Therapeutic response to chemotherapeutic drugs of glioma-PDX and correlation to common mutations identified by panel sequencing

A. Orthmann[1], A. Hoffmann[1], R. Zeisig[1], J. Haybäck[2], A. Jödicke[3], S. Kuhn[4], M. Linnebacher[5], J. Hoffmann[1], I. Fichtner[1]

[1]Experimental Pharmacology & Oncology GmbH, Germany; [2]Department of Pathology - Medical University of Graz, Austria;
[3]Clinic for Neurosurgery - Vivantes Klinikum, Germany; [4]Clinic for Neurosurgery - Klinikum Ernst von Bergmann, Germany;
[5]Molecular Oncology and Immunotherapy - Medical University of Rostock, Germany

Introduction

We have recently established 15 new patient derived xenografts (PDX) from glioblastoma multiforme (GBM). The PDX models are kept in early passages and coincide with the original tumor histology. As part of the PDX-model characterization, sensitivity of the glioma towards a panel of standard of care (temozolomide) and other drugs was tested including the chemotherapeutics irinotecan, the mTOR inhibitor everolimus, the multi-kinase inhibitor sorafenib, a monoclonal antibody targeting tumor angiogenesis (bevacizumab) and salinomycin.

Methods and Results

Drug Sensitivity

The glioma PDX displayed a heterogeneous response to the tested drug panel with strong initial sensitivity to the chemotherapeutics temozolomide (TMZ) and irinotecan. Based on a T/C value ≤ 30% as cutoff, 11 glioma were defined as responder and 4 glioma as non responder to TMZ. In general, the targeted drugs were less effective. Salinomycin and sorafenib had no meaningful effect on tumor growth at all.
To identify treatment options for the non responder, we correlated the response to TMZ with the effects of everolimus, bevacizumab and irinotecan. Whereas for two TMZ resistant glioma (Glio10535 and Glio11414) no alternative treatment option have been identified, for the other two resistant PDX models (Glio10995 and Glio10888) irinotecan was identified as a potential treatment alternative by our drug sensitivity screening.

Patient data and mutations of the PDX

PDX model	Histology	WHO-Grad	Patient gender	Mutations detected in PDX
10193	Glioblastoma multiforme	IV	male	FGFR3, KDR
10315	Glioblastoma multiforme	IV	male	PIK3CA, PTEN, TP53
10485	Glioblastoma multiforme	n/a	female	EGFR, MET
10535	Glioblastoma multiforme	IV	male	no mutations detected
10612	Glioblastoma multiforme	IV	male	no mutations detected
10618	Glioblastoma multiforme	IV	female	KDR, PIK3CA
10888	Glioblastoma multiforme	IV	male	KDR, TP53
10995	Glioblastoma multiforme	IV	female	SMAD4, TP53
11305	Glioblastoma multiforme	IV	male	JAK3, PTEN
11413	Glioblastoma multiforme	IV	female	n/a
11414	Glioblastoma multiforme	IV	female	EGFR, KRAS, TP53
11433	Glioblastoma multiforme	IV	male	KDR
11575	Glioblastoma multiforme	IV	male	PIK3CA, PTPN11
11874	Glioblastoma multiforme	IV	male	FGFR3, RB1
12032	Glioblastoma multiforme	IV	male	n/a

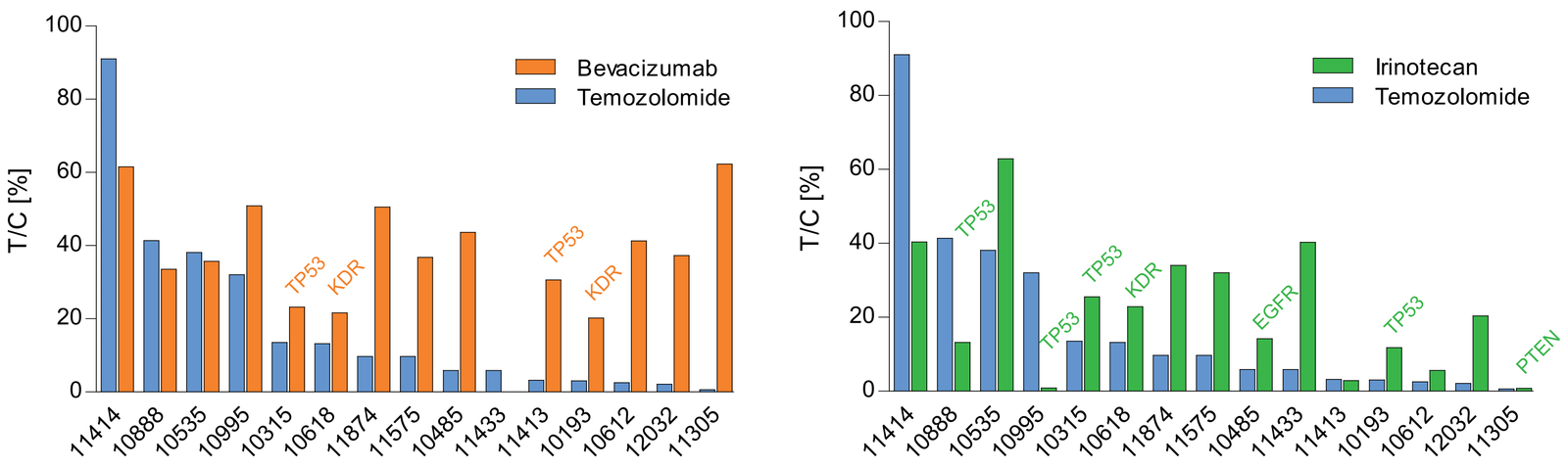
Chemosensitivity of the PDX (T/C values, %)

PDX model	Everolimus	Sorafenib	Bevazizumab	Irinotecan	Salinomycin	Temozolomide
10193	74,5	79,6	20,2	11,8	80	3
10315	83,4	60,5	23,2	25,5	78,4	13,5
10485	47,4	49,9	43,6	14,2	58,8	5,9
10535	84,2	103,9	35,7	62,9	117,1	*38,1*
10612	29,8	34	41,3	5,6	78,5	2,5
10618	47,7	64	21,6	22,9	63,7	13,2
10888	73,4	66,6	33,5	13,2	86,2	*41,4*
10995	28,1	73,7	50,9	0,9	71,4	*32*
11305	56,5	94,2	62,3	0,8	93	0,6
11413	37	74,6	30,6	2,9	61,8	3,2
11414	41,8	95,5	61,5	40,4	103,2	*91*
11433	-	-	-	40,3	85,9	5,9
11575	63,8	35,6	36,8	32	69,1	9,7
11874	81	60,9	50,5	34	67,6	9,7
12032	21,6	54,8	37,3	20,4	57,8	2,1

Correlation between mutation profile and chemosensitivity of the PDX

In a comparison of TMZ response and the glioma mutation profile we detected a TP53 mutation in 3/4 cases of the non-responder. Most interestingly these TMZ resistant glioma were found to be sensitive to treatment with irinotecan.

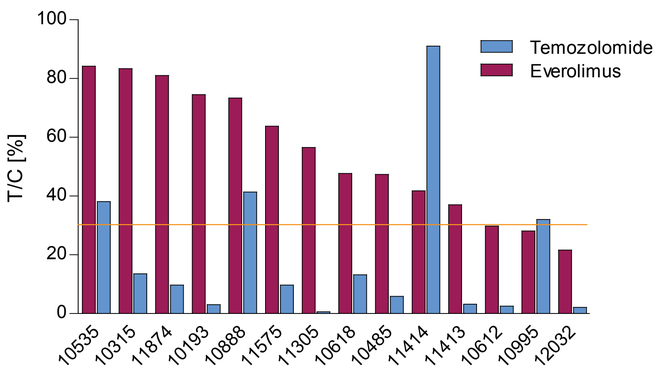
Respone of bevacizumab and irinotecan in the PDX models (T/C values, %)



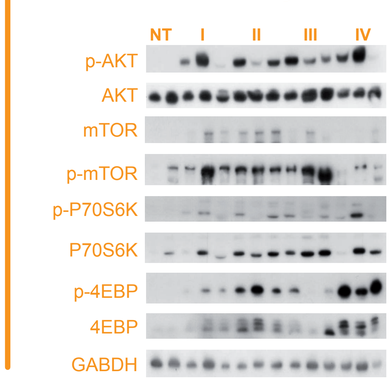
Activity of the mTOR pathway in glioma

In our drug sensitivity screening, there was almost no response to the mTOR pathway inhibitor everolimus observed. This led to the hypothesis, that in high grad glioblastoma the mTOR pathway is down regulated. We were able to analyze expression of mTOR pathway proteins in samples from patients with astrocytoma (WHO I-III) and GBM (WHO IV) and found a strong down regulation in the high grade population (GBM) only.

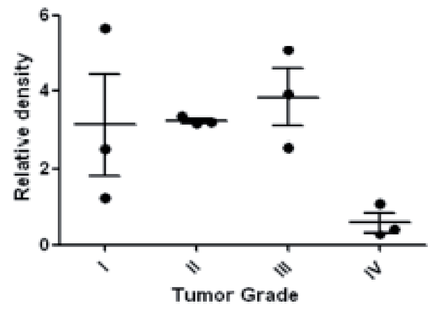
Respone of everolimus in the PDX models



Immunoblot analyses of mTOR signaling in patients

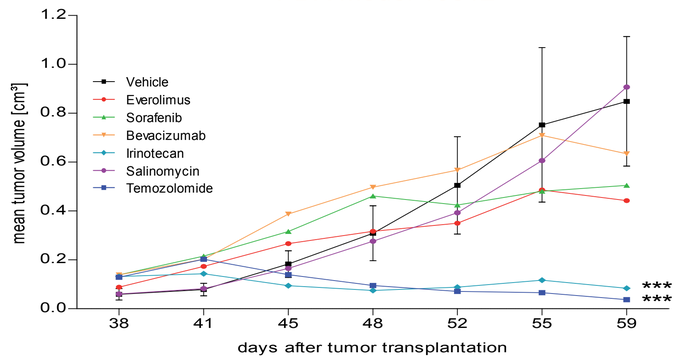


Density analyses of the mTOR immunoblots

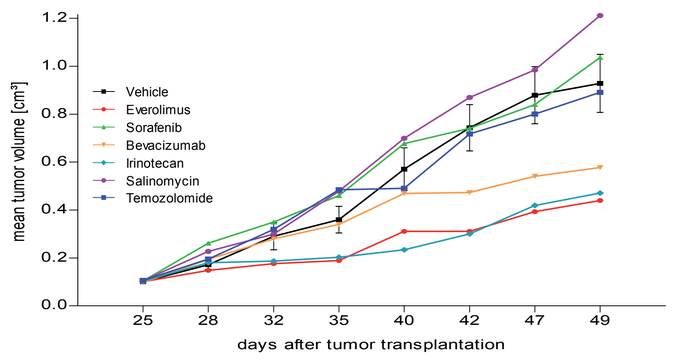


Examples for individual drug profiles

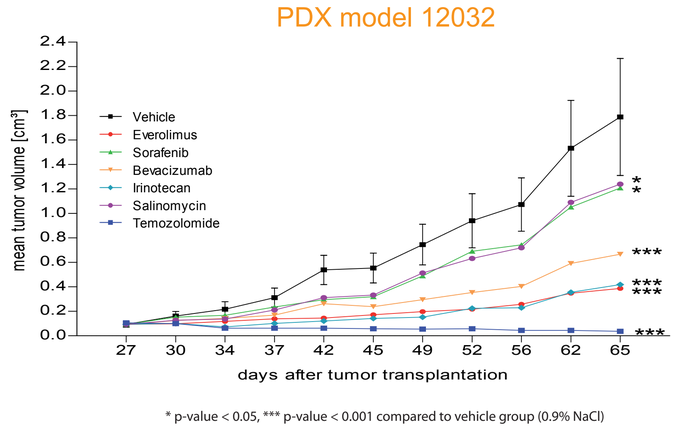
PDX model 10612



PDX model 11414



PDX model 12032



* p-value < 0.05, *** p-value < 0.001 compared to vehicle group (0.9% NaCl)

Conclusions

- we have developed a panel of 15 new GBM PDX models → these models have been used for drug sensitivity screening
- temozolomide and irinotecan have been identified as most active drugs whereas the targeted drugs are less effective
- TP53 mutation has been found in 3/4 of temozolomide resistant PDX with irinotecan as treatment alternative
- downregulation of the mTOR pathway has been identified as potential Mode of Action (MoA) for everolimus resistance

contact: andrea.orthmann@epo-berlin.com