GLIOMAS

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4th ESO-ESMO-RCE “Clinical update on rare adult solid cancers”
Milan, November 29, 2019
I have received grants and honoraria for Lectures and Advisory Boards from MSD, Roche, Merck Serono, Celldex Therapeutics, Novartis, Puma, Abbvie and Mundipharma.
OUTLINE

• New histological and molecular WHO classification of 2016.

• Reevaluation of clinical trial results across the new molecular subtypes.

• New tools for improving response evaluation.

• Update on glioblastomas.

• New insights.
# WHO classification of tumours of the central nervous system

<table>
<thead>
<tr>
<th>Diffuse astrocytic and oligodendroglial tumours</th>
</tr>
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<tbody>
<tr>
<td>Diffuse astrocytoma, IDH-mutant</td>
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<tr>
<td>Gemistocytic astrocytoma, IDH-mutant</td>
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<tr>
<td><strong>Diffuse astrocytoma, IDH-wildtype</strong></td>
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<tr>
<td>Diffuse astrocytoma, NOS</td>
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<tr>
<td>Anaplastic astrocytoma, IDH-mutant</td>
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<tr>
<td><strong>Anaplastic astrocytoma, IDH-wildtype</strong></td>
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<tr>
<td>Anaplastic astrocytoma, NOS</td>
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<tr>
<td>Glioblastoma, IDH-wildtype</td>
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<tr>
<td>Giant cell glioblastoma</td>
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<tr>
<td>Gliosarcoma</td>
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<tr>
<td><strong>Epithelioid glioblastoma</strong></td>
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<tr>
<td>Glioblastoma, IDH-mutant</td>
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<tr>
<td>Glioblastoma, NOS</td>
</tr>
<tr>
<td>Diffuse midline glioma, H3 K27M-mutant</td>
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<tr>
<td>Oligodendroglioma, IDH-mutant and 1p/19q-codeleted</td>
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<tr>
<td>Oligodendroglioma, NOS</td>
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<tr>
<td>Anaplastic oligodendroglioma, IDH mutant and 1p/19q-codeleted</td>
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<tr>
<td><strong>Anaplastic oligodendroglioma, NOS</strong></td>
</tr>
<tr>
<td>Oligoastrocytoma, NOS</td>
</tr>
<tr>
<td>Anaplastic oligoastrocytoma, NOS</td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>Other astrocytic tumours</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pilocytic astrocytoma</td>
</tr>
<tr>
<td>Pilomyxoid astrocytoma</td>
</tr>
<tr>
<td>Subependymal giant cell astrocytoma</td>
</tr>
<tr>
<td>Pleomorphic xanthoastrocytoma</td>
</tr>
<tr>
<td>Anaplastic pleomorphic xanthoastrocytoma</td>
</tr>
</tbody>
</table>
Figure 6. Clinical Outcomes.

Panel A shows Kaplan–Meier estimates of overall survival among patients with LGGs that are classified according to traditional histologic type and grade. GBM samples (from previously published Cancer Genome Atlas data) are also included for comparison. Panel B shows overall survival among patients with LGGs that are classified according to IDH mutation and 1p/19q codeletion status. GBM samples classified according to IDH mutation status are also included. The results of an age-adjusted analysis are provided in Table S2 in Supplementary Appendix 1, and further division according to histologic type, grade, and molecular subtype is shown in Fig. S22 in Supplementary Appendix 1.
### OVERALL SURVIVAL IN MOLECULARLY DEFINED ANAPLASTIC GLIOMAS IN LARGE PHASE III TRIALS

<table>
<thead>
<tr>
<th>STUDY</th>
<th>HISTOLOGY</th>
<th>MOLECULAR SUBTYPE</th>
<th>TREATMENT</th>
<th>N</th>
<th>MEDIAN OS</th>
<th>MEDIAN PFS</th>
</tr>
</thead>
<tbody>
<tr>
<td>EORTC 26951</td>
<td>Anaplastic oligodendroglioma</td>
<td>1p/19q codeleted IDHmt 1p/19q intact 7+/10q-/TERTpmt</td>
<td>RT/PCV</td>
<td>43</td>
<td>NR (&gt;14 yrs)</td>
<td>147</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RT/PCV</td>
<td>23</td>
<td>8.3 yrs</td>
<td>4.2 yrs</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RT or RT/PCV</td>
<td>55</td>
<td>1.13 yrs</td>
<td>NS</td>
</tr>
<tr>
<td>RTOG 9402</td>
<td>Anaplastic oligodendroglioma</td>
<td>1p/19q IDHmt (all)</td>
<td>RT/PCV</td>
<td>59</td>
<td>14.7 yrs</td>
<td>8.4 yrs</td>
</tr>
<tr>
<td>RTOG 9804</td>
<td>Anaplastic astrocytoma</td>
<td>IDH mt (IHC) IDHwt</td>
<td>RT/chemo</td>
<td>49</td>
<td>7.9 yrs</td>
<td>2.8 yrs</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>54</td>
<td>2.8 yrs</td>
<td></td>
</tr>
<tr>
<td>NOA4</td>
<td>Grade III</td>
<td>1p/19q codeleted IDHmt 1p/19q intact IDHwt</td>
<td>RT or chemo</td>
<td>66</td>
<td>NR</td>
<td>7.0-7.3 yrs</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>83</td>
<td>7.0-7.3 yrs</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>58</td>
<td>3.1 – 4.7 yrs</td>
<td></td>
</tr>
</tbody>
</table>

### ANAPLASTIC GLIOMA REPORTED MEDIAN SURVIVAL AFTER RT/CHEMO

<table>
<thead>
<tr>
<th>NUMERICAL REPORTED MEDIAN SURVIVAL AFTER RT/CHEMO</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oligodendroglioma, IDHmut &amp; 1p/19q codeleted</td>
</tr>
<tr>
<td>&gt; 14 years , 40% alive at 20 years (SNO 2019)</td>
</tr>
<tr>
<td>Astrocytoma, IDH mutated</td>
</tr>
<tr>
<td>7 - 8 years</td>
</tr>
<tr>
<td>Astrocytoma IDH wt</td>
</tr>
<tr>
<td>1 – 4.7 yrs</td>
</tr>
</tbody>
</table>
CATNON STUDY ON ANAPLASTIC GLIOMAS WITHOUT 1P/19Q LOSS: 2 X 2 DESIGN

- Pre-study 1p/19q testing
- Stratification: Methylation status
- Primary endpoint: overall survival

- Secondary endpoints:
  - Progression-free survival
  - Quality of life
  - Neurological deterioration free survival

- RT 59.4 GY + CONCURRENT TEMOZOLOMIDE
- NO ADJUVANT TREATMENT
- FOLLOW-UP

- RT 59.4 GY
- ADJUVANT TMZ
## OVERALL SURVIVAL FOLLOWING ADJUVANT TEMOZOLOMIDE:
### UNIVARIATE ANALYSIS (SECONDARY)

**Overall Survival**

<table>
<thead>
<tr>
<th>Adjuvant TMZ</th>
<th>Pts (N)</th>
<th>Observed events (O)</th>
<th>Non-parametric</th>
<th>Cox Model</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Median (95% CI) (Months)</td>
<td>Alive at 2 years (%) (95% CI)</td>
</tr>
<tr>
<td>No</td>
<td>372</td>
<td>129</td>
<td>41.1 (36.6, 60.7)</td>
<td>71.2 (65.3, 76.2)</td>
</tr>
<tr>
<td>Yes</td>
<td>373</td>
<td>92</td>
<td>Not Reached</td>
<td>75.6 (69.8, 80.4)</td>
</tr>
</tbody>
</table>

*Van den Bent et al, Lancet, 2017*
Phase III EORTC 22033-26033: radiotherapy vs temozolomide chemotherapy.

- PFS oligodendrogliomas IDH mutant and 1p/19q codeleted: 61.6 months for radiotherapy vs 55 months for temozolomide (ns).

- PFS astrocytomas IDH mutant and non 1p/19q codeleted: 55 months for radiotherapy vs 36 months for temozolomide (p=0.013).

- OS data still not mature

*Baumert et al, Lancet Oncol. 2016*
WHAT HAVE WE LEARNED FROM POST-HOC ANALYSES OF CLINICAL TRIALS ON THE IMPACT OF ADJUVANT TREATMENTS FOLLOWING SURGERY ACROSS MOLECULAR SUBTYPES OF HIGH RISK WHO GRADE II GLIOMAS?

Phase III RTOG 9402: radiotherapy vs radiotherapy + PCV

- PFS: 4 yrs with RT vs 10.4 yrs with RT+PCV
- OS: 7.8 yrs with RT vs 13.4 yrs vs RT+PCV
- Benefit of the addition of PCV to RT in both molecular subtypes but definitively longer in oligodendrogliomas IDH mutant and 1p/19q codeleted: OS 7.3 yrs with RT alone vs 14.7 with RT+PCV

WHAT HAVE WE LEARNED FROM POST-HOC ANALYSES OF CLINICAL TRIALS ON THE IMPACT OF ADJUVANT TREATMENTS FOLLOWING SURGERY ACROSS MOLECULAR SUBTYPES OF HIGH RISK WHO GRADE II GLIOMAS?

Phase II single arm UCSF trial with initial temozolomide alone:

• PFS longer in oligodendrogliomas 1p/19q codeleted and IDH mutant (4.9 yrs) as compared to astrocytomas 1p/19q non codeleted and IDH mutant (3.6 yrs); more than half of patients delaying RT for 5.8 years (Wahl et al, Neuro Oncol. 2017).

Italian AINO study with initial temozolomide alone:

• PFS longer in oligodendrogliomas 1p/19q codeleted and IDH mutant (4.2 yrs) as compared to astrocytomas 1p/19q non codeleted IDH mutant (3.6 yrs); 67% of patients with oligodendrogliomas did not recur with a median follow-up of 9.3 yrs and 59% did not receive RT with a median follow-up of 8.2 years (Rudà et al et al, J of Neurooncol. 2019).
QUESTIONS TO BE ANSWERED BY ONGOING RANDOMIZED CLINICAL TRIALS WITH MOLECULAR INCLUSION CRITERIA IN LOWER GRADE (GRADE II AND III) GLIOMAS

• Can temozolomide replace PCV in combination with radiotherapy?
  ↓

   Phase III RTOG/EORTC CODEL study in oligodendrogliomas 1p/19q codeleted and IDH-mutant: RT+PCV vs RT+TMZ.

• Can a shorter survival after initial chemotherapy alone be balanced by a better preservation of cognitive functions as compared to initial radiotherapy and chemotherapy?
  ↓

   Randomized POLO and POLCA trials in France in IDH-mutant and 1p/19q codeleted gliomas: PCV vs RT + PCV with overall survival with neurocognitive deterioration as primary endpoint.

• Which is better between observation with MRI or early RT and TMZ in astrocytomas IDH-mutant non 1p/19q codeleted?
  → phase III EORTC I-WOT study.
# AINO PHASE II STUDY ON INITIAL TEMOZOLOMIDE IN HIGH-RISK WHO GRADE II GLIOMAS

Primary Endpoint: response based on MRI RANO criteria

<table>
<thead>
<tr>
<th>Response</th>
<th>Count</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>CR</td>
<td>0/60</td>
<td></td>
</tr>
<tr>
<td>PR</td>
<td>23/60</td>
<td>38.3%</td>
</tr>
<tr>
<td>MR</td>
<td>12/60</td>
<td>20%</td>
</tr>
<tr>
<td>SD</td>
<td>21/60</td>
<td>35%</td>
</tr>
<tr>
<td>PD</td>
<td>4/60</td>
<td>6.7%</td>
</tr>
</tbody>
</table>

Median number of TMZ cycles: 11 (range 2-18)

Rudà et al, J Neurooncol. 2019
ASSOCIATION BETWEEN MRI RESPONSE AND MGMT METHYLATION

Rudà et al, J Neurooncol. 2019
# SEIZURE RESPONSE

## Seizure response

<table>
<thead>
<tr>
<th>Seizure response</th>
<th>Count</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Significant (&gt; 50% reduction of seizure frequency)</td>
<td>34/39</td>
<td>87.2%</td>
</tr>
<tr>
<td>Not significant</td>
<td>5/39</td>
<td>12.8%</td>
</tr>
<tr>
<td>Increase in seizure frequency</td>
<td>0/39</td>
<td>0%</td>
</tr>
</tbody>
</table>

- Seizure-free at 18 months 28/39 (71.8%)
- Significant reduction of AEDs 11/28 (39%)
- Withdrawal of AEDs 2/28 (7%)
- Significant association between seizure response and IDH 1-2 mutations

*Rudà et al, J Neurooncol. 2019*
Grade II IDH-wild type

With the current 2016 WHO Classification all IDH-wild type tumours are classified as astrocytomas, regardless of the tumour grade. Thus, cases originally diagnosed as either oligodendrogliomas or oligoastrocytomas according to WHO 2007 need to be reclassified as astrocytomas.

There is now increasing evidence of the molecular heterogeneity of the IDH-wild type grade II astrocytomas.

Due to the aggressive behaviour of these tumours an adjuvant treatment after surgery is always needed. Which is the best treatment options should be analysed in prospective multicenter trials based on molecular characterisation.

There is now increasing evidence of the molecular heterogeneity of the IDH-wild type grade II astrocytomas. Aibadula et al, Neuro Oncol 2017;19(10), 1327 - 1337.

**Diffuse IDH-wild type astrocytomas with molecular features of GBM:**
- EGFR amplification
- TERTp mutation
- H3F3A-K27 mutation
- (BRAF-wild type)
- (MYB amplification)
- +7/-10 chromosomes alterations

Diffuse astrocytic glioma, IDH-wildtype, with molecular features of glioblastoma, WHO grade IV

Aibadula et al., Neuro Oncol. 2017;19(10), 1327-1337.
HOW TO BEST ASSESS THE RESPONSE TO TREATMENTS ON NEUROIMAGING:

NEW TOOLS
Evaluation of low-grade glioma structural changes after chemotherapy using DTI-based histogram analysis and functional diffusion maps

Antonella Castellano¹, Marina Donatiivi², Roberta Rudà³, Giorgio De Nunzio²,⁴, Marco Riva⁵, Antonella Iadanza¹, Luca Bertero³, Matteo Rucco⁶, Lorenzo Bello⁵, Riccardo Soffietti³, Andrea Falini¹

Received: 11 January 2015 / Revised: 16 July 2015 / Accepted: 20 July 2015
© European Society of Radiology 2015
AXIAL FLAIR @ BASELINE

AXIAL FLAIR AFTER 6 CYCLES TMZ: SD (RANO BIDIMENSIONAL CRITERIA)
FDM ON DIFFUSIVITY MAPS AFTER 3 CYCLES OF TMZ: INITIAL REDUCTION OF ISOTROPY

FDM ON DIFFUSIVITY MAPS AFTER 6 CYCLES TMZ: FURTHER REDUCTION OF ISOTROPY
MR SPECTROSCOPY FOR 2-HYDROXYGLUTARATE

Amino acid positron emission tomography to monitor chemotherapy response and predict seizure control and progression-free survival in WHO grade II gliomas

Ulrich Roelcke†, Matthias T. Wyss†, Martha Nowosielski, Roberta Rudä, Patrick Roth, Silvia Hofer, Norbert Galldiks, Flavio Crippa, Michael Weller, and Riccardo Soffietti

Department of Neurology and Brain Tumor Center, Cantonal Hospital, Aarau, Switzerland (U.R.); Institute for Pharmacology and Toxicology, ETH and University of Zürich, Zürich, Switzerland (M.T.W.); Neuroscience Center, ETH and University of Zürich, Zürich, Switzerland (M.T.W.); Department of Neurology, Medical University, Innsbruck, Austria (M.N.); Department of Neuro-Oncology, University Hospital, Torino, Italy (R.R., R.S.); Department of Neurology, University Hospital, Switzerland (P.R., M.W.); Department of Oncology, University Hospital, Zürich, Switzerland (S.H.); Department of Neurology, University Hospital, Cologne, Germany, and Research Center, Jülich, Germany (N.G.); Medicina Nucleare, Istituto Nazionale dei Tumori, Milano, Italy (F.C.)

Corresponding Author: Ulrich Roelcke, MD, Department of Neurology & Brain Tumor Center, Cantonal Hospital, Aarau, 5001 Aarau, Switzerland (roelcke@ksa.ch).
†These authors contributed equally to this study.

• 33 patients monitored with both aminoacid PET and MRI during treatment with temozolomide.

• A 25% reduction of the metabolically active tumor volume on PET was observed after 2,3 months compared with 25% reduction on MRI after 16,8 months.

• The reduction of the metabolically active tumor volume on PET, but not tumor volume reduction on MRI, correlated with improved seizure control (p=0.012).
Tracking tumour evolution in glioma through liquid biopsies of cerebrospinal fluid

Alexandra M. Miller,1,2,3 Bonak H. Shah,2,3,12 Elena I. Pantolov,2,3 Maryam Pourmalek,1,2 Samuel Briggs,2,3 Natalie Dellafavour,4 Yuxuan Zheng,2,3 Anna Skulidou,2,3 Saman A. Mehta,1 Carl Campos,2,3 Wan-Ying Hsieh,5 S. Duygu Secuklu,2,3 Lilian Ling,2 Fadil Meng,2 Xiaohong Jiao,2 Aliaxandra Samolla,2 Tejus A. Bale,2 Dana Y. V. Tsu2,4 Christian Grommes,2 Arno Vials2,3 Mark M. Soenwirdano,2,3,4 Wimse Tabara,2 Carmen W. Bubnan,2,3 Lisa M. DeAngelis,2 Robert I. Young,2,3 Michael F. Berger2,3,4,5,6,7 and Ingo K. Meilinghoff2,3,4

Diffuse gliomas are the most common malignant brain tumours in adults and include glioblastomas and World Health Organization (WHO) grade II and grade III tumours (sometimes referred to as lower grade gliomas). Genetic tumour profiling is used to classify disease and guide therapy,2,3 but involves brash surgery for tissue collection; repeated tumour biopsies may be necessary for accurate genetic testing over the course of the disease.10,11 While the detection of circulating tumour DNA (ctDNA) in the blood of patients with primary brain tumours remains challenging11,12, sequencing of ctDNA from the cerebrospinal fluid (CSF) may provide an alternative way to genotype gliomas with lower morbidity and cost.13,14 We therefore evaluated the representation of the glioma genome in CSF from 85 patients with glioma who underwent a lumbar puncture because they showed neurological signs or symptoms. Here we show that tumour-derived DNA was detected in CSF from 42 of 85 patients (49.4%) and was associated with disease burden and adverse outcome. The genetic lesions in the glioma in the CSF included a broad spectrum of genetic alterations and closely resembled the genomes of the tumour biopsies. Alterations that occur early during tumorigenesis, such as inactivation of tumour suppressor genes (TSGs), were detected in the CSF of patients with glioblastoma with the same frequency and time scale as in glioblastoma biopsies. These findings suggest that CSF is a potential source of ctDNA to monitor glioma progression. The ability to monitor the evolution of the glioma genome through a minimally invasive technique could advance the clinical development and use of genotypically-directed therapies for glioma, one of the most aggressive human cancers.

Eighty-five adults with glioma underwent collection of CSF as part of their routine clinical evaluation for neurological signs or symptoms. Diagnoses included WHO grade IV glioblastoma (46/85; 54%), WHO grade III glioma (26/85; 31%) and WHO grade II glioma (13/85; 15%). The median duration of disease before CSF collection was 355 days for IDH wild-type glioblastomas (GBMs) (43 days for both IDH1 and IDH2), 473 days for IDH wild-type lower-grade gliomas (LGGs), and 2,077 days for IDH mutant LGGs (Extended Data Fig. 1). Indicators for lumbar puncture included signs or symptoms of CNS infection, leptomeningeal tumour spread, or increased intracranial pressure. All samples were analysed using MSK IMPACT, a custom, FDA-authorized next generation sequencing-based tumour sequencing assay.2,3 We detected at least one tumour-derived genetic alteration in the CSF from 42 of 85 patients with glioma (49.4%). By contrast, we did not detect ctDNA variants in CSF from individuals with non-malignant neurological conditions (Extended Data Table 1). In our patients with glioma, several radiographic findings were associated with shedding of tumour DNA in the CSF, including tumour progression ($p < 0.0005$, Fisher’s exact test), tumour burden ($p = 0.000003$, Wilcoxon rank sum test and spread of tumour towards the ventricular system or subarachnoid space ($p = 0.02$, Fisher’s exact test; Table 1, Extended Data Fig. 2). The latter finding is consistent with a prior study that collected CSF during surgery, in which tumour DNA was more commonly detected in CSF from patients with glioma abutting a CSF reservoir or cortical surface.15 The presence of tumour DNA in CSF was associated with shorter survival following CSF collection (Extended Data Fig. 2). In a multivariable analysis, the presence of CSF DNA remained a statistically significant prognostic factor, even after adjustment for per cent extent of resection at original glioma tumour burden at CSF collection, and IDH status (Extended Data Table 2, Supplementary Table 1). Subjects who had CSF DNA in their CSF experienced a fourfold higher risk of death than subjects who did not ($p = 0.000003$). We found no significant association between CSF DNA positive CSF and glioma grade, disease duration, or prior therapy. Most patients with CSF DNA-positive CSF (35/59, 90%) did not have detectable malignant cells in the CSF as assessed by standard CSF cytopathological analysis.

Cetrtan genetic alterations occur at the earliest stages of glioma development. These alterations are viewed as ‘truncal’ events during tumour evolution and are used to define genotypically distinct subtypes of glioma.9,14 For example, the detection of IDH1 or IDH2 mutations in glioma is used to define glioma subtypes,9,14 and these mutations also have clinical implications for treatment (Extended Data Table 1). The relationship between CSF DNA and disease burden is supported by the observation that patients with CSF DNA-positive CSF had a shorter survival compared to those without CSF DNA-positive CSF (Extended Data Table 2). We examined whether these combinations of genetic alterations (TGG signatures) could be detected in the CSF and matched the genetic signature of the original tumour. We sequenced all available tumour biopsies from patients with CSF DNA-positive CSF. These included ten LGGs, twenty GBMs, and six tumours with DNA hypermutation in tissue or CSF in all cases (100%) of the LGG patients without DNA
Detection of Histone H3 mutations in cerebrospinal fluid-derived tumor DNA from children with diffuse midline glioma

Tina Y. Huang, Andrea Piunti, Rishi R. Lulla, Jin Qi, Craig M. Horbinski, Tadanori Tomita, C. David James, Ali Shilatifard and Amanda M. Saratsis

Abstract

Diffuse midline gliomas (including diffuse intrinsic pontine glioma, DIPG) are highly morbid glial neoplasms of the thalamus or brainstem that typically arise in young children and are not surgically resectable. These tumors are characterized by a high rate of histone H3 mutation, resulting in replacement of lysine 27 with methionine (K27M) in genes encoding H3 variants H3.3 (H3F3A) and H3.1 (HIST1H3B). Detection of these gain-of-function mutations has clinical utility, as they are associated with distinct tumor biology and clinical outcomes. Given the paucity of tumor tissue available for molecular analysis and relative morbidity of midline tumor biopsy, CSF-derived tumor DNA from patients with diffuse midline glioma may serve as a viable alternative for clinical detection of histone H3 mutation. We demonstrate the feasibility of two strategies to detect H3 mutations in CSF-derived tumor DNA from children with brain tumors (n = 11) via either targeted Sanger sequencing of H3F3A and HIST1H3B, or H3F3A c.83 A > T detection via nested PCR with mutation-specific primers. Of the six CSF specimens from children with diffuse midline glioma in our cohort, tumor DNA sufficient in quantity and quality for analysis was isolated from five (83%), with H3.3K27M detected in four (66.7%). In addition, H3.3G34V was identified in tumor DNA from a patient with supratentorial glioblastoma. Test sensitivity (87.5%) and specificity (100%) was validated via immunohistochemical staining and Sanger sequencing in available matched tumor tissue specimens (n = 8). Our results indicate that histone H3 gene mutation is detectable in CSF-derived tumor DNA from children with brain tumors, including diffuse midline glioma, and suggest the feasibility of “liquid biopsy” in lieu of, or to complement, tissue diagnosis, which may prove valuable for stratification to targeted therapies and monitoring treatment response.

Keywords: Cerebrospinal fluid, Liquid biopsy, Diffuse midline glioma, Diffuse intrinsic pontine glioma (DIPG), H3K27M
LIMITED PROGRESS IN TREATING GLIOBLASTOMAS

**FDA approvals**

- **Radiotherapy**
- **Lomustine**
- **Carmustine**
- **Gliadel wafer** (carmustine implant)
- **Macdonald criteria**: MRI + steroids; WHO Pathology Criteria
- **Stupp regimen** as standard treatment for GBM
- **Temozolomide** for relapsed AA
- **Bevacizumab** for recurrent GBM
- **Temozolomide** for relapsed AA
- **Novo TTF-100A**

**Technology Advances**

- **1970**: First US commercial CT
- **1980**: First US commercial MRI
- **1990**: Levin criteria: CT scans
- **1990**: Macdonald criteria: MRI + steroids; WHO Pathology Criteria
- **2000**: Brain Tumor Clinical Trial Endpoints Workshop 1
- **2010**: Response Assessment in Neuro-Oncology (RANO) criteria
- **2020**: Do not duplicate or distribute without permission from author and ESO
EFFECT OF TUMOR-TREATING FIELDS PLUS MAINTENANCE TEMOZOLOMIDE VS MAINTENANCE TEMOZOLOMIDE ALONE ON SURVIVAL IN PATIENTS WITH GLIOBLASTOMA

Median PFS from randomization for the tumor-treating fields (TTFields) plus TMZ group was 6.7 months and was 4.0 mo. for the temozolomide-alone group (hazard ratio [HR], 0.63; 95% CI, 0.52-0.76; \( P < .001 \)). B, Median survival from randomization was 20.9 for the TTFields plus TMZ group vs 16.0 mo. for the temozolomide-alone group (HR, 0.63; 95% CI, 0.53-0.76; \( P < .001 \)).

Stupp et al, JAMA 2015
SHORT-COURSE RADIATION PLUS TEMOZOLOMIDE IN ELDERLY PATIENTS WITH Glioblastoma Aged > 65 YRS

Table 3. Exploratory Analyses of Overall Survival Rate at 12, 18, and 24 Months According to Treatment Group and MGMT Status.

<table>
<thead>
<tr>
<th>Population</th>
<th>At 12 Months</th>
<th>At 18 Months</th>
<th>At 24 Months</th>
</tr>
</thead>
<tbody>
<tr>
<td>All patients</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Radiotherapy alone</td>
<td>22.2 (17.5−27.3)</td>
<td>10.8 (7.4−14.8)</td>
<td>2.8 (1.2−5.4)</td>
</tr>
<tr>
<td>Radiotherapy plus temozolomide</td>
<td>37.8 (32.1−43.6)</td>
<td>20.0 (15.5−24.9)</td>
<td>10.4 (7.1−14.5)</td>
</tr>
<tr>
<td>Patients with unmethylated MGMT</td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Radiotherapy alone</td>
<td>21.3 (13.7−30.0)</td>
<td>12.7 (6.9−20.3)</td>
<td>3.8 (1.1−9.6)</td>
</tr>
<tr>
<td>Radiotherapy plus temozolomide</td>
<td>32.3 (23.0−42.0)</td>
<td>13.4 (7.3−21.2)</td>
<td>6.7 (2.7−13.1)</td>
</tr>
<tr>
<td>Patients with methylated MGMT</td>
<td></td>
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</tr>
<tr>
<td>Radiotherapy alone</td>
<td>29.9 (19.9−40.5)</td>
<td>13.6 (7.0−22.4)</td>
<td>4.1 (1.1−10.4)</td>
</tr>
<tr>
<td>Radiotherapy plus temozolomide</td>
<td>55.7 (44.7−65.3)</td>
<td>34.1 (24.4−44.0)</td>
<td>17.8 (10.5−26.7)</td>
</tr>
</tbody>
</table>

Fig. 1. Immunosuppression in the glioblastoma microenvironment. (A) Partially driven by increased STAT3 expression, glioblastoma cells secrete immunosuppressive factors such as TGF-β-2, PGE, IL-1, IL-10 and FGL2, all of which suppress the activity of effector cells. PD-L1 expressed on its surface also engages PD-1 to suppress effector activity. M-CSF, TGF-β-1 and IL-10 skew macrophages to the immunosuppressive M2 phenotype. (B) M2 Macrophages secrete TGFβ-1 and IL-10, suppressing effector cells further. (C) Regulatory T cells secrete TGFβ-1 and IL-10 as well, further suppressing immune reactivity, while also expressing PD-L1. (D) M-CSF, TGFβ-1 and IL-10, secreted by the glioblastoma, skew tumor associated macrophages to the immunosuppressive M2 phenotype. (D) Antigen is presented to T cells by antigen presenting cells within an MHC molecule, but a costimulatory signal from CD80/86 to CD28 is required for activation. CD80/86 can also suppress activity by engaging the CTLA4 receptor on the activated T cell.
APPROACHES OF IMMUNOTHERAPY FOR GLIOBLASTOMA

- Vaccination against EGFRvIII (Rindopepimut, Celldex)
- Peptide vaccination (IMA950, Immatics)
- DC/lysate-based immunotherapy (DCVax, NW Biotherapeutics)
- DC/peptide-based immunotherapy (ICT-107, Immunocellular)
- DC/CMV-targeted immunotherapy (Duke)

- TGF-β antisense oligonucleotide (Trabedersen, Antisense Pharma/Isarna)

- TGF-β receptor antagonists (LY2157299, Lilly)

- Nivolumab (anti-PD-1) (BMS)
- Anti-PD-L1 (MPDL3280A) (Roche)
- Pembrolizumab (anti-PD-1) (MK-3475, MSD)
- Ipilimumab (anti-CTLA-4) (Yervoy®, BMS)
“Rindopepimut with bevacizumab for patients with relapsed EGFRvIII-expressing glioblastoma (ReACT): results of a double-blind randomized phase 2 trial”,

Reardon et al, 2017 (in press)
“Rindopepimut with temozolomide for patients with newly diagnosed, EGFRvIII-expressing glioblastoma (ACT IV): results of a randomized, double-blind, international phase 3 trial”

Weller et al, Lancet Oncol, 2017
PHASE III STUDY OF NIVOLUMAB vs BEVACIZUMAB IN GLIOBLASTOMA AT FIRST RECURRENCE (ChecKMate 143)

• No OS differences (10 months vs 9-8 months)
• PFS significantly longer (~ 3 months) for bevacizumab.
• Overall Response Rate 7.8% vs 11.3%
• Duration of response 11 months vs 5.3 months.

Reardon et al, World Federation of Neuro-Oncology meeting, May 2017
(manuscript submitted, 2019)
CheckMate 498 CA209-498 (NCT02617589): Phase 3 Randomized, Open-Label Study of RT in Combination With Nivolumab or TMZ in Newly Diagnosed MGMT-Unmethylated GBM

Key inclusion criteria
- Newly diagnosed brain cancer or tumor called GBM or GBM
- Males and females ≥ 18 years old
- Tumor test result shows MGMT unmethylated
- KPS ≥ 70%

Experimental: Nivolumab IV infusion + RT Q2W (dose as specified); then nivolumab Q4W

Active Comparator: Standard therapy with TMZ + RT (dose as specified)

Estimated enrollment N = 550

Start Date: February 2016
Estimated Study Completion Date: October 2019
Estimated Primary Completion Date: March 2019
Status: currently recruiting participants
Study Sponsor: Bristol-Myers Squibb
Collaborator: Ono Pharmaceutical Co. Ltd

Primary Outcome Measure: 3-year OS
Secondary Outcome Measure: PFS; 2-year OS

Neoadjuvant anti-PD-1 immunotherapy promotes a survival benefit with intratumoral and systemic immune responses in recurrent glioblastoma


Glioblastoma is the most common primary malignant brain tumor in adults and is associated with poor survival. The Ivy Foundation Early Phase Clinical Trials Consortium conducted a randomized, multi-institution clinical trial to evaluate immune responses and survival following neoadjuvant and/or adjuvant therapy with pembrolizumab in 35 patients with recurrent, surgically resectable glioblastoma. Patients who were randomized to receive neoadjuvant pembrolizumab, with continued adjuvant therapy following surgery, had significantly extended overall survival compared to patients that were randomized to receive adjuvant, post-surgical programmed cell death protein 1 (PD-1) blockade alone. Neoadjuvant PD-1 blockade was associated with upregulation of T cell- and interferon-γ-related gene expression, but downregulation of cell-cycle-related gene expression within the tumor, which was not seen in patients that received adjuvant therapy alone. Focal induction of programmed death-ligand 1 in the tumor microenvironment, enhanced clonal expansion of T cells, decreased PD-1 expression on peripheral blood T cells and a decreasing monocytic population was observed more frequently in the neoadjuvant group than in patients treated only in the adjuvant setting. These findings suggest that the neoadjuvant administration of PD-1 blockade enhances both the local and systemic antitumor immune response and may represent a more efficacious approach to the treatment of this uniformly lethal brain tumor.
First results on survival from a large Phase 3 clinical trial of an autologous dendritic cell vaccine in newly diagnosed glioblastoma

DCVax-L

**PROS**

- The largest number of patients enrolled in a DC IT trial. In a 2:1 ratio: SOC + DC n = 222; SOC + placebo n = 99.
- Median OS of all 331 patients enrolled 23.1 months (95% CI 21.2 – 25.4).
- 30% of the ITT patients showed prolonged survival: 40.5 months at Kaplan-Meier analysis.

**CONS**

- No survival data for the two patients arms available «because the number of the events (i.e. deaths) in the trial was not sufficient to justify unbliding».
- No information on the IDH status.
- All patients could receive DCVax-L at recurrence.
Recurrent Glioblastoma Treated with Recombinant Poliovirus

Annick Desjardins, M.D., Matthias Gromeier, M.D., James E. Herndon II, Ph.D., Nike Beaubier, M.D., Dani P. Bolognesi, Ph.D., Allan H. Friedman, M.D., Henry S. Friedman, M.D., Frances McSherry, M.A., Andrea M. Muscat, B.Sc., Smita Nair, Ph.D., Katherine B. Peters, M.D., Ph.D., Dina Randazzo, D.O., John H. Sampson, M.D., Ph.D., Gordana Vlahovic, M.D., William T. Harrison, M.D., Roger E. McLendon, M.D., David Ashley, M.B., B.S., Ph.D., and Darell D. Bigner, M.D., Ph.D.

Figure 1. Overall Survival among Patients Who Received PVSRIPO and Historical Controls.

Tick marks indicate censored data. PVSRIPO denotes recombinant nonpathogenic polio–rhinovirus chimera.
Regression of Glioblastoma after Chimeric Antigen Receptor T-Cell Therapy

Christine E. Brown, Ph.D., Darya Alizadeh, Ph.D., Renate Starr, M.S., Lihong Weng, M.D., Jamie R. Wagner, B.A., Araceli Naranjo, B.A., Julie R. Ostberg, Ph.D., M. Suzette Blanchard, Ph.D., Julie Kilpatrick, M.S.N., Jennifer Simpson, B.A., Anita Kurien, M.B.S., Saul J. Priceman, Ph.D., Xiuli Wang, M.D., Ph.D., Todd L. Harshbarger, M.D., Massimo D'Apuzzo, M.D., Julie A. Ressler, M.D., Michael C. Jensen, M.D., Michael E. Barish, Ph.D., Mike Chen, M.D., Ph.D., Jana Portnow, M.D., Stephen J. Forman, M.D., and Behnam Badie, M.D.
CONCLUSIONS

• Molecular biology has definitely established the value of standard treatments for molecular subtypes of lower grade gliomas.

• Glioblastomas are “cold tumors”: new approaches (i.e. combination strategies) may hopefully improve the results following immunotherapy.

• Cross-talks between tumor cells and normal cells of microenvironment (neurons, astrocytes, microglia) are of rising importance.