PEDIATRIC GLIOMAS—A CASE-BASED, PRACTICAL APPROACH TO USING THE WHO 2021 CLASSIFICATION

CYNTHIA HAWKINS, MD, PHD, FRCPC

DIVISION OF PATHOLOGY, LABATT BRAIN TUMOUR RESEARCH CENTRE, THE HOSPITAL FOR SICK CHILDREN, UNIVERSITY OF TORONTO, CANADA
DISCLOSURE

- I have nothing to declare
OBJECTIVES - AT THE END OF THIS TALK YOU SHOULD BE ABLE TO:

1. Approach the workup of a pediatric glioma
2. Apply the changes in the new WHO classification of CNS tumors as it applies to gliomas
3. Integrate molecular and morphologic data to generate a layered neuropathologic diagnosis
WHO 2016

- Major goal of 2016 WHO was formulating concept of how CNS tumour diagnoses could be structured in the molecular era
- Integrated Diagnosis – incorporating molecular and morphologic data
- Incorporation of molecularly defined entities
Major restructuring of diffuse gliomas:
- Incorporation of distinct pediatric-type vs adult-type entities
- Additional molecularly-defined entities

Restructuring of ependymomas to recognize distinct location-based and molecular entities

Additional molecularly-defined embryonal tumors

Move away from assigned grades based on entity name to grading within an entity
CASE 1

- 3-month-old girl presented with diencephalic syndrome and nystagmus
CASE 1

- Rapid clinical deterioration despite chemotherapy (Vinblastine)
- Biopsy
CASE 1 - BIOPSY
CASE 1 - BIOPSY
CASE 1 - BIOPSY
CASE 1 - BIOPSY
Most common CNS neoplasm in children

Distinct from adult “lower grade glioma”

Histologically diverse group of tumors arising throughout CNS

Now categorised under one of Paediatric-type diffuse low-grade gliomas, Circumscribed astrocytic gliomas or Glioneuronal and neuronal tumours
CLASSIFICATION OF GLIOMAS

- **Adult-type diffuse gliomas**
  - Astrocytoma, IDH-mutant
  - Oligodendroglioma, IDH-mutant and 1p/19q-codeleted
  - Glioblastoma, IDH-wildtype

- **Paediatric-type diffuse high-grade gliomas**
  - Diffuse midline glioma, H3 K27-altered
  - Diffuse hemispheric glioma, H3 G34-mutant
  - Diffuse paediatric-type high-grade glioma, H3-wildtype and IDH-wildtype
  - Infant-type hemispheric glioma

- **Paediatric-type diffuse low-grade gliomas**
  - Diffuse astrocytoma, MYB- or MYBL1-altered
  - Angiocentric glioma
  - Polymorphous low-grade neuroepithelial tumour of the young
  - Diffuse low-grade glioma, MAPK-altered

- **Circumscribed astrocytic gliomas**
  - Pilocytic astrocytoma
  - High-grade astrocytoma with piloid features
  - Pleomorphic xanthoastrocytoma
  - Subependymal giant cell astrocytoma
  - Chordoid glioma
  - Astroblastoma

- **Glioneuronal and neuronal tumours**
  - Ganglioglioma
  - DIG/DIA
  - DGONC
  - DNT
  - Papillary glioneuronal tumour
  - RGNT
  - MGNT
  - DLGNT
  - Central neurocytoma
  - Extraventricular neurocytoma
Molecular vs Histology
Do I really need molecular?
What does the molecular mean?
# The Molecular Landscape of PLGG

<table>
<thead>
<tr>
<th>Alteration</th>
<th>Common pLGG Drivers</th>
<th>Other MAPK Drivers</th>
<th>FGF Receptors</th>
<th>Receptor Tyrosine Kinases</th>
<th>Non-MAPK Oncogenes</th>
<th>Secondary</th>
</tr>
</thead>
<tbody>
<tr>
<td>BRAF p.V600E</td>
<td>NF1</td>
<td>Other BRAF SNV</td>
<td>FGF1 SNV</td>
<td>ROS1 Fusion</td>
<td>IDH1 p.R132H</td>
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<tr>
<td>NF1</td>
<td>KRA5 SNV</td>
<td>MAP2K1 Indel</td>
<td>FGF1-TACC1</td>
<td>NTRK Fusion</td>
<td>GNA11 SNV</td>
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<td>FGF2 Fusion</td>
<td>PDGFRA SNV</td>
<td>PIK3CA SNV</td>
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<td>PDGFRB Fusion</td>
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<td>KIT SNV</td>
<td>MYC</td>
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<td>MET SNV</td>
<td>MYB</td>
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<td>RET SNV</td>
<td>MYB1</td>
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<td>Undetermined</td>
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</tbody>
</table>

- KIAA1549-BRAF: 35%
- BRAF p.V600E: 17%
- NF1: 17%
- Other MAPK Drivers: 2.7%
- FGF Receptors: 6.1%
- Non-MAPK Oncogenes: 4.6%
- Other Receptor Tyrosine Kinases: 3.4%
- Undetermined: 16%
## Molecular vs Histology: Low Grade Gliomas with Enriched Molecular Alterations

<table>
<thead>
<tr>
<th>Tumour</th>
<th>Characteristic Gene</th>
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<tbody>
<tr>
<td>Papillary glioneuronal tumour</td>
<td>PRKCA</td>
</tr>
<tr>
<td>Rosette-forming glioneuronal tumour</td>
<td>FGFR1 &amp; PIK3CA</td>
</tr>
<tr>
<td>Myxoid glioneuronal tumour</td>
<td>PDFGRA</td>
</tr>
<tr>
<td>Diffuse leptomeningeal glioneuronal tumour</td>
<td>KIAA1549-BRAF fusion, 1p del</td>
</tr>
<tr>
<td>Astroblastoma, MN1-altered</td>
<td>MN1 fusion</td>
</tr>
<tr>
<td>Pleomorphic xanthoastrocytoma</td>
<td>BRAF &amp; CDKN2A/B hom del</td>
</tr>
<tr>
<td>Pilocytic astrocytoma</td>
<td>KIAA1549-BRAF, BRAF, NF1</td>
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<tr>
<td>Angiocentric glioma</td>
<td>MYB fusion</td>
</tr>
<tr>
<td>Diffuse astrocytoma, MYB- or MYBL1-altered</td>
<td>MYB, MYBL1 fusion</td>
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<tr>
<td>PLNTY</td>
<td>FGFR2 fusion</td>
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</table>
MUTATION VS HISTOLOGY IN PAEDIATRIC LOW GRADE GLIOMA – MIX AND MATCH

Ryall et al. Acta Neuropath Comms, 2020
DO I REALLY NEED MOLECULAR TESTING FOR PLGG?

- For morphologically classic entities with gross total resection, histology alone may be sufficient
- Situations where molecular characterisation is helpful:
  - Consideration is being given to radiation and need further prognostic guidance
  - Small biopsy and unsure if low grade vs high grade
  - Growing, incompletely resected lesion with potential for targeted therapeutics
SURVIVAL VARIATES BY MOLECULAR STATUS
RESPONSE TO CHEMOTHERAPY VARIES BY MOLECULAR STATUS

Fisher Exact test p=0.0226
RELAPSED KIAA1549-BRAF PATIENTS AND RESPONSE TO CHEMOTHERAPY VS MEKI

Fangusaro et al. Lancet Oncology, 2019
RESPONSE TO TARGETED THERAPEUTICS MAY BE BETTER THAN TO STANDARD CHEMOTHERAPY

6-month response to chemo  6-month response to BRAFi

Nobre et al. JCO Precision Oncology, 2020
LGG TESTING STRATEGY

- LGG (IDH wildtype)
  - BRAFV600E (IHC)
    - Fusion sensitive NGS*
    - CDKN2A deletion
      - If midline H3K27M (IHC)

Include:
- MYB and MYBL1 fusions
- FGFR2 fusions
- FGFR1 fusions, ITD, SNVs
- MN1 fusions
- PRKCA fusions
- BRAF fusions and SNVs
- PDGFRA SNVs
- PIK3CA SNVs
CASE 1 - BRAFV600E
Biopsy, Optic pathway/ hypothalamic mass:

- Ganglioglioma
  - WHO grade I
  - BRAF p.V600E mutant (IHC)
  - CDKN2A not deleted (FISH)
CASE 1

- BRAF inhibitor started when the patient was critically ill in ICU
- Prompt clinical and radiological response
- Improvement of diencephalic syndrome (calorimetry normal), normal vision
- Currently on therapy 6 years later doing well
CASE 2

12 yr old female presented with

- Headaches and vomiting x 4 week
- Diploplia and blurry vision x 2 weeks

- Examination
  - Bilateral papilledema
  - No other neurological deficit
CASE 2 - BIOPSY
CASE 2- IHC
PEDIATRIC-TYPE DIFFUSE HIGH-GRADE GLIOMAS

- Less common than low-grade glioma
- Usually are not the result of progression from low-grade counterpart (except BRAFV600E)
- Molecularly distinct from adult-type, by definition IDH WT
- Types defined based on characteristic age, location and molecular alterations
DIFFUSE HIGH-GRADE GLIOMAS: TUMOR TYPES

Adult-type
- Astrocytoma, IDH-mutant
- Oligodendroglioma, IDH-mutant and 1p/19q co-deleted
- Glioblastoma, IDH-wildtype

Pediatric-type
- Diffuse midline glioma, H3 K27-mutant
- Diffuse midline glioma, H3 G34-mutant
- Diffuse hemispheric glioma, H3 G34-mutant
- Diffuse high-grade paediatric-type glioma, H3-wildtype
- Infantile-type hemispheric glioma, H3-wildtype

Anaplastic glioma or GBM
- Diffuse midline glioma, H3 K27-mutant
- Diffuse midline glioma, H3 G34-mutant
- Diffuse hemispheric glioma, H3 wildtype with EZHIP over-expression
Histone mutations are most frequent recurrent alterations
PEDIATRIC-TYPE DIFFUSE HIGH-GRADE GLIOMAS: DIAGNOSTIC APPROACH

**HISTOLOGY**

- Anaplastic glioma or GBM

**LOCATION**

- Midline
- Hemispheric

**TESTING**

- P53, ATRX, BRAFV600E
- H3K27M
- H3K27me3
- EGFR mutation
- EZHIP

**INTEGRATED DIAGNOSIS**

- Diffuse midline glioma, H3 K27-mutant
- Diffuse midline glioma, EGFR-mutant
- Diffuse midline glioma, H3-wildtype with EZHIP over-expression
- Diffuse high-grade paediatric-type glioma, H3-wildtype
- Diffuse hemispheric glioma, H3 G34-mutant
- Infantile-type hemispheric glioma, H3-wildtype

**Fusion testing:** NTRK, ROS, MET, ALK
CASE 2- IHC

H3K27me3

H3K27M
CASE 2 - IHC

ATRX

P53
Biopsy, Thalamic mass:
  - Diffuse midline glioma, H3 K27-altered
    - WHO grade 4
    - H3.3 p.K27M (IHC)
### Molecular Results

<table>
<thead>
<tr>
<th>Protein Length</th>
<th>Custom Mutation</th>
<th>COSMIC</th>
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<tbody>
<tr>
<td>K28M</td>
<td>1 mutation</td>
<td>309</td>
</tr>
<tr>
<td>50</td>
<td>100</td>
<td>150</td>
</tr>
</tbody>
</table>
Biopsy, Thalamic mass:
- Diffuse midline glioma, H3 K27-altered
  - WHO grade 4
  - H3.3 p.K27M (IHC and NGS)
  - P53 p.H179del (NGS)
DIFFUSE MIDLINE GLIOMA, H3 K27M-MUTANT

- Enriched in childhood but can occur at any age, particularly outside the pons
- H3.3 and H3.1
- Very poor outcome

Solomon et al. Brain Path 2015

Mackay et al. Cancer Cell 2017
DIFFUSE MIDLINE GLIOMA, H3 K27M-MUTANT

- cIMPACT/WHO recommendation is to limit this diagnosis to diffuse gliomas
- Even if DA or circumscribed/ grade I histology, these are not low-grade gliomas
H3K27M INHIBITS PRC2 FUNCTION AND LEADS TO LOSS OF H3K27ME3
Some DMGs have loss of H3K27me3 but no H3K27M

EZHIP inhibits PRC2 through an H3K27M-like mechanism

Molecular heterogeneity and CXorf67 alterations in posterior fossa group A (PFA) ependymomas  
Acta Neuropathol 136(2):211-26, 2018

Kristian W. Pailler,1,2,3,9 Ji Wan,4,20 Martin Sill,1,2,20 Tong Lin,5,20 Wida Oriejon,4 Bo Tang,4 Jenia-Martin Hübner,1,2 Vilay Ramaswamy,6,7 Suljana Jib,1 James D. Dalton,4 Kelly Hoogendam,6 Hazel A. Rogers,9 Chandanamali Panchilshwar,4 Ryan Lee,4 John Easton,9 Gane Wu,9 Timothy A. Ritzmann,8 Rebecca Chanman,3 Lukas Chavez,1,2 Fredrick A. Boop,10 Paul Klimo Jr.,16 Noah D. Sabin,11 Robert Ogg,11 Stephen C. Mack,7,12 Brian D. Freibaur,13 Hong Joo Kim,13 Harshik Witt,1,2,3 David T.W. Jones,1,2 Baichuan Yu,14 Amar Gujjar,15 Blan Pounds,2 Azru Omar-Thomas,5 Martina F. Roussel,14 Jinghui Zhang,9 J. Paul Taylor,13,16 Thomas E. Merchant,17 Richard Grundy,5 Ruth G. Tatevosian,4 Michael D. Taylor,7 Stefan M. Pfister,1,2,3 Andrey Korsunov,18,19 Marcel Kool,1,2,21 and David W. Ellison4,21,7

Histone H3 wild-type DIPG/DMG overexpressing EZHIP extend the spectrum diffuse midline gliomas with PRC2 inhibition beyond H3-K27M mutation  
Acta Neuropathol 139: 1109–1113, 2020

David Castel1,2· Thomas Kergrohen1,2· Arnauld Tauziède-Espariat3,4· Alan Mackay5· Samia Ghermaoui1· Emmanuèle Lechapt3,4· Stefan M. Pfister6,7,8· Christof M. Kramm9· Nathalie Boddaert10· Thomas Blauwblomme11· Stéphanie Puget11· Kévin Beccaria1,11· Chris Jones5· David T. W. Jones6,12· Pascale Varlet3,4· Jacques Grill1,2· Marie-Anne Debily1,13
H3 WILD-TYPE DMG WITH EZHIP OVER-EXPRESSSION

- Outcome and age distribution similar to H3K27M
- These likely belong in DMG, H3 K27M mutant group
Pediatric bithalamic gliomas have a distinct epigenetic signature and frequent EGFR exon 20 insertions resulting in potential sensitivity to targeted kinase inhibition.
Category expanded to recognise all diffuse gliomas with H3K27me3 loss

- DMG, H3.3 K27-mutant
- DMG, H3.1/3.2 K27-mutant
- DMG, EGFR-mutant
- DMG, H3-wildtype with EZHIP overexpression
CASE 3

- 7 month-old boy presented with poor feeding and increased head circumference
- Partial resection
CASE 3
CASE 3 IHC
DIFFUSE HIGH-GRADE GLIOMAS: TUMOR TYPES

Anaplastic glioma or GBM

Adult-type

- Astrocytoma, IDH-mutant
- Oligodendroglioma, IDH-mutant and 1p/19q co-deleted
- Glioblastoma, IDH-wildtype

Pediatric-type

Midline

- Diffuse midline glioma, H3 K27-mutant
- Diffuse midline glioma, H3 G34-mutant
- Diffuse midline glioma, H3-wildtype with EZHIP overexpression

Hemispheric

- Diffuse hemispheric glioma, H3-wildtype
- Diffuse high-grade paediatric-type glioma, H3-wildtype
- Infantile-type hemispheric glioma, H3-wildtype

AYA

child

infant
3 MAIN SUBGROUPS OF INFANT GLIOMAS

Stucklin, Ryall et al. Nature Commun, 2019
INFANT-TYPE HEMISPHERIC GLIOMA

- Hemispheric, high-grade gliomas arising in early childhood, mostly < 1 year
- Typically harbor receptor tyrosine kinase fusions: NTRK, ALK, ROS1, MET
- Better outcome than high grade gliomas in older children
- RTK fusions may be therapeutically targeted
RNA sequencing revealed \textit{CLIP2-MET} fusion
Resection, Left hemispheric mass:
- Infantile hemispheric glioma
  - *CLIP2-MET* fusion positive (RNAseq)
CASE 3

- Treated with chemotherapy (carbo/vincristine)
- Currently off-treatment, doing well
OBJECTIVES - AT THE END OF THIS TALK YOU SHOULD BE ABLE TO:

1. Approach the workup of a pediatric glioma

2. Apply the changes in the new WHO classification of CNS tumors as it applies to gliomas

3. Integrate molecular and morphologic data to generate a layered neuropathologic diagnosis
QUESTIONS?
References

1. Ellison et al., Acta Neuropathologica 137:683-687, 2019
4. Louis et al. Neuro Oncol 23(8):1231-51, 2021
5. Louis et al. Brain Pathology 29(4):469-72, 2019
7. Guerreiro-Stucklin et al. Nature Communications. 10(4343), 2019