Provincial Health Services Authority

Constitutional genetic testing in BC: updates to ordering molecular genetic and cytogenetic tests at the Division of Genome Diagnostics.

Dr. Lindsay Brown, PhD, FCCMG Clinical Molecular Geneticist and Cytogeneticist
Dr. Elizabeth Digby, MD, FRCPC, FCCMG Medical Geneticist
Dr. Emma Strong, PhD, FCCMG Clinical Molecular Geneticist

Disclosures

Provincial Health Services Authority

- The authors declare no conflicts of interest.

We are grateful to speak to you today from the traditional, ancestral and unceded territory of the Musqueam, Squamish, and Tsleil-Waututh First Nations.

First Nations have been responsible for stewarding this land for all time and we give thanks as uninvited guests on these lands.

Provincial Health Services Authority

Learning objectives

Provincial Health Services Authority

- Recognize when specific genetic tests may or may not be indicated for your patients;
- Identify and utilize resources that support ordering constitutional genetic tests;
- Support multidisciplinary partnership between Pediatrics and Medical Genetics by identifying clinical scenarios that may require additional testing prior to referral to Medical Genetics.

Outline

Provincial Health Services Authority

1) Brief introduction to Division of Genome Diagnostics and the Provincial Medical Genetics Program;

2) Review common clinical scenarios, genetic tests to consider and requirements or considerations prior to referral to medical genetics;

3) Review how to order genetic testing and improvements made to this process;

4) Highlight resources available to support ordering of genetic tests.

Introductions

Provincial Health Services Authority

Division of Genome Diagnostics, Provincial Laboratory

- Constitutional molecular genetic and pediatric cytogenetic testing across lifetime
- Provide consultative support to Physicians;
- Test development and implementation

Provincial Medical Genetics Program

- Evaluation and diagnosis of congenital anomalies and genetic disease across lifetime
- Genetic counselling regarding chance of occurrence or recurrence of genetic disease
- Inpatient, outpatient, outreach consultations
- Education about medical genetics for health care professionals and others

Division of Genome Diagnostics

Provincial Health Services Authority

Molecular Genetic tests:

- Fragile X syndrome;
- Prader-Willi syndrome;
- Spinal Muscular Atrophy (SMA);
- Type 1 Myotonic Dystrophy

Cytogenetic tests:

- Chromosomal microarray (CMA);
- Karyotype;
- Targeted fluorescent *in-situ* hybridization (FISH)

Provincial Health Services Authority

Case example 1

Case example 1

Provincial Health Services Authority

- 3 year old male with global developmental delay.

Genetic tests to consider:

- Chromosomal microarray (CMA)
- Fragile X syndrome

Indications for CMA

Provincial Health Services Authority

Currently the clinical utility of CMA has been recognized by many professional societies and recommended as a ***first-tier genetic test*** for patients with ***unexplained GDD/ID, ASD, and/or multiple congenital anomalies***

Indications for Fragile X syndrome testing

Provincial Health Services Authority

- Males or females with global developmental delay (GDD) or intellectual disability (ID) of unknown etiology, with or without Autism Spectrum Disorder;
- *For individuals with developmental delay, ID or GDD must be suspected (multiple domains impacted)

Utility of Fragile X syndrome testing

Provincial Health Services Authority

- In individuals with ID, the diagnostic rate has been reported as **0-2.5%** in recent literature*
- The diagnostic yield increases to **9.5-17%** with increased specificity of testing (PMIDs: 19786505, 29624914, 8859272):
  - males with intellectual disability AND
  - characteristic physical and behavioural features OR
  - family history suggestive of Fragile X syndrome

* PMIDs: 32152462, 28541279, 28914265, 28933791, 28933790. See also Fragile X FAQ at www.genebc.ca

Common clinical features of Fragile X syndrome

Provincial Health Services Authority

Clinical feature	Frequency in Fragile X syndrome	Frequency in ID (without FXS)	Clinical Score
Skin soft and velvety on the palms with redundancy on the dorsum of the hands	88%	52%	2
Flat feet	70%	37%	2
Large and prominent ears	84%	22%	2
Plantar crease	86%	23%	1
Large testicles (post-puberty)	71%	10%	1
Family history of ID	81%	24%	1
Autistic-like behaviour	76%	25%	1

- Adapted from Table 2 in Lubala *et al* 2018
  PMID: 29624914
- Meta-analysis of clinical features of Fragile X syndrome
- ID was a prerequisite for inclusion in this study
- A score ≥ 5 had a significant yield of Fragile X diagnoses

Case example 1

Provincial Health Services Authority

- 3 year old male with developmental delay spanning multiple domains.
- Meets classification of global developmental delay.

Prior to referral to Medical Genetics

- Arrange (and provide and results of) available in province genetic testing
- Provide copies of any developmental assessments/psychoeducational testing
- Consultation note documenting any congenital anomalies, distinctive features, and/or pertinent family history
  - Relevant imaging studies and consultations, if applicable

Provincial Health Services Authority

Case example 2

Case example 2

Provincial Health Services Authority

- 7 year old male with mild autism spectrum disorder.
- No history of GDD, learning difficulties in school (reading) but no intellectual disability.

Genetic tests to consider:

- Chromosomal microarray

Clinical Utility of CMA

Provincial Health Services Authority

- Diagnostic yields associated with phenotypes (PMID: 31781653)
  - Isolated GDD/ID – 18%
  - GDD/ID with epilepsy – 17%
  - GDD/ID with multiple congenital anomalies – 35%
  - Isolated ASD – 4%
- Isolated ASD (~4-5%) had lower diagnostic yield than those patients with ASD and syndromic features (~25%) (PMID: 27975050, 29564645, 33394245)
- Higher diagnostic yield has been reported for moderate to severe ID (20-30%) than for mild ID (12-19%) (PMID: 26511719, 24297458)

Severe and complex phenotypes have a higher likelihood of identifying a genetic etiology

Utility of Fragile X syndrome testing

Provincial Health Services Authority

- Evidence-based publications do not support testing for Fragile X syndrome in individuals with isolated ASD
  - The diagnostic rate has been reported as **0%** in recent literature (PMID: 28541279)

Case example 2

Provincial Health Services Authority

- 7 year old male with mild autism spectrum disorder.
- No history of GDD, learning difficulties in school (reading) but no intellectual disability.

Prior to referral to Medical Genetics

- Referral largely not indicated
- Evaluate for additional features suspicious for underlying syndromic diagnosis
  - ID, congenital anomalies, distinctive features, very severe autism diagnosed at a very young age (particularly affecting girls), multiple severely affected siblings, concomitant ID cannot be ruled out and is highly suspected
- Consider providing recurrence chance counselling based on empiric data*

* PMIDs: 28973142

Provincial Health Services Authority

Case example 3

Case example 3

Provincial Health Services Authority

- 8 year old male with isolated ADHD.
- No history of GDD, intellectual disability or congenital anomalies.

Genetic testing not indicated.

Isolated behavioural disorders

Provincial Health Services Authority

- No studies evaluating diagnostic yield of CMA for isolated speech or motor delay or learning disability
- Lack of demonstrated utility for isolated behavioural disorders.
  - One study reported detection rate of ~8-9% in patients with ADHD (PMID: 31602316)
    - Lack of clinical information to determine if ADHD isolated or associated with GDD/ID
    - Many CNVs were inherited - clinical significance unclear and do not distinguish pathogenic vs VUS
  - Studies have shown that patients with ADHD had similar frequency of rare CNVs to that of controls (PMID: 19546859, 21832240).

Case example 3

Provincial Health Services Authority

- 8 year old male with isolated ADHD.
- No history of GDD, intellectual disability, or congenital anomalies.

Referral to Medical Genetics not indicated

Provincial Health Services Authority

Case example 4

Case example 4

Provincial Health Services Authority

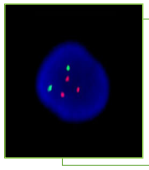
- 7 year old female with ID, CMA shows copy number variant of uncertain significance (VUS)

Genetic testing to arrange

- Parental follow-up variant assessment, as directed by test report
- May need samples from both parent and proband

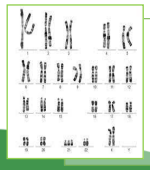
Parental follow up variant assessment

Provincial Health Services Authority



FISH
- Requires NaHep sample
- Sample from parents and proband

CMA
- Do not require proband sample
- EDTA sample from parents needed



Karyotype
- Requires NaHep sample
- Sample from parents and proband

CNV Classification

Provincial Health Services Authority

Recommended variant classification categories
(ACMG technical standards; PMID: 31690835)

- Pathogenic
- Likely pathogenic
- Uncertain significance (VUS)
- Likely benign*
- Benign*

*not included on clinical report

Interpretation and reporting of CNVs

Provincial Health Services Authority

Variants of uncertain significance (VUS)

- CNV described in a small number of cases in general population
- Contains genes that are unknown if dosage sensitive
- CNV described in multiple contradictory publicns and/or databases

Reporting of VUS can be challenging – goal of clinical interpretation and reporting is maximize return of useful diagnostic findings. Also to eliminate uncertainty and burden of further testing for families (avoid misinterpretation).

Parental follow up variant assessment

Provincial Health Services Authority

914 CNVs

~40% Path (367)		~60% VUS (547)	
128 parental follow up testing		198 with parental follow up testing	
~55% De novo (70)	~45% Inherited (58)	~14% de novo (28)	~86% inherited (170)

Interpretation and reporting of CNVs

Provincial Health Services Authority

Variants of uncertain significance (VUS)

- >80% are inherited (likely from healthy parent) and follow up testing does not change variant classification (i.e. does not change classification to likely pathogenic)
  - Questions the utility of this testing
- Finding of a VUS (or even a 'negative' CMA) does not exclude a genetic diagnosis
  - Many VUS are unlikely to be disease causing
  - Additional testing may be warranted to identify genetic etiology for patients clinical features
    - Referral to PMGP

Slide 30

NT[3 edit?
Nelson, Tanya [CWBC], 5/10/2022

Case example 4

Provincial Health Services Authority

- 7 year old female with ID, CMA shows copy number variant of uncertain significance

Prior to referral to Medical Genetics

- Please arrange parentals and wait for them to be reported **prior** to initiating referral as this is necessary for triage
- Provide copies of any developmental assessments/psychoeducational testing
- Consultation note documenting any congenital anomalies, dysmorphic features, and/or pertinent family history
  - Relevant imaging studies and consultations, if applicable
- ***Exception*** you do not need to arrange parentals for pathogenic, likely pathogenic, or neurosusceptibilty copy number variants
  - These parents may benefit from consenting discussion prior to appointment and these referral are prioritized

Provincial Health Services Authority

Case examples 5 & 6

Case example 5

Provincial Health Services Authority

- Newborn infant with hypotonia

Genetic testing to consider prior to referral to Medical Genetics

- CMA
- Prader-Willi Methylation Studies
- Spinal Muscular Atrophy Exon 7-8 Deletion Testing
- Myotonic Dystrophy Type 1 Repeat Expansion Testing
- Pompe enzyme assay

Case example 6

Provincial Health Services Authority

- 16M with tall stature and hypermobility ?Marfan syndrome

Prior to referral to Medical Genetics

- Calculate systemic score: https://marfan.org/dx/
- Arrange and provide echocardiogram
- Arrange and provide ophthalmology assessment
- Provide pertinent family history

No genetic testing indicated prior to referral

Scoring of systemic features.

Feature	YES	NO
Wrist AND thumb sign	3	0
Wrist OR thumb sign	1	0
Pectus carinatum deformity	2	0
pectus excavatum or chest asymmetry	1	0
Hindfoot deformity	2	0
Plain flat foot	1	0
Pneumothorax	2	0
Dural ectasia	2	0
Protrusio acetabulae	2	0
Reduced US/LS and increased armspan/height	1	0
Scoliosis or thoracolumbar kyphosis	1	0
Reduced elbow extension	1	0
3/5 facial features	1	0
Skin striae	1	0
Myopia	1	0
Mitral valve prolapse	1	0

How do I order genetic testing?

Provincial Health Services Authority

- All test requests require the appropriate test requisition form and clinical indications for testing must be provided

Quality improvement updates:

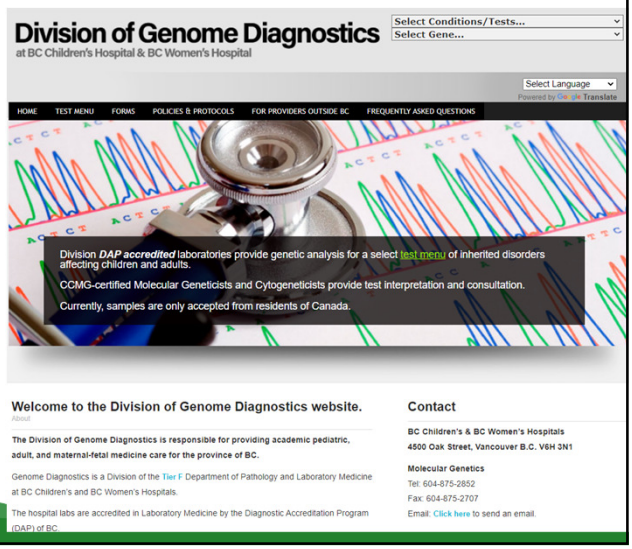
- 1 requisition: constitutional genetic test requisition
- Many clinical indications provided on 2nd page for ease of completion
- Only 2 mL EDTA necessary (unless NaHep also required)

Where can I find more information?

Provincial Health Services Authority

- www.genebc.ca

- Test requisition form
- Tests currently available
- Test information, including indications
- FAQs for common scenarios
- Information on how to apply for out-of-province testing
- Contact information for further enquiries



Where can I find more information?

Provincial Health Services Authority

www.genebc.ca

If you would like a word version of our requisition to build into your EMR, please contact us at:

moleculargenetics@cw.bc.ca