Brain Tumour Cells Can Survive and Thrive in the Hypoxic Solid Tumour Microenvironment

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Abstract

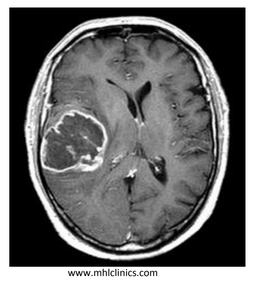
Background: Hypoxia is associated with the increased malignancy of a broad range of tumours. Paradoxically, hypoxia has also been shown to induce cell cycle arrest, although the impact of hypoxia on tumour cell proliferation is poorly understood.

Aim: To investigate the effects of different oxygen levels on brain tumour cell proliferation and survival.

Results & Conclusion: Only severe hypoxia (<0.1% O_2) caused reduced cell proliferation and survival. This suggests that the majority of cells in brain tumours will proliferate and survive despite exposure to hypoxia.

Glioblastoma Multiforme (GBM)

- Glioblastoma Multiforme is the most common malignant brain tumour. Average patient survival time is just 12-14 months
- Treatment resistance is a major obstacle and is at least partly responsible for poor prognosis



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Tumour Hypoxia

- The blood vessels within tumours are abnormal, leading to an inadequate supply of oxygen (hypoxia). Normal brain tissue contains 5-8% O_2 while on average brain tumours contain just 1% O_2
- Hypoxia increases tumour cell invasion and resistance to treatment. It is associated with a poorer patient prognosis
- Severe (<0.1% O_2) hypoxia can cause some cells to stop growing but it is not known how the oxygen levels in brain tumours (1% O_2) affect tumour growth

Methods

- Cells were exposed to different levels of oxygen using a Don Whitley Scientific H35 HypOxystation
- Cells were stained with PI +/- Annexin V-FITC and analysed by flow cytometry

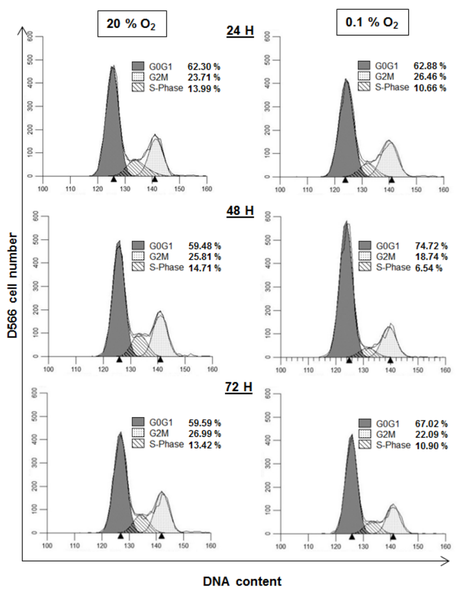
- Gene expression was assessed using qRT-PCR

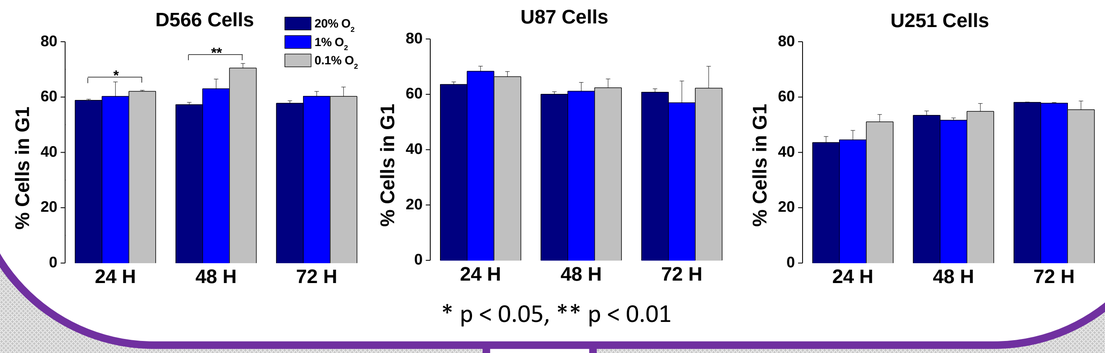
GBM cell cycle is not affected by moderate hypoxia

There was no change in cell cycle following exposure to moderate hypoxia in any cell lines (*N* =3).

D566 cells showed a transient G_1 phase arrest in response to severe hypoxia.

U87 and U251 cell cycle was not affected by severe hypoxia.

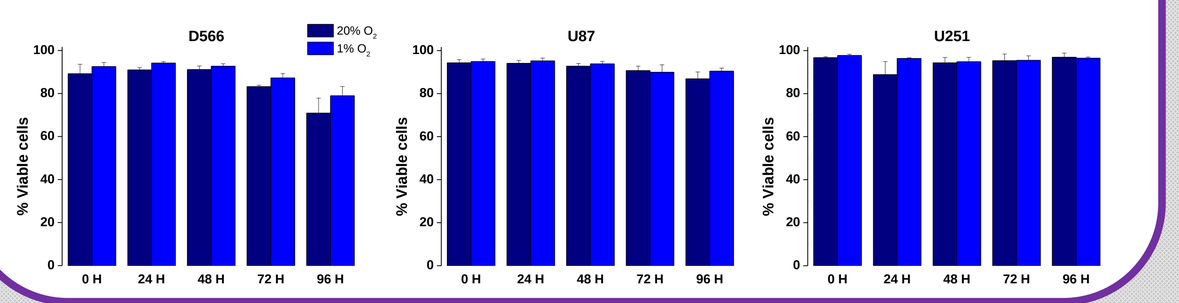




* p < 0.05, ** p < 0.01

GBM cell survival is not affected by moderate hypoxia

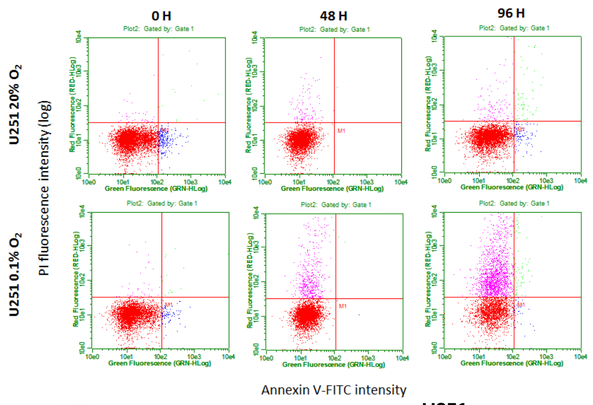
There was no change in cell survival following up to 5 days exposure to 1% O_2 in any of the cell lines (*N* = 3).

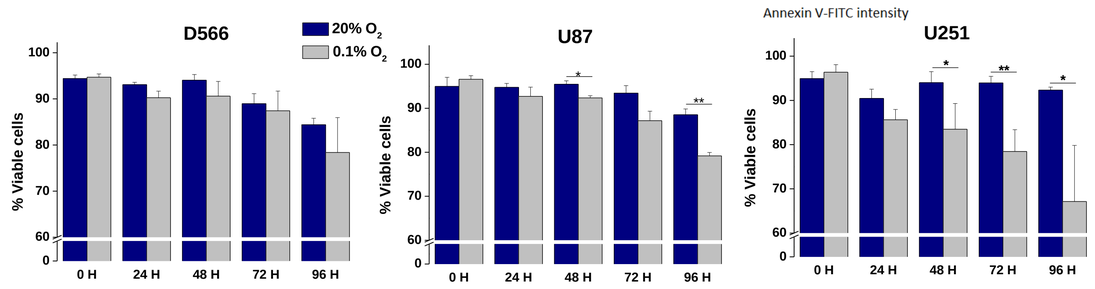


Severe hypoxia causes GBM cell death

Both U251 and U87 cells displayed increased cell death following exposure to severe hypoxia.

There was no change in D566 cell survival (*N* = 3).

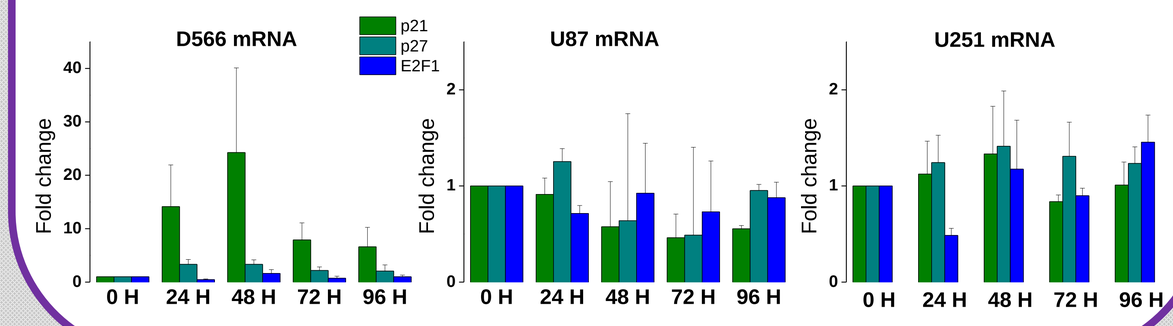




* p < 0.05, ** p < 0.01

p21 expression is associated with cell cycle arrest in response to severe hypoxia

D566 cells (*N* = 3) showed an increase in the expression of the cell cycle inhibitor p21 following exposure to severe hypoxia. There was no change in the expression levels of cell cycle related genes in U251 (*N* = 3) or U87 cells (*N* = 2).



Conclusion

- Only severe hypoxia (<0.1% O_2) causes GBM cell cycle arrest
- Only a small proportion of cells in brain tumours are exposed to hypoxia this severe, therefore the majority of cells will proliferate and survive despite exposure to hypoxia
- These findings help to explain how hypoxic tumours can be incredibly fast growing and aggressive as is the case with GBM