

High Grade Glioma Through the Ages



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Although brain tumours represent <2% of all human cancers, they are the leading cause of cancer-related deaths in patients under 40. Of these, the high grade gliomas represent the most significant burden to health, with ~90% of patients with glioblastoma multiforme (GBM), the most malignant of these tumours, dying within two years of diagnosis [1]. Genetic analysis conducted at The Institute of Cancer Research in London and other organisations has begun to reveal the many genetic variants of high grade glioma, including particular differences between its adult and paediatric forms. These findings are important because ultimately they are likely to have a major impact on disease management.

Glioblastoma – even more ‘multiforme’ than we thought

Glioblastomas can be thought of as a disease of the elderly, the majority of cases developing after the age of 55, with a peak incidence at 70-80 years. Recent large-scale molecular profiling of these tumours has uncovered in exquisite detail the key genetic aberrations driving and maintaining tumorigenesis, as well as providing novel therapeutic targets for drug development [2].

As well as the specific genetic changes present at the DNA level, mRNA expression profiling has revealed the existence of several subtypes of high grade glioma based solely on their gene expression signatures [3]. These have been defined according to the predominant functions of the specific genes involved, and are known as ‘proneural’, ‘proliferative’ and ‘mesenchymal’. Such molecular fingerprints may reflect important differences in the origin and progression between different tumours that would otherwise appear indistinguishable.

In addition to adult primary glioblastomas, there are distinct clinical presentations which we are only now beginning to recognise as different diseases from the most common form. These include so-called secondary glioblastoma, which arises in young adults (35-45 years) on the back of a pre-existing lower grade lesion. They have long been hypothesised as belonging to a different genetic developmental pathway, but it is only a recent discovery of the highly restricted mutations in IDH1/2 in these cancers that has provided confirmation [4].

High grade gliomas of childhood - an unmet clinical and biological need

Ideas about high grade gliomas that arise in children have, in contrast, been controversial. Although these

tumours are rare (0.5 cases per 100,000 person-years), as in adults they make up ~75% of all malignant brain tumours. Their low incidence has made it difficult for single institutions to carry out sufficient studies to provide robust evidence for how biologically different, or similar, they may be to the adult disease(s).

Despite this, clinical evidence suggests that they are different. Gliomas in children have distinct patterns of presentation in the brain, they rarely undergo malignant transformation like the secondary glioblastomas of young adulthood, and very young children appear to respond better to chemotherapy with an improved clinical outcome compared with older patients.

With the flood of data on adult glioblastoma, the ease and accessibility of genome-wide profiling techniques, and an increased awareness of the gap in our understanding of this key element of gliomagenesis, the timing seems right for us to finally make some progress in our understanding of these devastating childhood tumours.

Paediatric high grade gliomas have a distinct genetic make-up

2010 is shaping up to be the year that paediatric high-grade glioma research went prime-time. After years of neglect, several studies have been, or are due to be, published that shed light into the genomics of these enigmatic tumours. From smaller single institution studies [5,6] to large multi-centre collaboratives, the amount of DNA copy number and expression profiling data has increased by an order of magnitude. For our own part, besides taking part in the largest study published to date along with the Children's Cancer and Leukaemia Group in the UK and St Jude Children's Research Hospital in the US [7], we have provided the first large validation set of paediatric high-grade glioma amenable to copy number profiling by array comparative genomic hybridisation from archival formalin-fixed paraffin-embedded specimens [8].

These datasets reveal significant differences in the DNA copy number alterations in childhood versus adult high-grade gliomas. The most common chromosomal abnormalities in adult glioblastoma, i.e. gains of chromosome 7 and losses of 10q (seen in >75% of cases), are considerably less frequent in the paediatric setting. Conversely, childhood tumours have high levels of 1q gain and losses of 4q and 16q, all of which are rare in adults.

Perhaps the most striking of the differences in chromosomal changes has been an absence – the finding that ~1/7th of paediatric high grade gliomas

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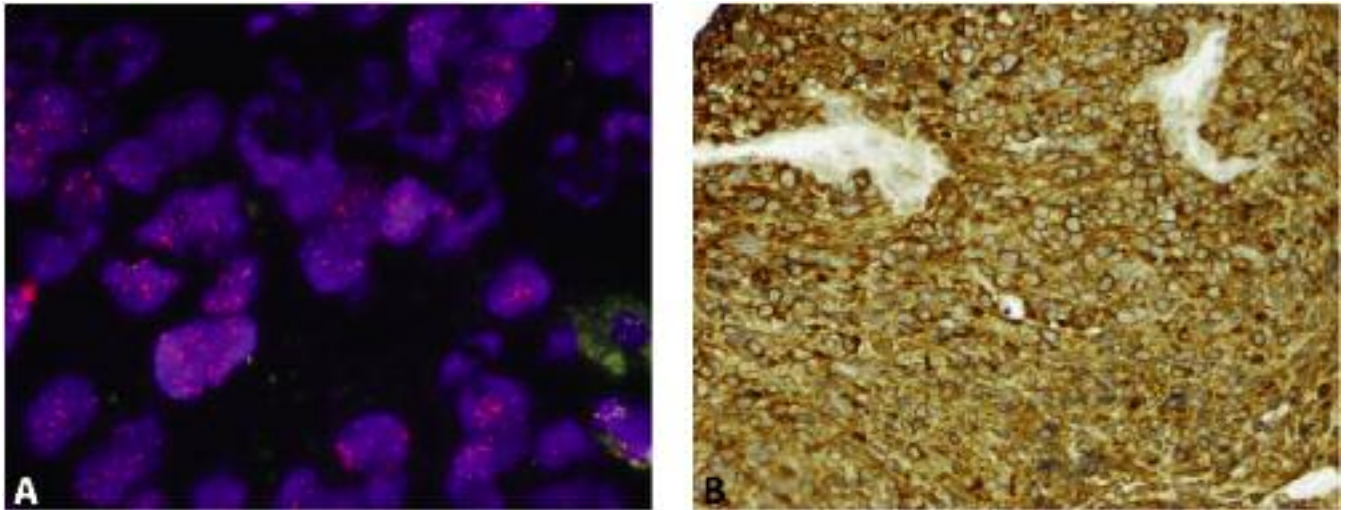


Figure: PDGFRA is amplified and overexpressed in paediatric high-grade glioma. (A) fluorescent in situ hybridisation showing an increased copy number of specific probes directed against the PDGFRA gene (pink). (B) Immunohistochemical staining using an antibody specific to PDGFRA protein showing widespread receptor expression in an amplified tumour.

have no detectable changes in DNA copy number following profiling on numerous array platforms. This is a surprising observation for such malignant tumours, and one that is not seen in adult lesions. Whether there are epigenetic or more subtle mutational changes occurring in these cases is not yet clear; however, it is intriguing that these stable genome tumours have also been reported in paediatric ependymoma and supratentorial primitive neuroectodermal tumours, perhaps hinting at some commonality in paediatric brain tumour biology.

Platelet-derived growth factor alpha (PDGFRA) comes in from the cold

In terms of bona fide high level gene amplifications, there is a clear winner in paediatric high grade glioma. Although numerous genes reported in adult glioblastoma are also represented in children, most only occur at very low frequencies, often in single cases. In contrast, amplification of PDGFRA at chromosome 4q12 is by far the most common, being amplified in fully 20% of paediatric glioblastoma. This event is even more common in two childhood-specific instances of the disease – diffuse intrinsic pontine glioma, which affects the brainstem, and post-irradiation glioma, which is a second malignancy arising after cranio-spinal irradiation for an earlier cancer. In these subtypes, PDGFRA amplification rises to ~50% of cases.

PDGFRA amplification had previously been noted in glioblastoma, and specific small molecule inhibitors were used in early phase clinical trials with little success. To determine whether the gene was playing a critical role in these tumours, or just an incidental role, expression profiles of childhood and adult tumours were compared. Surprisingly, when

a set of genes defined as being upregulated after PDGFRA had been amplified were examined in the two age groups, completely distinct sets of genes were differentially expressed in the paediatric versus adult tumours. Furthermore, this paediatric-specific PDGFRA signature was active in over half the childhood tumours, and was therefore present even in the absence of gene amplification.

In adults, those (less frequent) cases with PDGFRA amplification were associated with the ‘proneural’ subclass, as defined by expression profiling. This group of tumours was also tightly linked to IDH1/2 mutations, and thus to the secondary glioblastoma pathway. In childhood high-grade glioma, IDH1/2 mutations are almost entirely absent, creating another clear distinction from a form of the adult disease. It is perhaps not surprising then, that PDGFRA in children is not associated with the proneural group, but rather with expression of numerous cell cycle-associated genes which place these tumours firmly in the ‘proliferative’ subclass.

In retrospect, this makes sense, as these tumours in adults appear to be driven by the most commonly amplified gene in that age group, viz. EGFR. Such an abnormality is considerably less common in paediatric cases, and instead the proliferative pathways are driven by the distinct receptor tyrosine kinase, PDGFRA. Clearly, a fresh look at strategies targeting this receptor in children is warranted, where novel predictive markers may be at play.

High-grade glioma across all ages comprises a genomic spectrum of disease

Although clear evidence is starting to emerge for differences between childhood and adult high-grade gliomas, it is important to recognise that there is still a high degree

of overlap between the two age groups. Of particular note, there are a proportion of paediatric tumours which do look rather like their adult counterparts – EGFR amplified, with gains of chromosome 7, losses at 10q, etc., and with a ‘classical’ or ‘mesenchymal’ gene expression signature. However, these tumours are in the minority.

Similarly, there are (less frequent) tumours from elderly patients whose genetics places them firmly of the type that is rather prevalent in children – PDGFRA amplified, with a specific proliferative signature, and possibly with gains of 1q and losses of 16q. We have come to call these cases ‘Group P’ – predominantly paediatric, PDGFRA-driven and proliferative. What is fundamental is that these tumours are clearly distinct from the adult proliferative subtype, which is EGFR-driven, and also separate from the proneural groupings. In adults, this subtype is associated with IDH1/2 mutations, absent in children; in the paediatric disease, however, the proneural gene expression group is still evident, but not associated with PDGFRA.

It seems clear that rather than a small number of highly segregated subclasses, high-grade glioma across all the groups forms a genomic spectrum of disease, and only by studying tumours at whatever age they arise will we fully understand all the different subtleties of how they fit together.

Perspectives on progress in a particularly paediatric puzzle

Having defined this ‘Group P’ end of the genomic spectrum of high-grade glioma, how can we use this knowledge to understand the differences between similar tumours that arise at different stages of life? There are already hints from secondary

Table: Key genetic differences between paediatric and adult high-grade glioma

	Paediatric	Adult
Predominant amplification	PDGFRA	EGFR
Chromosomal gains	1q	7
Chromosomal losses	16q	10q
IDH1/2 mutation	No	Yes (Secondary GBM)
Stable genomes	Yes	No
Expression signatures	PDGFRA:proliferative	PDGFRA & IDH1/2: proneural

versus primary adult glioblastoma, as well as the IDH1/2 mutation story, that very distinct development processes play a key role in disease initiation. Given the short time to malignancy apparent in paediatric tumours, this is probably even more critical in children.

The differential targeting of receptor tyrosine kinases in childhood and adult tumours suggests that the developing brain may be more sensitive to PDGFRA-directed cancer

initiation than more mature cells of the central nervous system. This in turn may reflect a unique cell of origin for the majority of childhood high-grade gliomas in comparison with their adult counterparts. Identifying the key genetic differences which separate these diseases is only the first part in the process of understanding the prospect of treating and preventing these highly aggressive, and currently incurable, tumours. ■

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