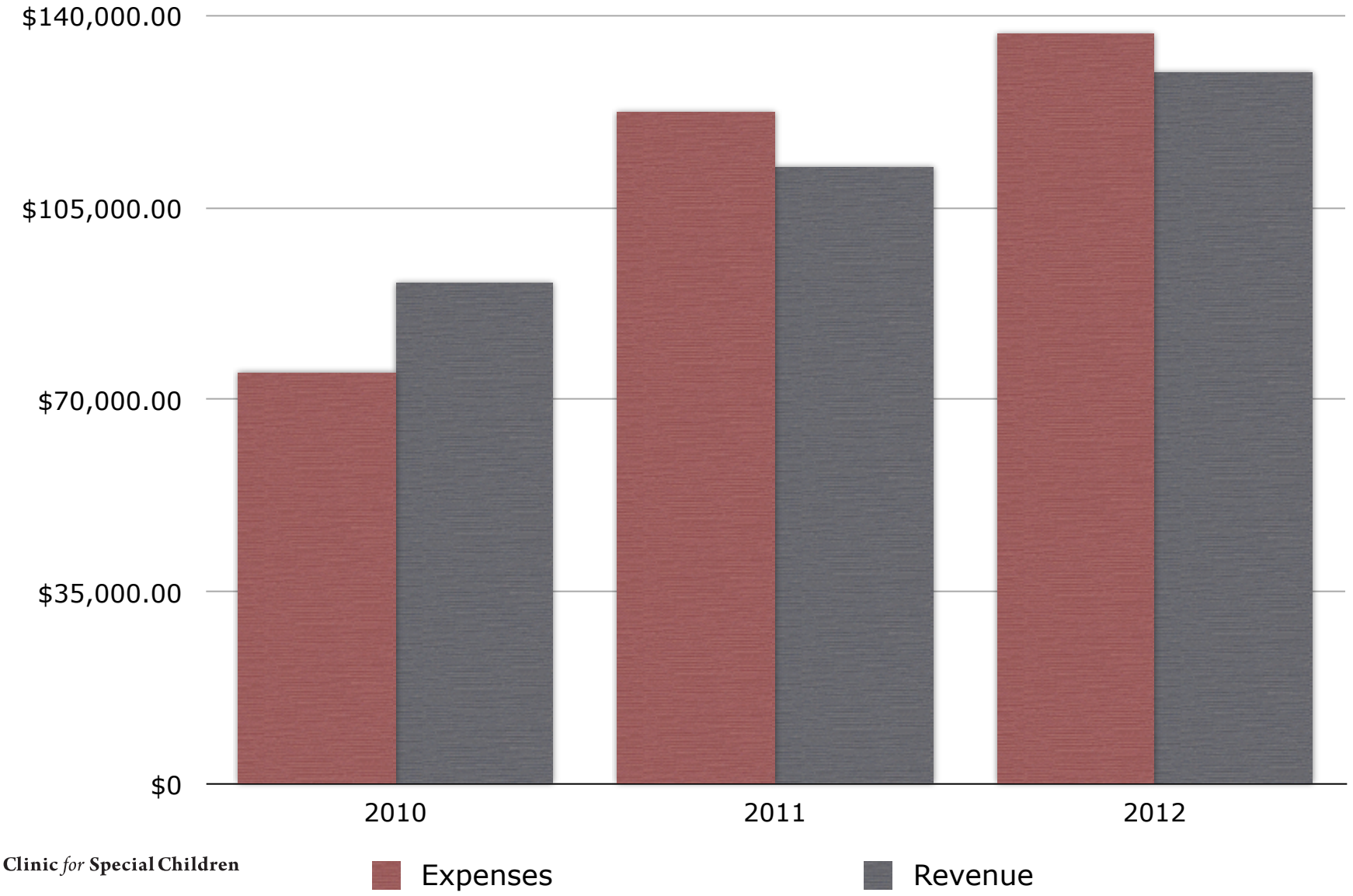
Clinic *for* Special Children

Expenses and Revenue, FY 2012

	Expenses*	Revenue
FY 2010	$74,924.79	$91,324.00
FY 2011	$122,337.41	$112,330.00
FY 2012	$136,639.05	$129,618.00

*excludes payroll

Expenses and Revenue, FY 2012



Clinic *for* Special Children

Research

Clinic for Special Children-Broad Institute Collaboration

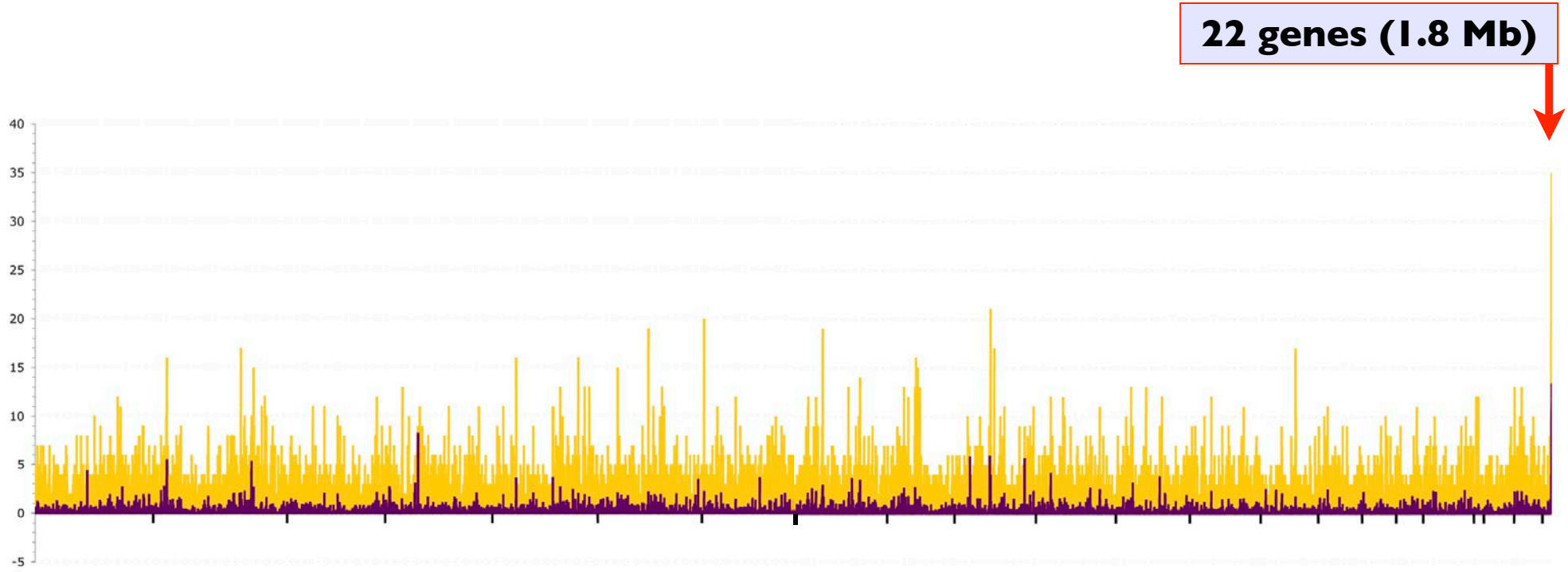
Unfinished Mapping Studies (November 2009)

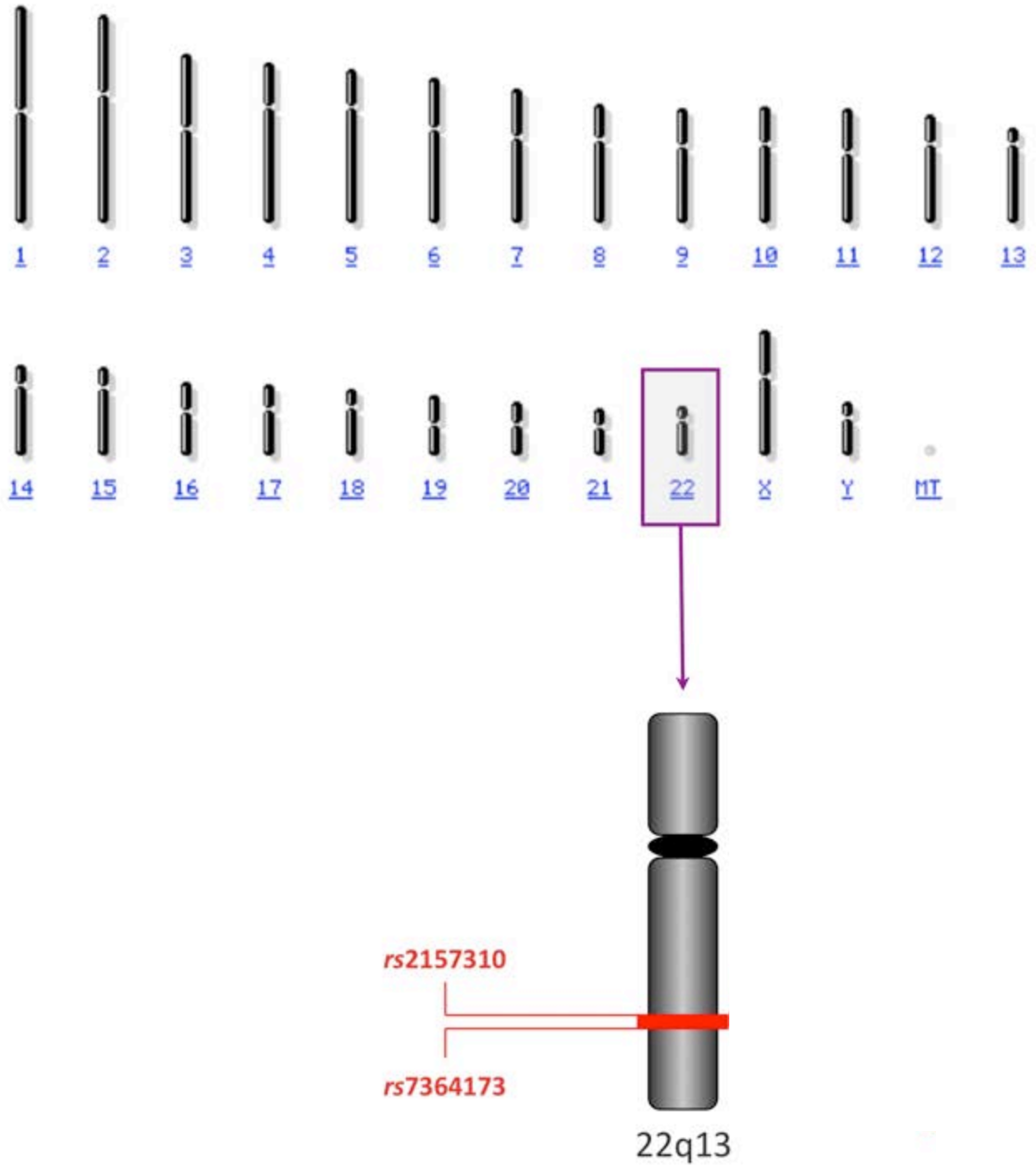
Disease	OMIM	Chr	Start	Stop	Mb	Genes
Posterior column ataxia with retinitis pigmentosa	609033	1	207.3	211.4	4.2	55
Craniosynostosis, Miller-Schrock	-	1	36.6	43.3	6.6	108
Usher-like syndrome	-	5	134.6	143.1	8.4	172
Lethal seizure syndrome	-	7	1.5	3.1	1.6	31
Mental retardation, non-syndromic	-	12	91.0	94.7	3.6	37
Yoder dystonia	-	15	79.2	83.1	4.0	69
Microcephaly with chorioretinopathy	251270	22	47.2	49.0	1.8	22
Hurst dystonia	-	-	-	-	-	-
Junctional ectopic tachycardia (JET)	-	-	-	-	-	-

Mennonite Microcephaly, 1966

Mennonite Microcephaly, 2006

Microcephaly with Chorioretinopathy: *50K Mapping*

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22
X
Y
MT
rs2157310
rs7364173
22q13

Microcephaly with Chorioretinopathy

CHANGE	ENTIRE EXOME		MAPPED INTERVAL ON 22q13	
	All	Novel *	All	Novel*
Inter-genic	19	7		
3'-UTR	26	3		
5'-UTR	22	2		
Intron	26	5		
Missense	8701	2562		
Nonsense	69	32		
Promoter	5	1	**4**	
Read-through	13	5	**1**	**1**
Splice site	66	46		
Synonymous	8288	830	**8**	
miRNA vicinity	35	19		
TOTAL	**17,270**	**3,512**	**13**	**1**

Eight Disorders in Twelve Months

Disease	OMIM	Gene	DNA Variant	Protein Variant
Posterior column ataxia with retinitis pigmentosa	609033	*FLVCR1*	c.361A>G	p.Asn121Asp
Craniosynostosis, Miller-Schrock	-	*SNIP1*	c.1097A>G	p.Glu366Gly
Usher-like syndrome	-	*HARS*	c.1361A>C	p.Tyr454Ser
Lethal seizure syndrome	-	*C7orf27*	c.638_639insA	
Mental retardation, non-syndromic	-	*CRADD*	c.382G>C	p.Gly128Arg
Yoder dystonia	-	*XXXXX*	xxxxxx	
Microcephaly with chorioretinopathy	251270	*TUBGCP6*	c.5458T>G	p.Ter1820Gly
Hurst dystonia	-	*SLC6A3*	IVS9+1G>T	
Junctional ectopic tachycardia (JET)	-	-	?	?

Efficiency of Exome Sequencing

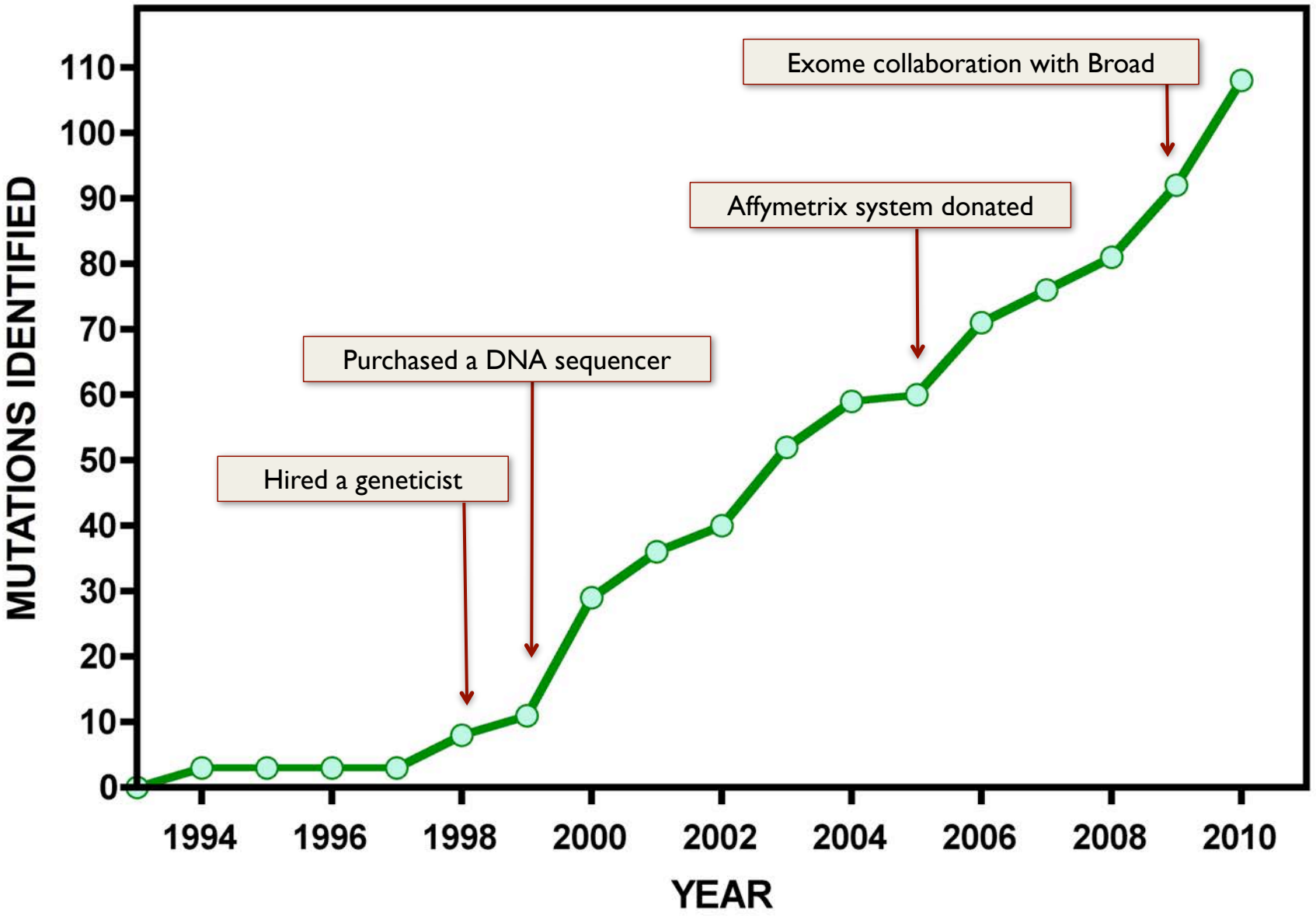
Mapped/multiple affecteds

>95%

Trios (singleton, parents)

~55% (first pass)

Cataloging Population-Specific Genetic Risk



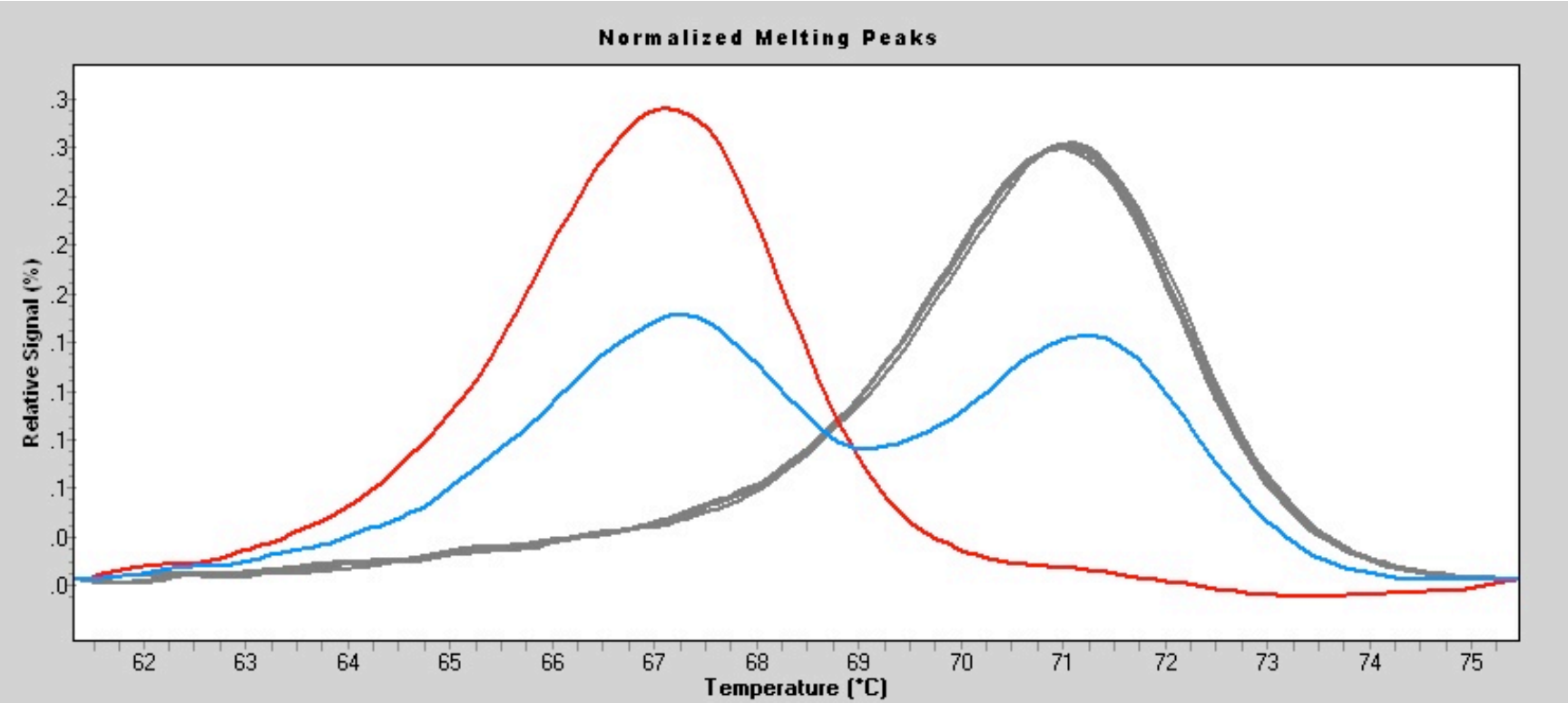
1000 Active Patients, **115** Genetic Disorders

ABCG8 1720G>A
ACADM 985A>G
ACADM IVS4-30A>G
ADA 646G>A
ADAMTS10 17,346 bp deletion
AMN 44 bp deletion
APOA4 552_749dup
ATP8B1 923G>T
BAAT 226A>G
BBS1 1169T>G
BCKDHA 1312T>A
BRAT1 638_639insA
BTD 1459T>C
BTD 1368A>C
BTD 1330G>C
C7orf10 895C>T
CAPN3 2306G>A
CFP 379T>G
CHST3 1298C>T
CLCNKB 22,508 bp deletion
CNGA3 1126G>A
CNTNAP2 3709delG
COL1A2 2098G>T
CRADD 382G>C
CYBB 1335C>A
CYP11B1 1343G>A
CYP11B2 5 bp deletion
DNAH5 4348C>T
EDNRB 828G>T
ERCC6 IVS14+1G>T
EVC IVS13+5G>T
F11 1327C>T
F5 1601G>A
FLVCR1 361A>G
FMR1 (CGG)n expansion
GALT 563A>G
GALT 940A>G
GCDH 1262C>T
GJB2 35delG
GJC2 203A>G
GLB1 902C>T
GLDC 2186delC
GLDC 128delA
HARS 1361A>C
HFE 187C>G
HFE 845G>A
HPD 85G>A
HPD 479A>G
HPD 1005C>G
HSD3B2 35G>A
IL7R 2T>G
ITCH 394_395insA
KRIT1 47G>C
LAMB2 440A>G
LMNA 568C>T
LRP5 1225A>G
LRP5 1275G>A
MCCC2 295G>C
MCCC2 518insT
MCCC2 687A>C
MKKS [250C>T + 724G>T]
MTHFR 1129C>T
MVK 803T>C
MVK 1174G>A
NPHS1 1481delC
NPHS1 3250delG
NPHS2 413G>A
NTRK1 IVS12+1G>A
PAH 280_282delATC
PAH 782G>A
PAH IVS10-11G>A
PAH IVS12+1GA
PAH 782G>A
PCCB 1606A>G
PEPD 793C>T
PKLR 1436G>A
PYGL IVS13+1G>A
RAG1 2974A>G
RMRP 70A>G
SERPINA1 1096G>A
SGCB 452C>G
SGCB 271C>T
SLC12A3 1924C>G
SLC12A3 8,627 bp deletion
SLC17A5 115C>T
SLC25A19 530G>C
SLC25A4 523delC
SLC3A1 IVS6+2T>C
SLC3A1 1354C>T
SLC6A3 [1408T>A + 1409A>G]
SLC6A3 IVS9+1G>T
SLC7A9 201C>T
SLC7A9 1166C>T
SMN1 exon 7 deletion
SPG20 1110delA
ST3GAL5 694C>T
STRADA 7 kb deletion
TERT 1710C>G
TH 698G>A
TJP2 143T>C
TMCO1 139_140delAG
TNFRSF1A 362G>A
TNNT1 505G>T
TOR1A GAG deletion
TSPYL1 457_458insG
TUBGCP6 5458T>G
UGT1A1 222C>A
ZMPSTE24 54_55insT

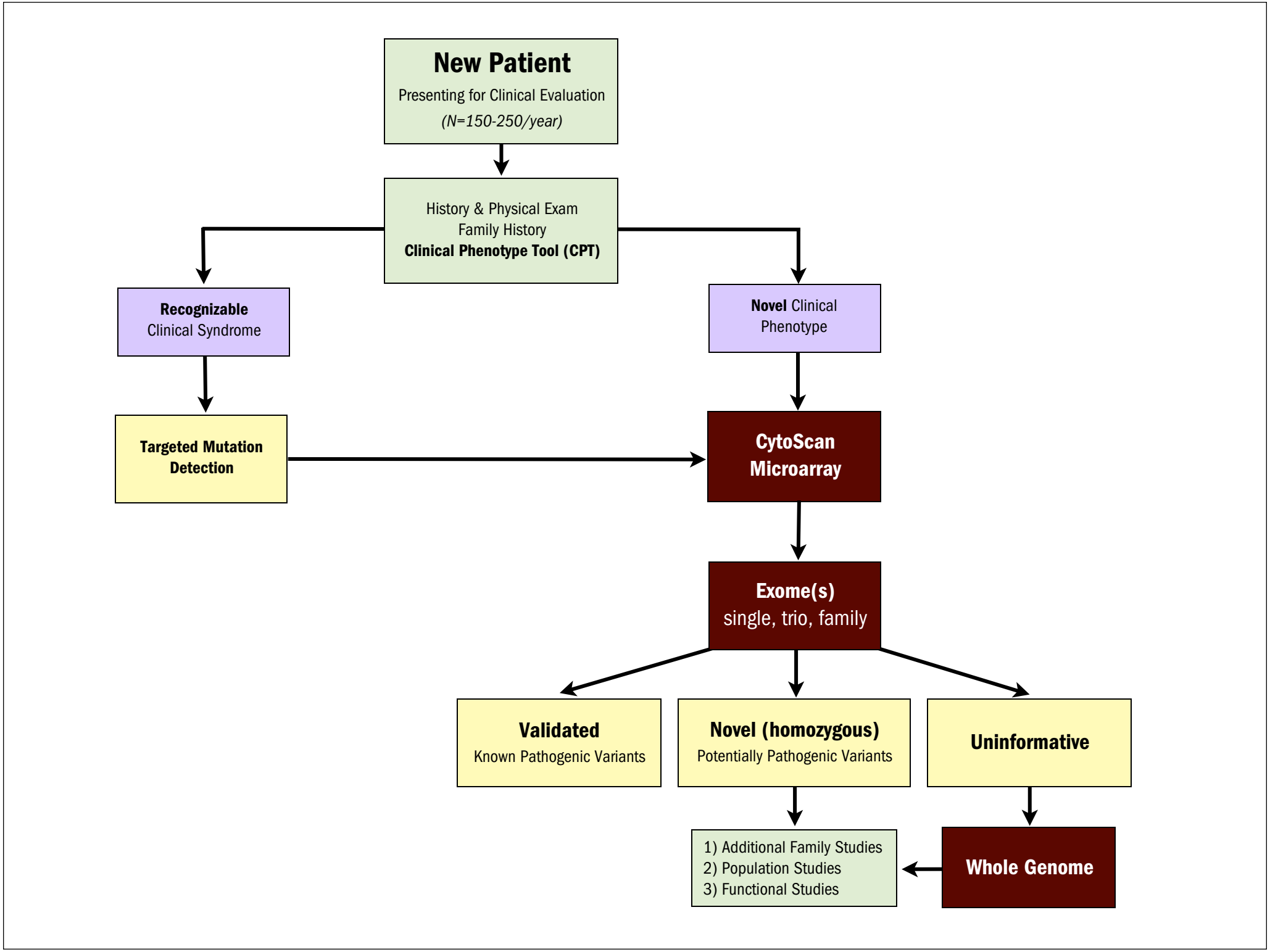
LightScanner 32

LightScanner 32

***TJP2* c.143T>C LunaProbe assay**




Institutional Memory


New Patient
Presenting for Clinical Evaluation
(N=150-250/year)
History & Physical Exam
Family History
Clinical Phenotype Tool (CPT)
Recognizable
Clinical Syndrome
Novel Clinical
Phenotype
Targeted Mutation
Detection
CytoScan
Microarray
Exome(s)
single, trio, family
Validated
Known Pathogenic Variants
Novel (homozygous)
Potentially Pathogenic Variants
Uninformative
1) Additional Family Studies
2) Population Studies
3) Functional Studies
Whole Genome

Why Does This Work?

- Commitment to provide accessible and affordable care
- Close interaction between physician and researcher
- Integration of basic science with patient care
- Continuity of care
- Patient-centered research
- Flexible laboratory
- Strong community support
- External collaborations