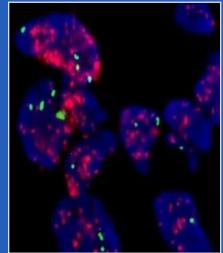
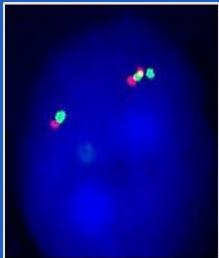




dkfz.

UniversityHospital Heidelberg

THE MOLECULAR DISSECTION OF MEDULLOBLASTOMA



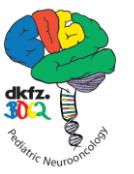
Vienna, September 29, 2012 ESMO Meeting

Stefan Pfister

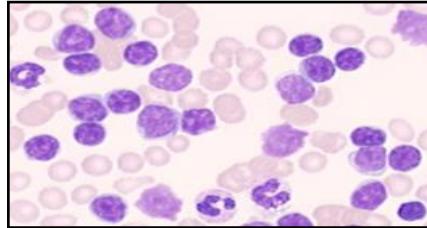
Division of Pediatric Neurooncology

www.pediatric-neurooncology.com

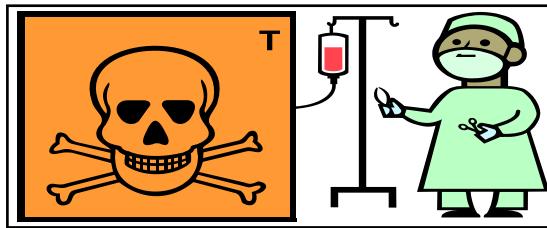
Vision: Molecular Diagnostics & Therapeutic strategies



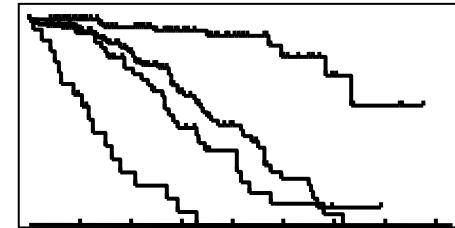
Single diagnosis



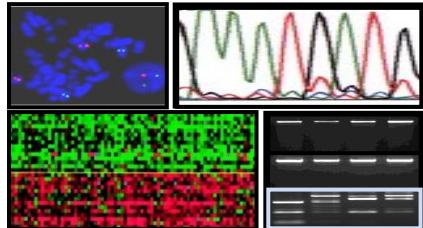
Uniform therapy



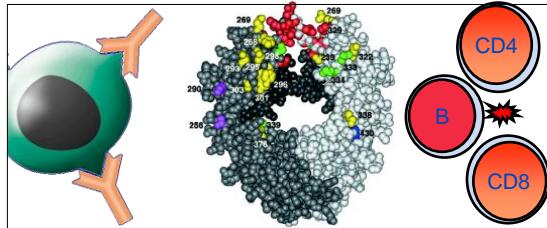
Variable results



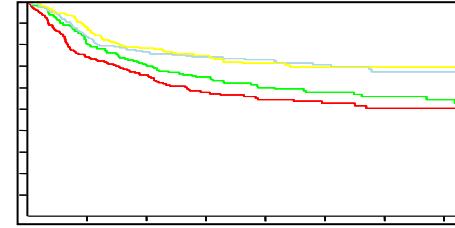
Molecular subtypes



Targeted therapy



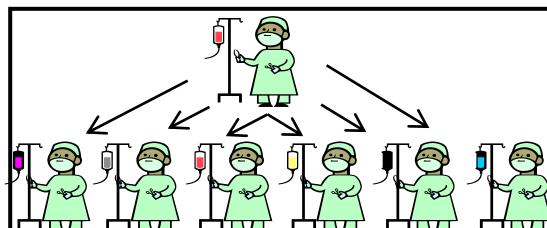
Better results



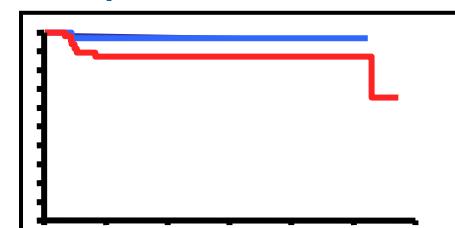
Individual profiles



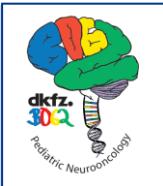
Personalized therapy



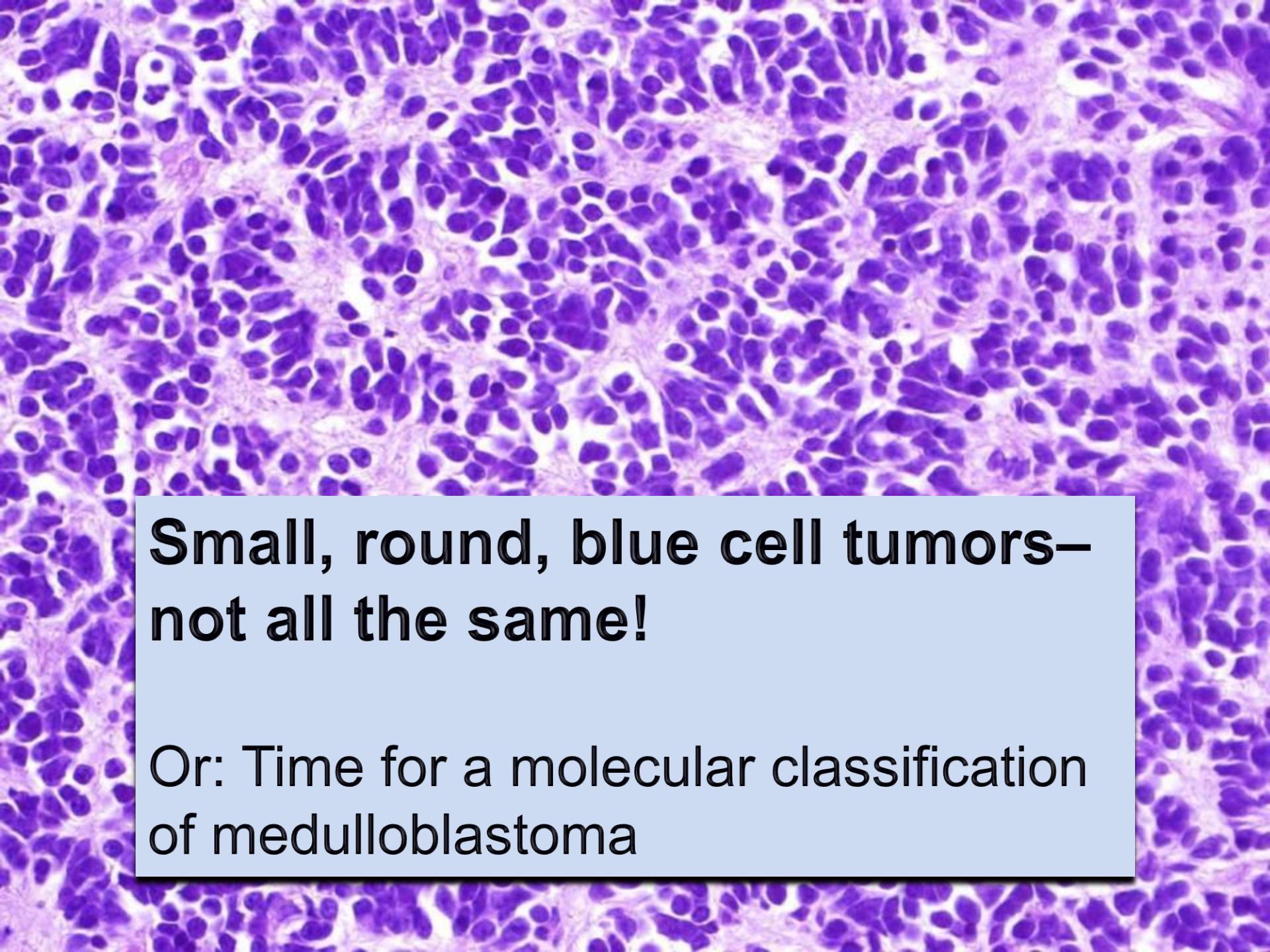
Optimal results



Agenda



- 1.) Introduction: Current risk stratification & molecular classification of MB
- 2.) News from the ICGC project PedBrain Tumor
 - Li-Fraumeni-associated MB
 - Novel recurrent mutations and common themes
 - Tetraploidy as an early event in tumorigenesis
- 3.) Outlook



**Small, round, blue cell tumors—
not all the same!**

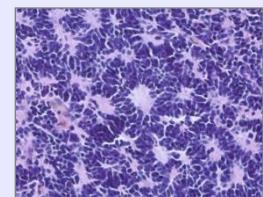
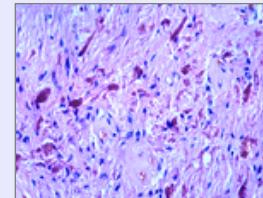
Or: Time for a molecular classification
of medulloblastoma

Childhood brain tumors (WHO)

Main histologic subtypes, WHO grade, and frequency of central nervous system tumors in children

(Data from Kaatsch et al., 2001)

	WHO grade	frequency (%)
Tumor entity		
Gliomas	Astrocytomas	I-IV
	Ependymomas	I-III
	Gangliogliomas*	I
	Oligodendrogiomas	II-III
Non-glial tumors	Embryonal tumors	IV
	Craniopharyngeomas	I
	Pineal tumors	I-IV
	Meningeomas	I-III
	Others (e.g., lymphomas, germ cell tumors, metastases)	n/a
		25,7
		4,4
		1,3
		1,2
		11,0

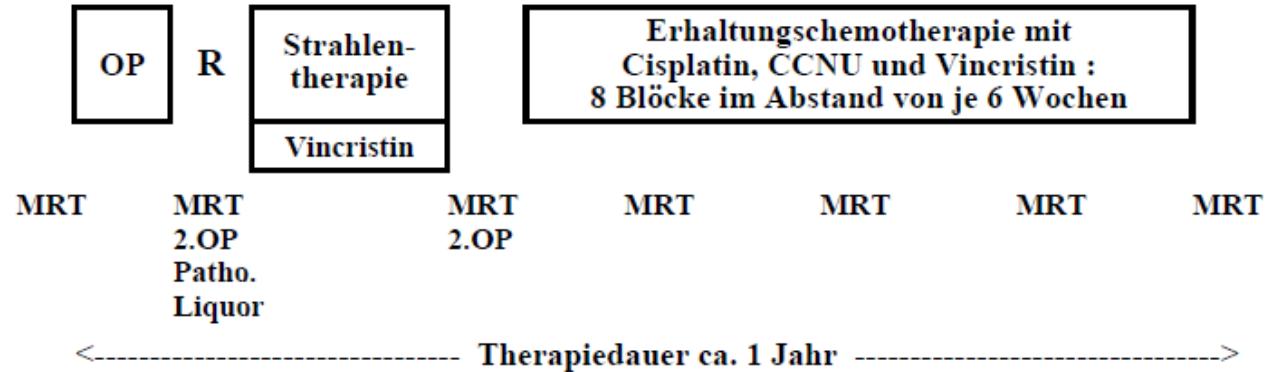


*Gangliogliomas are mixed neuronal-glial tumors

Current status of stratification (1)

Medulloblastoma

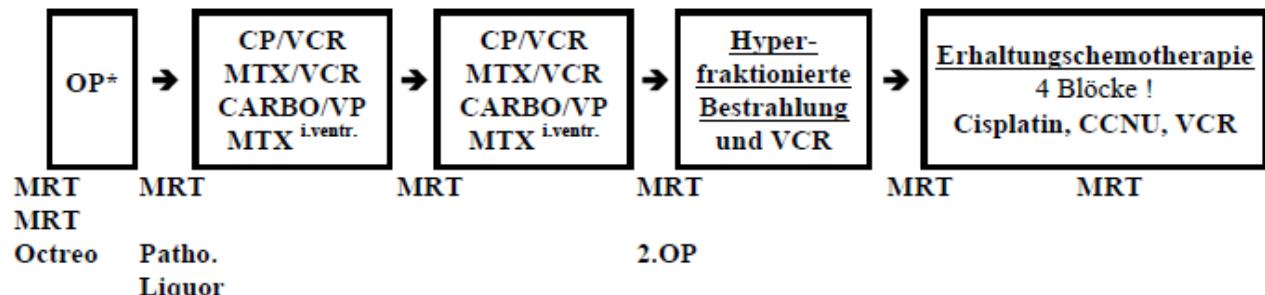
M-



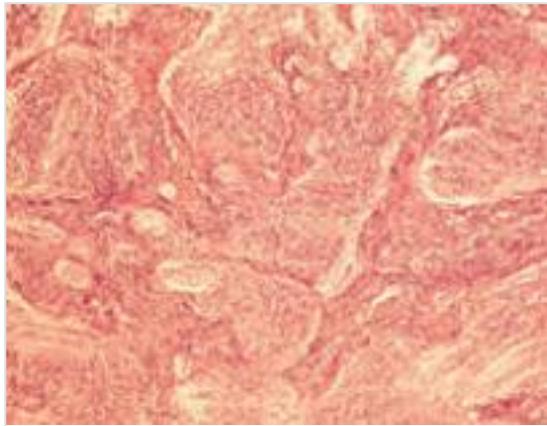
optional (wie bisher):
bei CR / >2/3 PR nach 1.SKK-Zyklus

**HDCT
(CARBO/VP/
THIOTEPAN)**

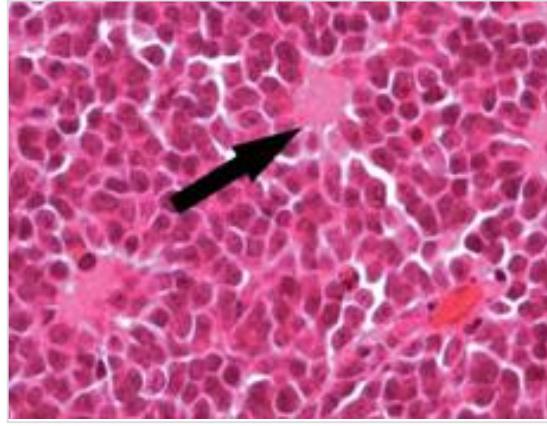
M+



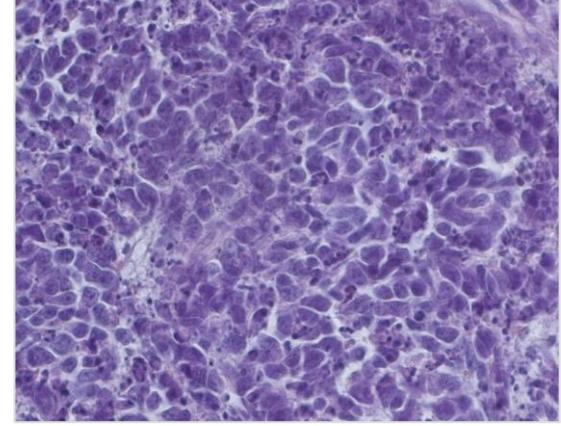
Current status of stratification (2)



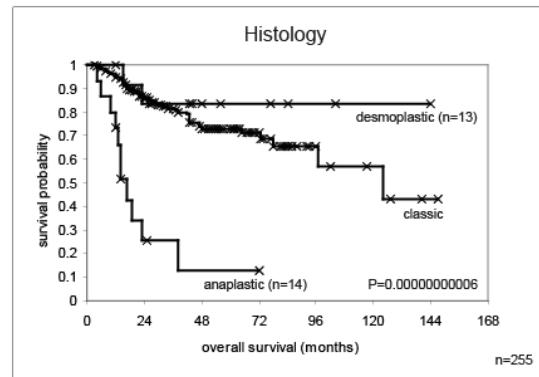
Desmoplastic
& extensive nodular (5-15%)



classic (70-90%)



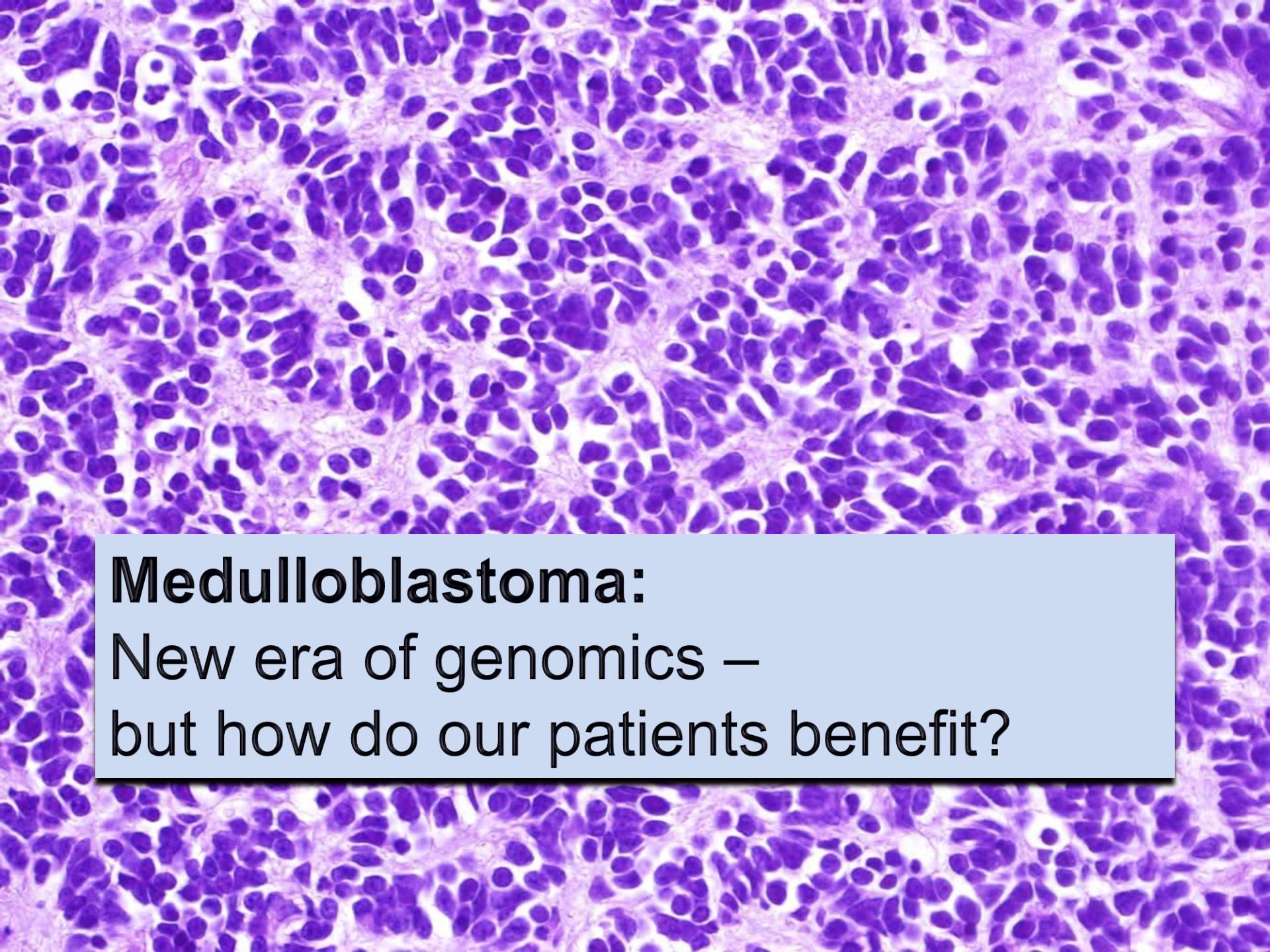
Large cell
& anaplastic (5-15%)



→ Unsatisfactory!

Current molecular classification of MB

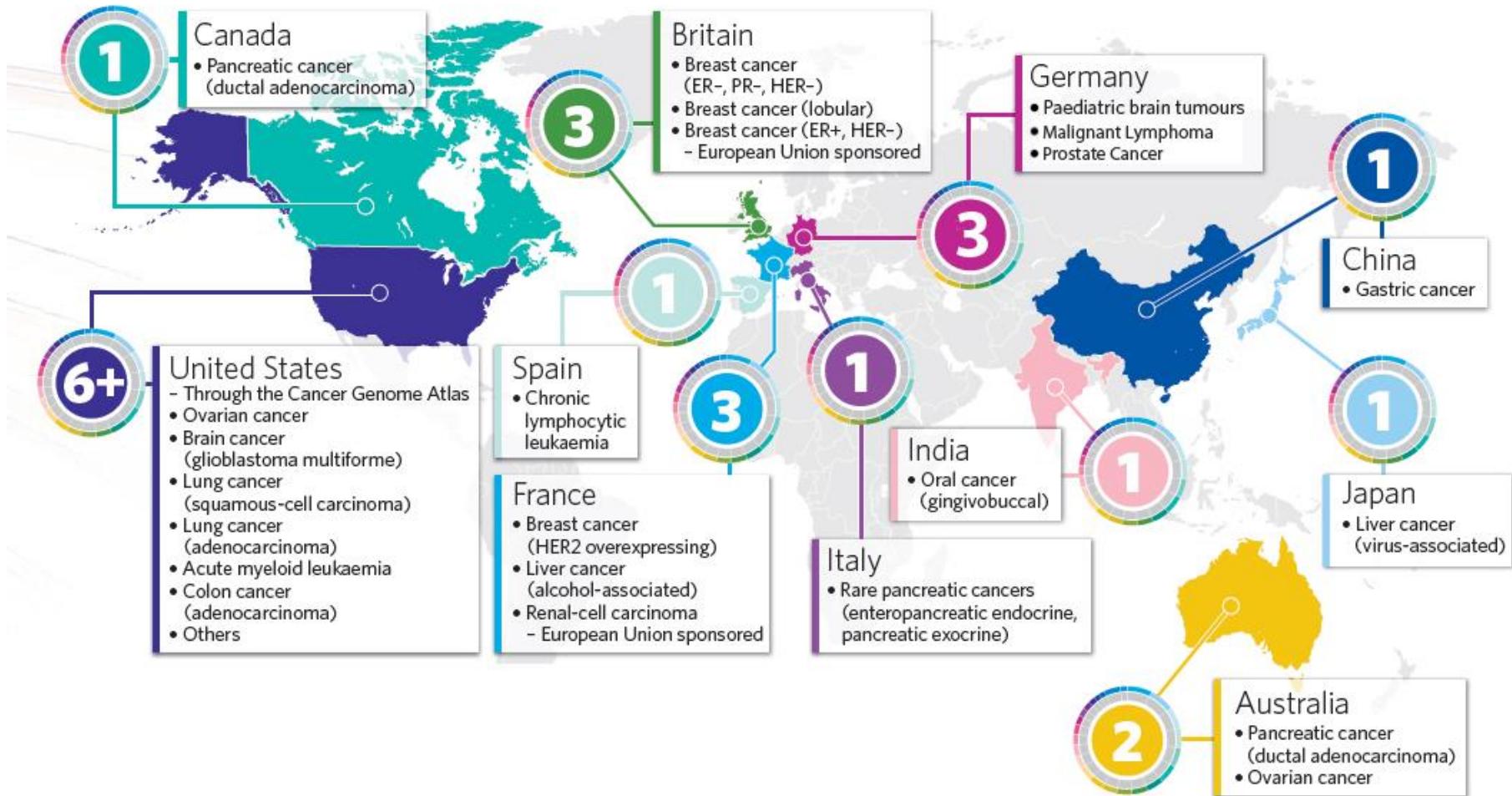
Subgroup	WNT	SHH	Group 3	Group 4
Gender ratio				
Age distribution				
Histology	Classic; very rare LCA	Classic > Nodular > LCA > MBEN	Classic > LCA	Classic; rarely LCA
Metastasis @ Dx	~5-10%	~15-20%	~40-45%	~35-40%
Patient Survival	~95% OS	~75% OS	~50% OS	~75% OS
Proposed Cell of origin	Lower rhombic lip progenitor cells	CGNPs of the EGL and cochlear nucleus; neural stem cells of SVZ	Prominin1(+), lineage(-) neural stem cells; CGNPs of the EGL	Unknown



Medulloblastoma:

New era of genomics – but how do our patients benefit?

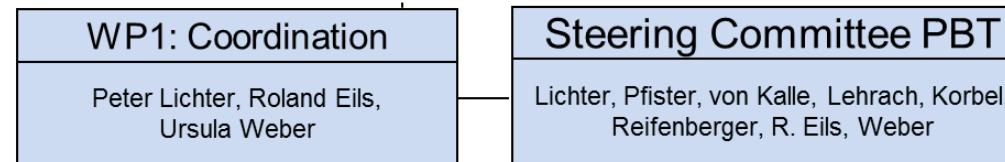
The ICGC Landscape



The ICGC PedBrain consortium



Sequencing
300 Medulloblastoma
300 Pilocytic Astrocytoma



WP 2 Banking	WP 3 Qual. Cont.	WP 4 Isolation	WP 5 Gen. Seq.	WP 6 PEM	WP 7 Methylome	WP 8 Transcript.	WP 9 miRNA	WP 10 Bioinform.	WP 11 Data Man.
Korshunov Witt Pfister Taylor	Reifenberger	von Kalle Schmidt	Pfister Licher	Korbel	Radlwimmer	Yaspo Lehrach	Landgraf Borkhardt Reifenberger	Brors Zapata R. Eils	R. Eils J. Eils Lawerenz

dkfz.

RUPRECHT-KARLS-UNIVERSITÄT HEIDELBERG EXZELLENZUNIVERSITÄT

HEINRICH HEINE UNIVERSITÄT DÜSSELDORF

MPI Berlin

SickKids FOUNDATION

NCT

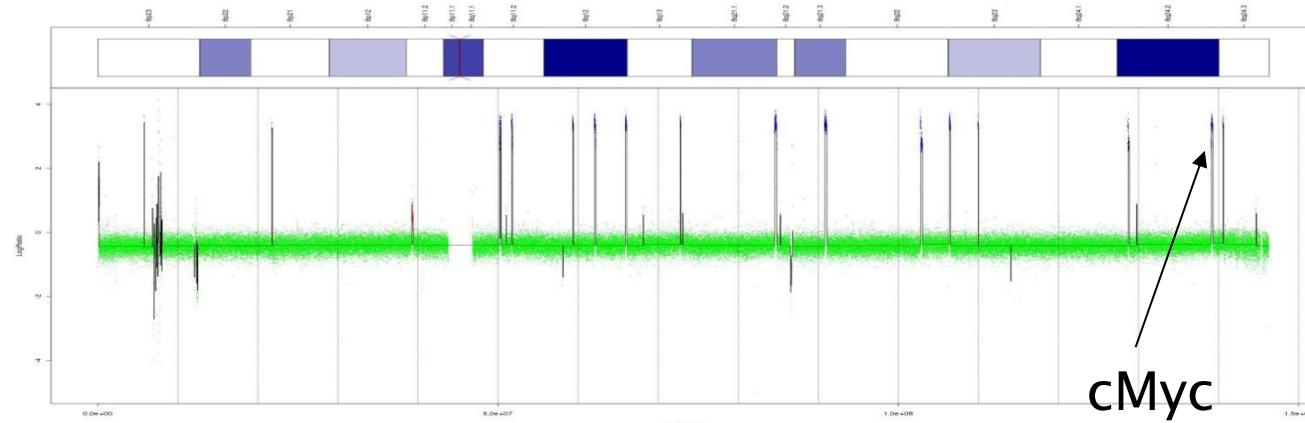
EMBL

dkfz.

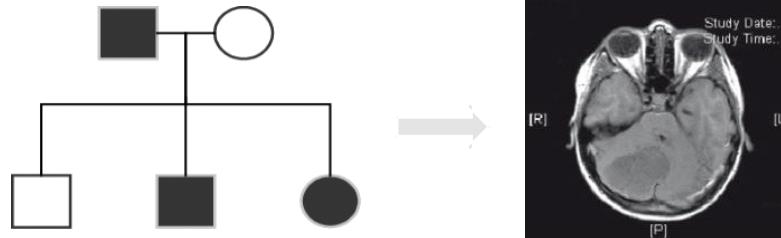
ICGC Pilot study: Li-Fraumeni Syndrome



Chromosome 8

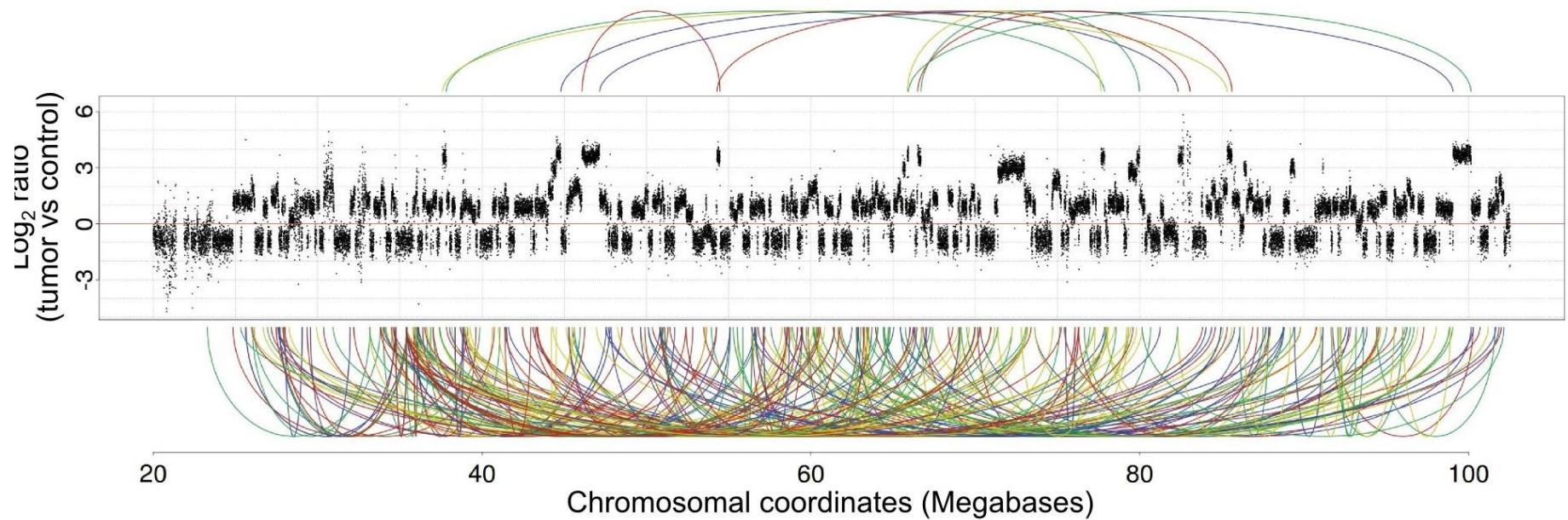


First sequenced LFS-MB



Further unexpected results...

Chromosome 15

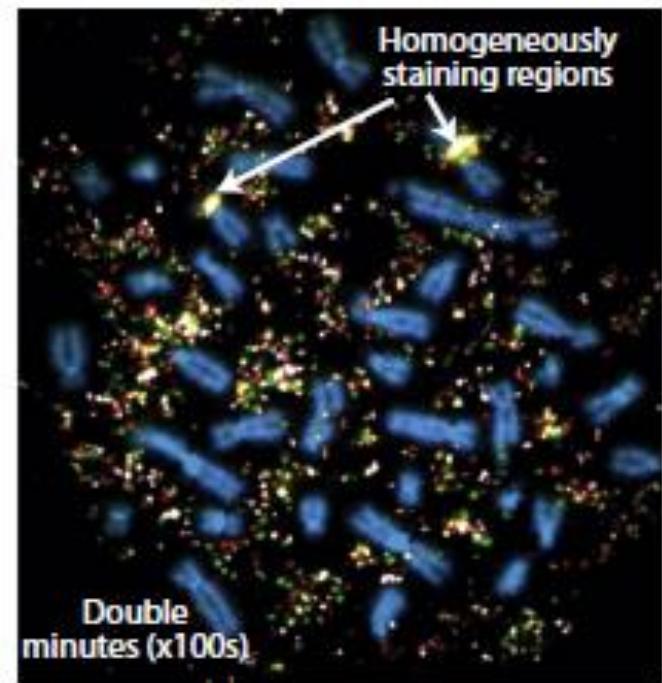
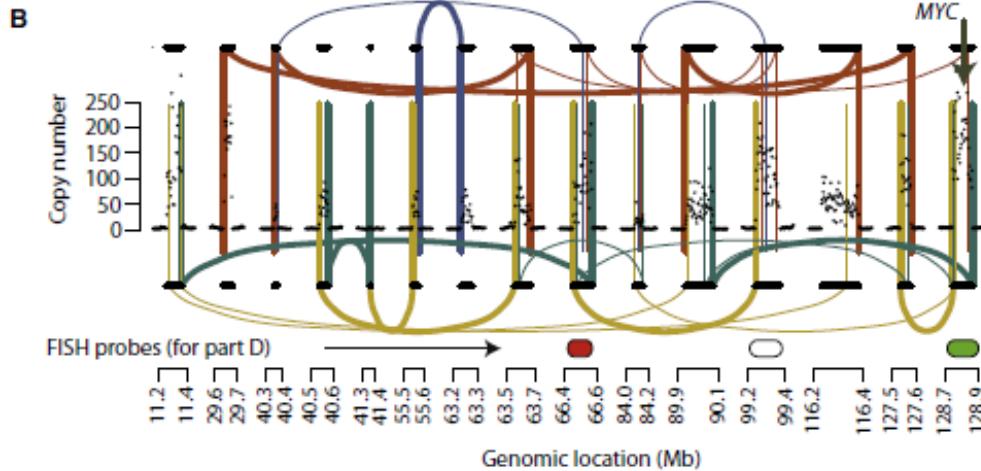


Chromothripsis

Chromos = chromosome + Thripsy = fall apart into pieces

Massive Genomic Rearrangement Acquired in a Single Catastrophic Event during Cancer Development

Philip J. Stephens,¹ Chris D. Greenman,¹ Beiyuan Fu,¹ Fengtang Yang,¹ Graham R. Bignell,¹ Laura J. Mudie,¹ Erin D. Pleasance,¹ King Wai Lau,¹ David Bearle,¹ Lucy A. Stebbings,¹ Stuart McLaren,¹ Meng-Lay Lin,¹ David J. McBride,¹ Ignacio Varela,¹ Serena Nik-Zainal,¹ Catherine Leroy,¹ Mingming Jia,¹ Andrew Menzies,¹ Adam P. Butler,¹ Jon W. Teague,¹ Michael A. Quail,¹ John Burton,¹ Harold Swerdlow,¹ Nigel P. Carter,¹ Laura A. Morsberger,² Christine Iacobuzio-Donahue,² George A. Follows,³ Anthony R. Green,^{3,4} Adrienne M. Flanagan,^{5,6} Michael R. Stratton,^{1,7} P. Andrew Futreal,¹ and Peter J. Campbell^{1,3,4,*}

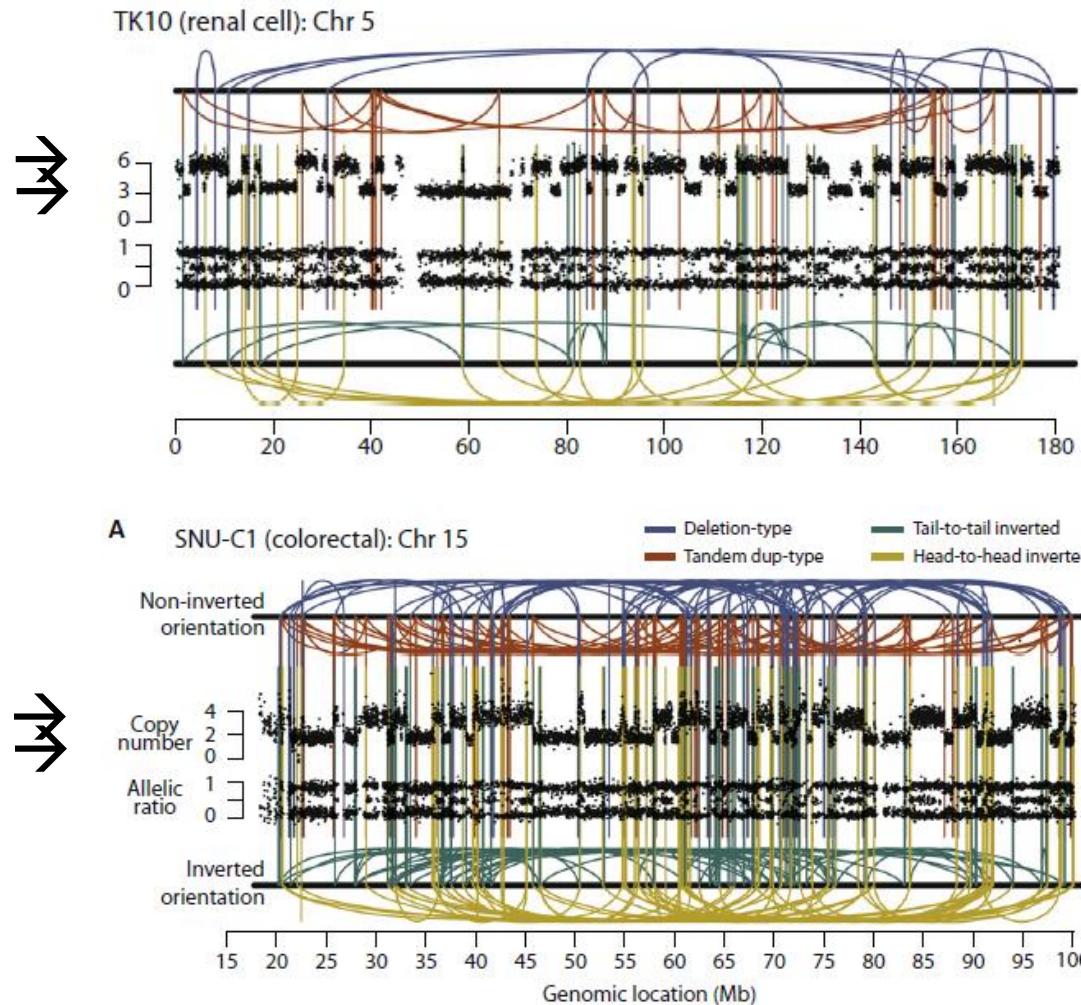


Cell 144, 27-40, January 7, 2011

→ 2-3% of all tumors

1. Pattern: Co-amplification of oncogenes

2. Pattern: „Shattering“



= Alternation between two copy-number states

Textbook: Multiple-Hit Model



Progressive rearrangements model

Germline



Tandem duplication CDEF



Inversion EFGH



Deletion EI



Tandem duplication BC



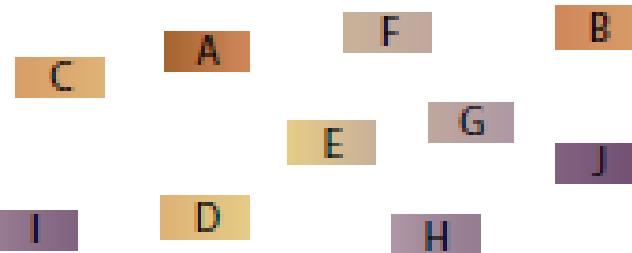
Alternative model: single catastrophic event



Catastrophe model



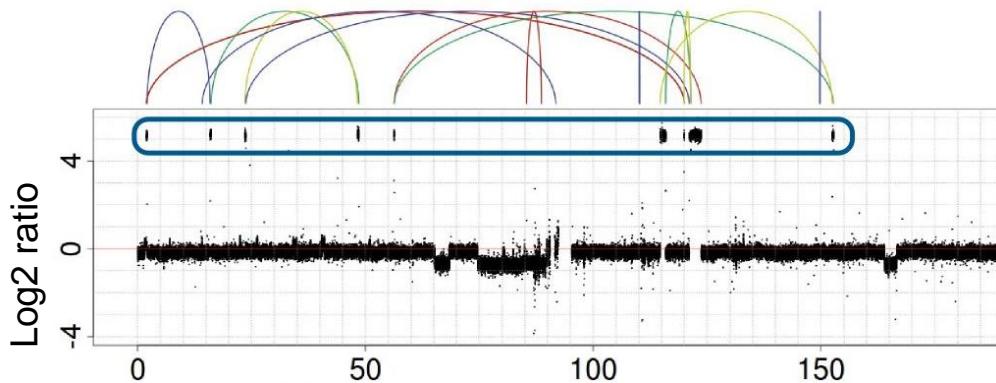
Catastrophic chromosome breakage



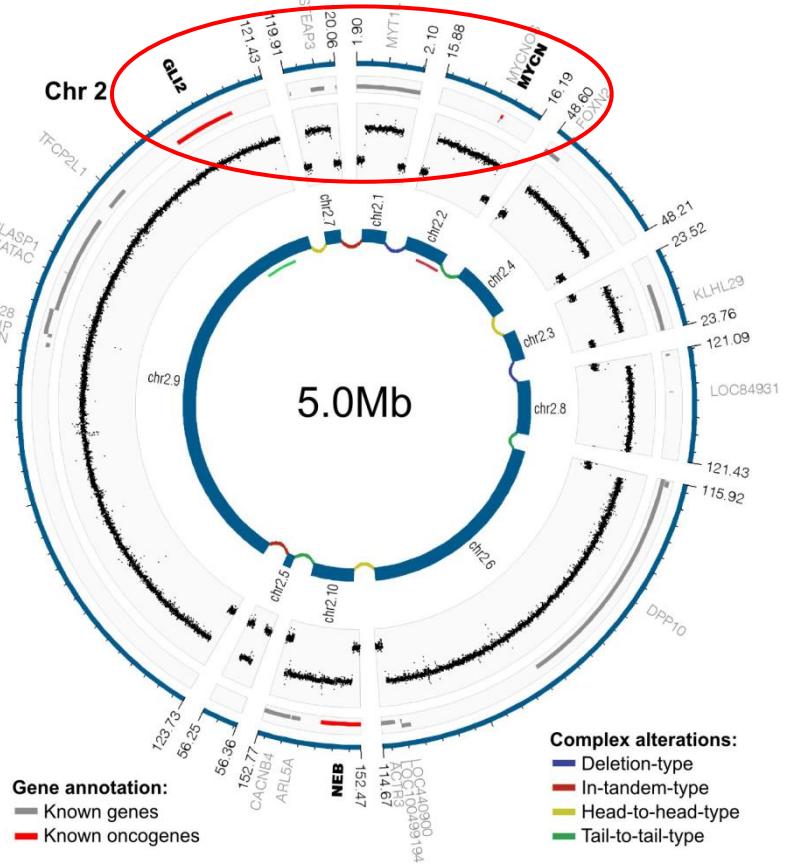
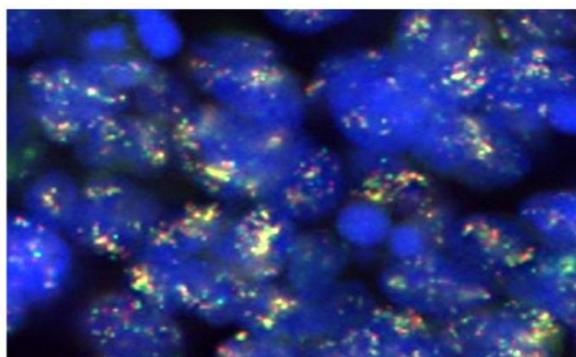
Non-homologous end-joining



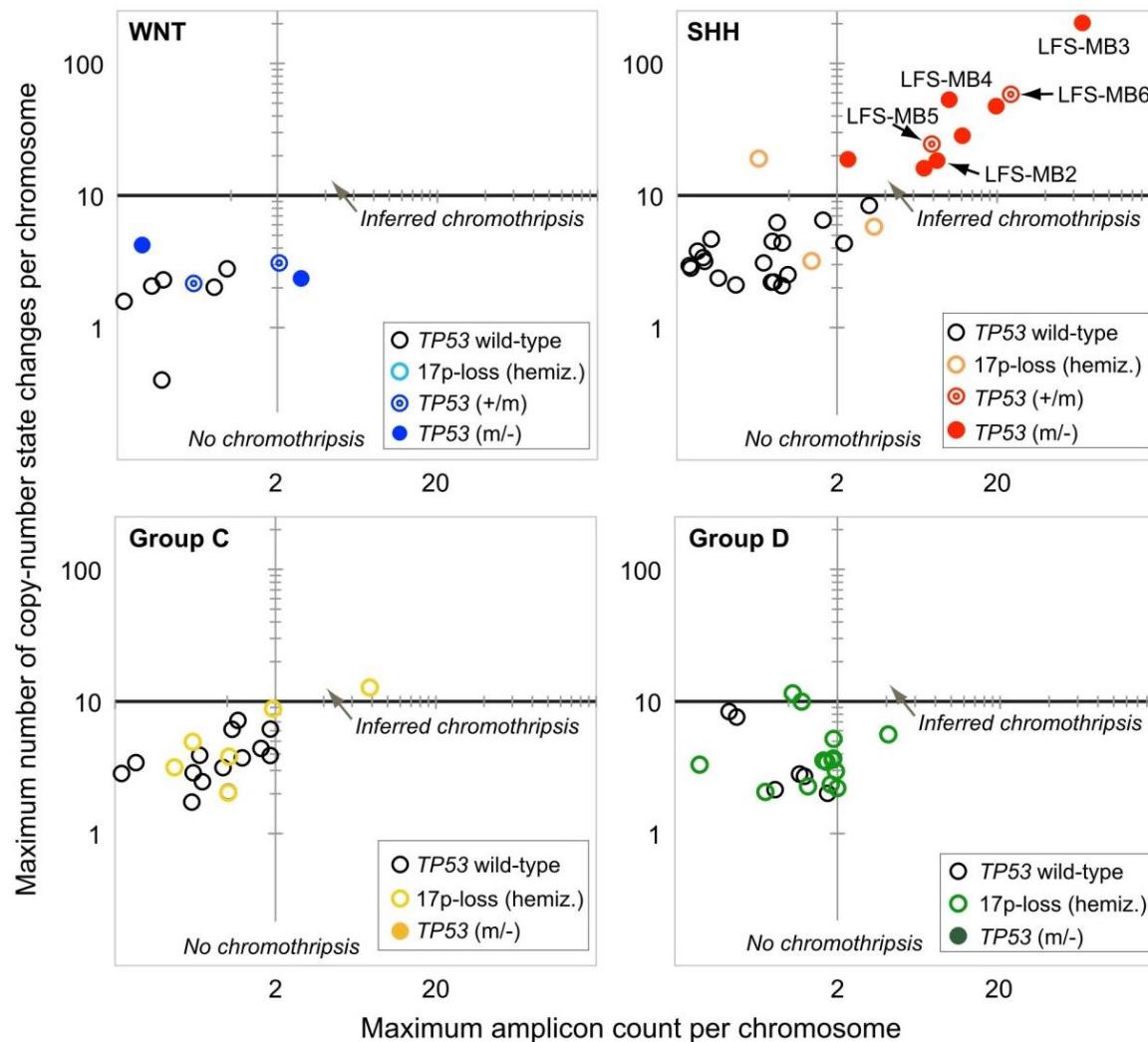
Co-amplification of SHH oncogenes in MB



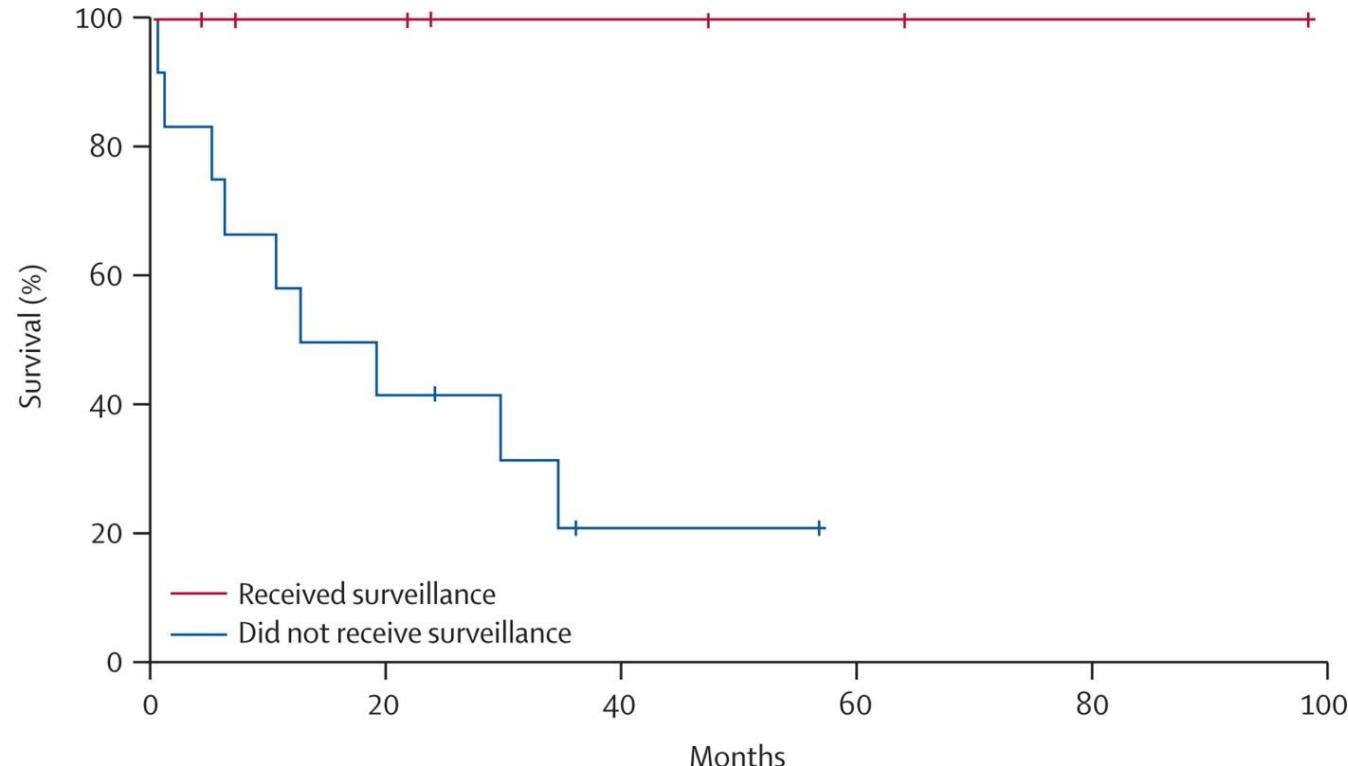
Two known MB oncogenes are getting co-amplified in one mini-chromosome



LFS – more common than anticipated!



Clinical relevance: Surveillance



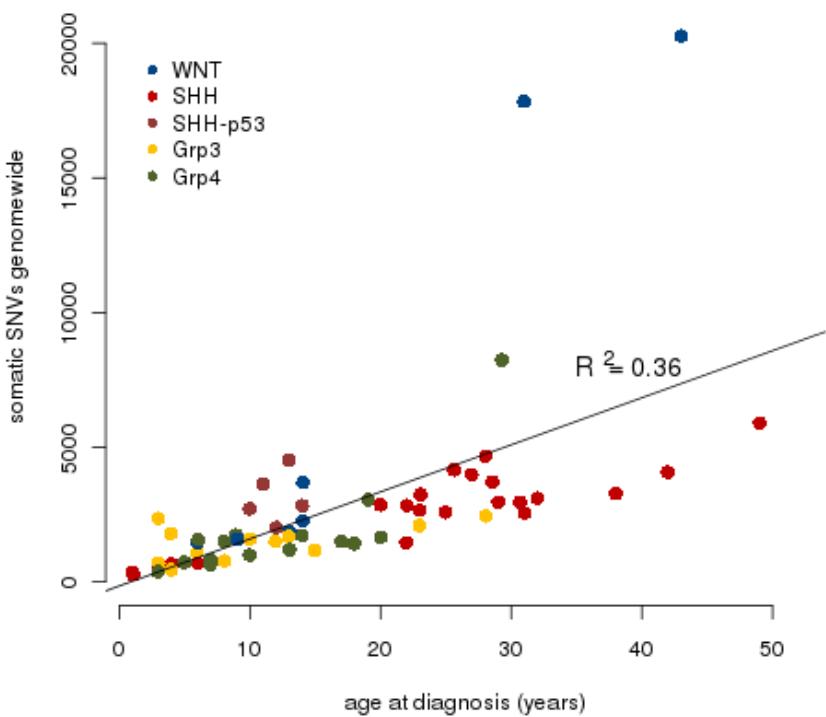
Number at risk

Received surveillance	10	8	5	3	1	0
Did not receive surveillance	12	5	1	0		

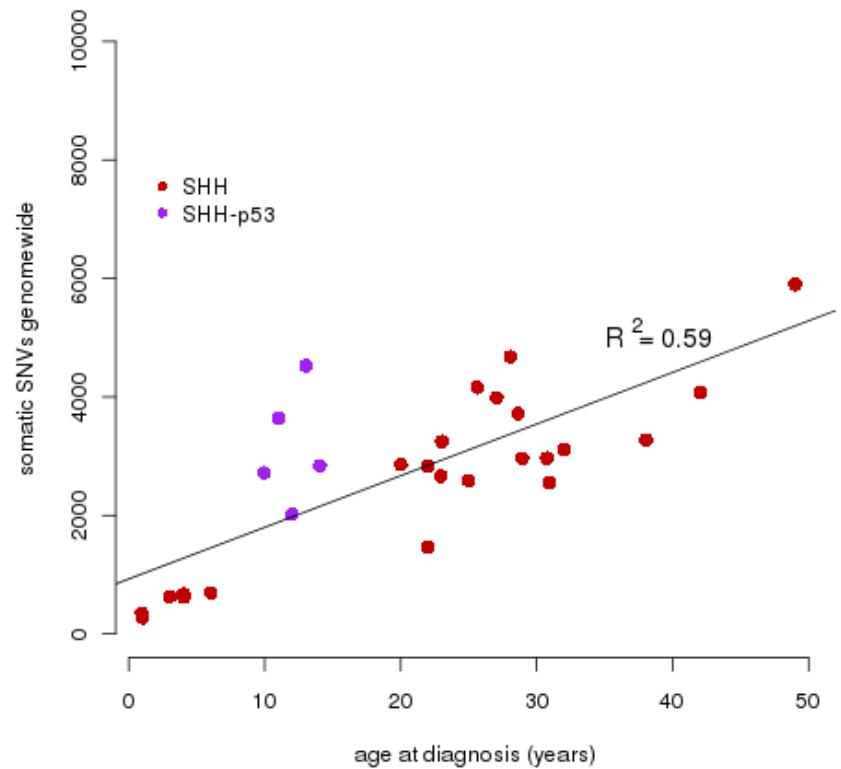
Sporadic MB: Mutation rate ~ age



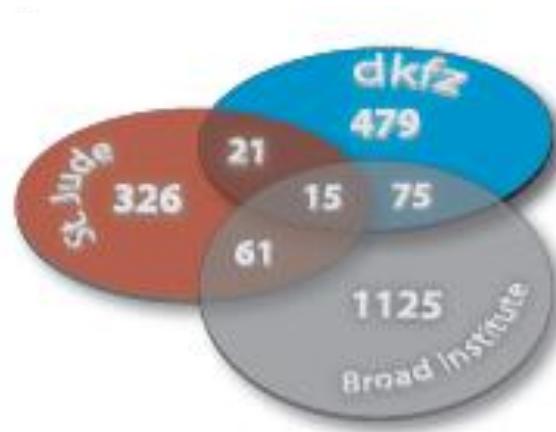
All medulloblastomas



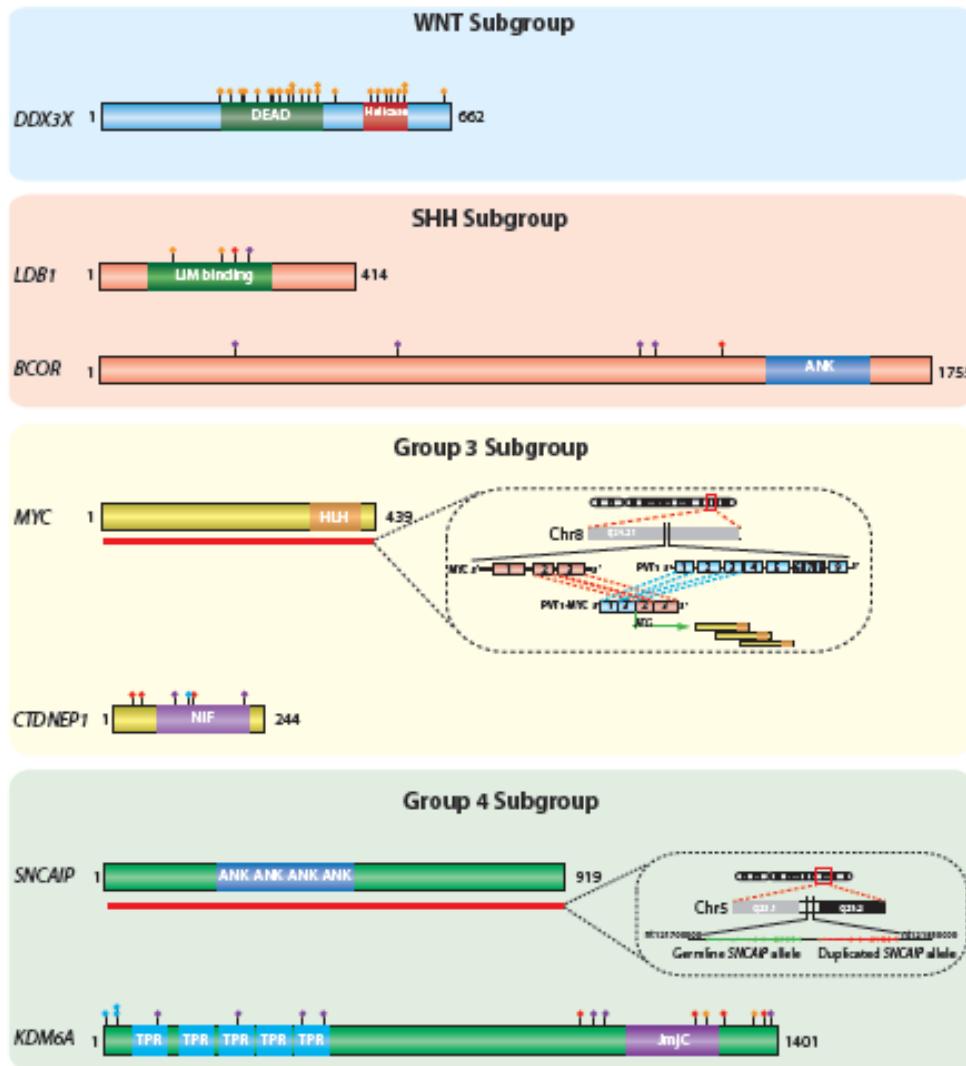
Only SHH medulloblastomas



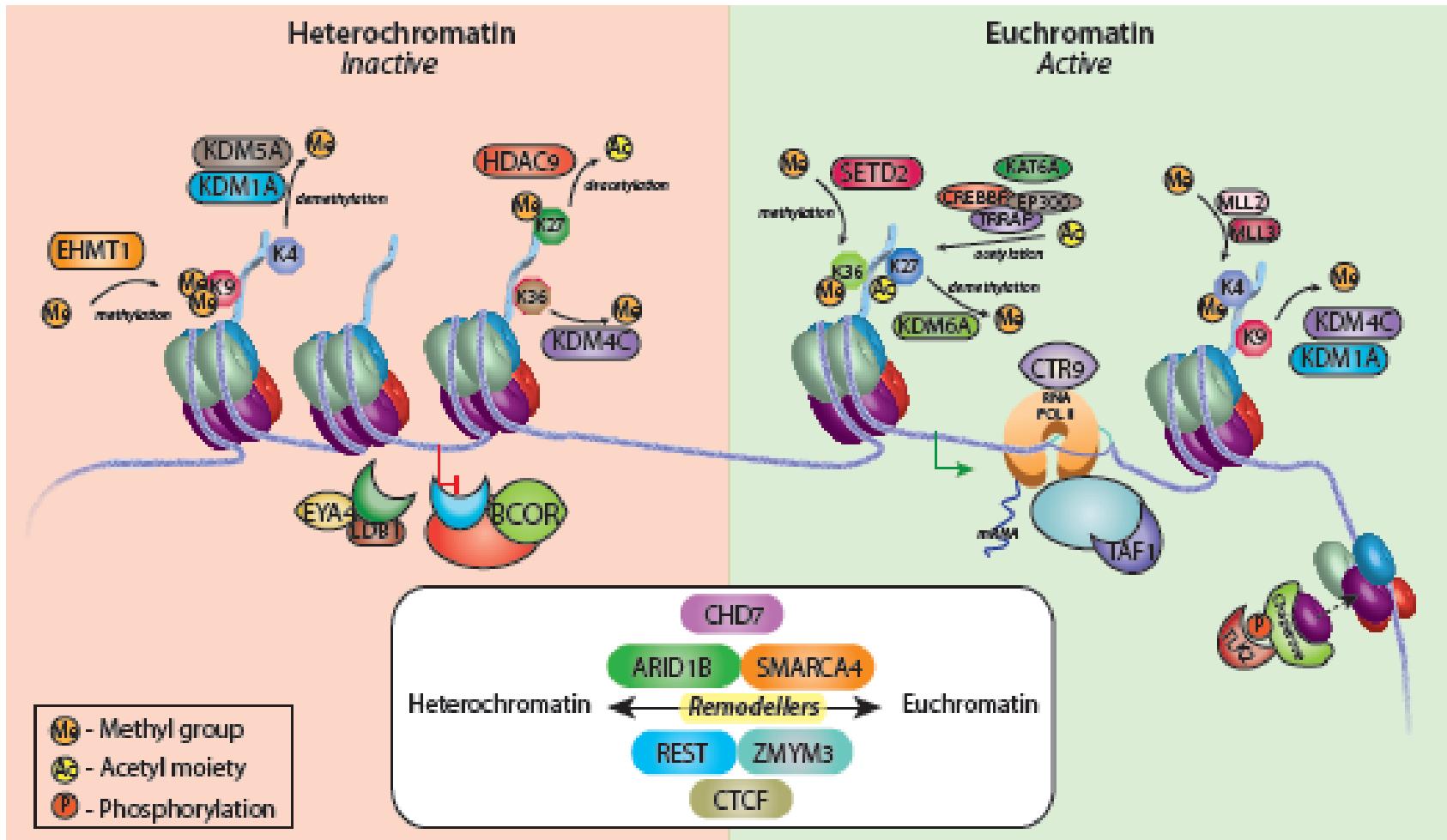
Novel recurrent mutations in MB



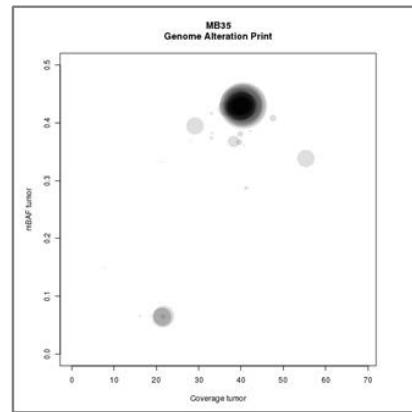
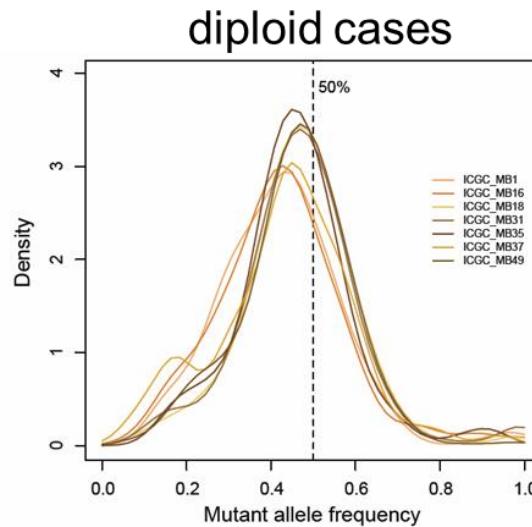
n=189



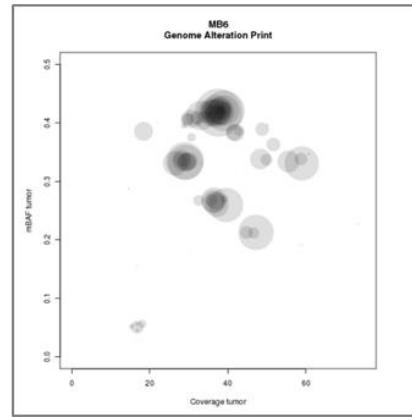
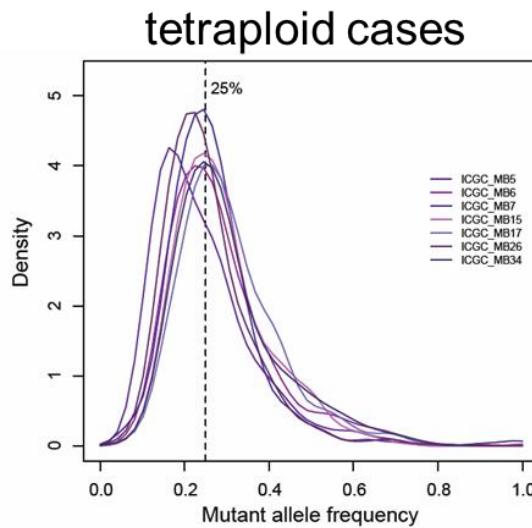
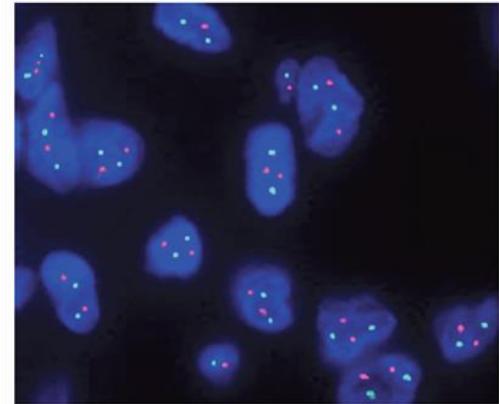
Common topic: Chromatin modifiers



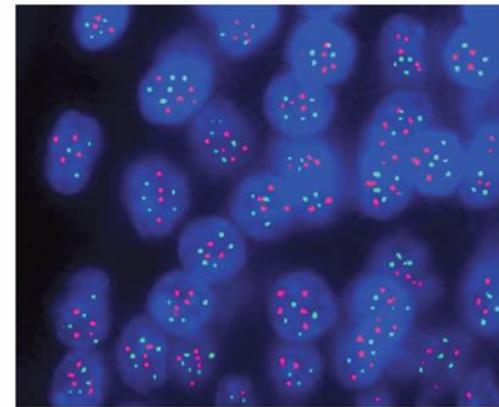
Tetraploidy highly enriched in groups 3 & 4



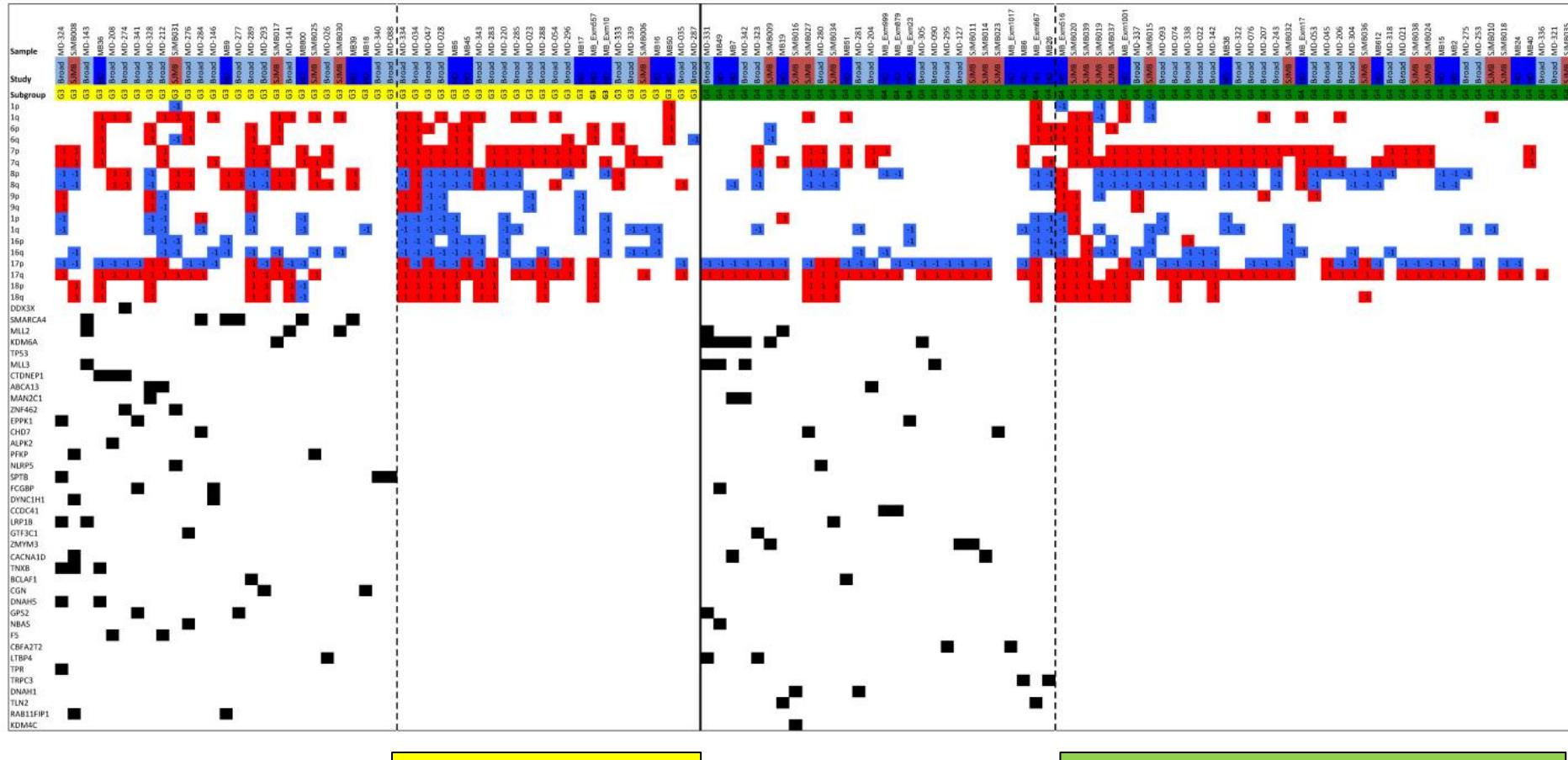
MB37 (diploid)



MB15 (tetraploid)



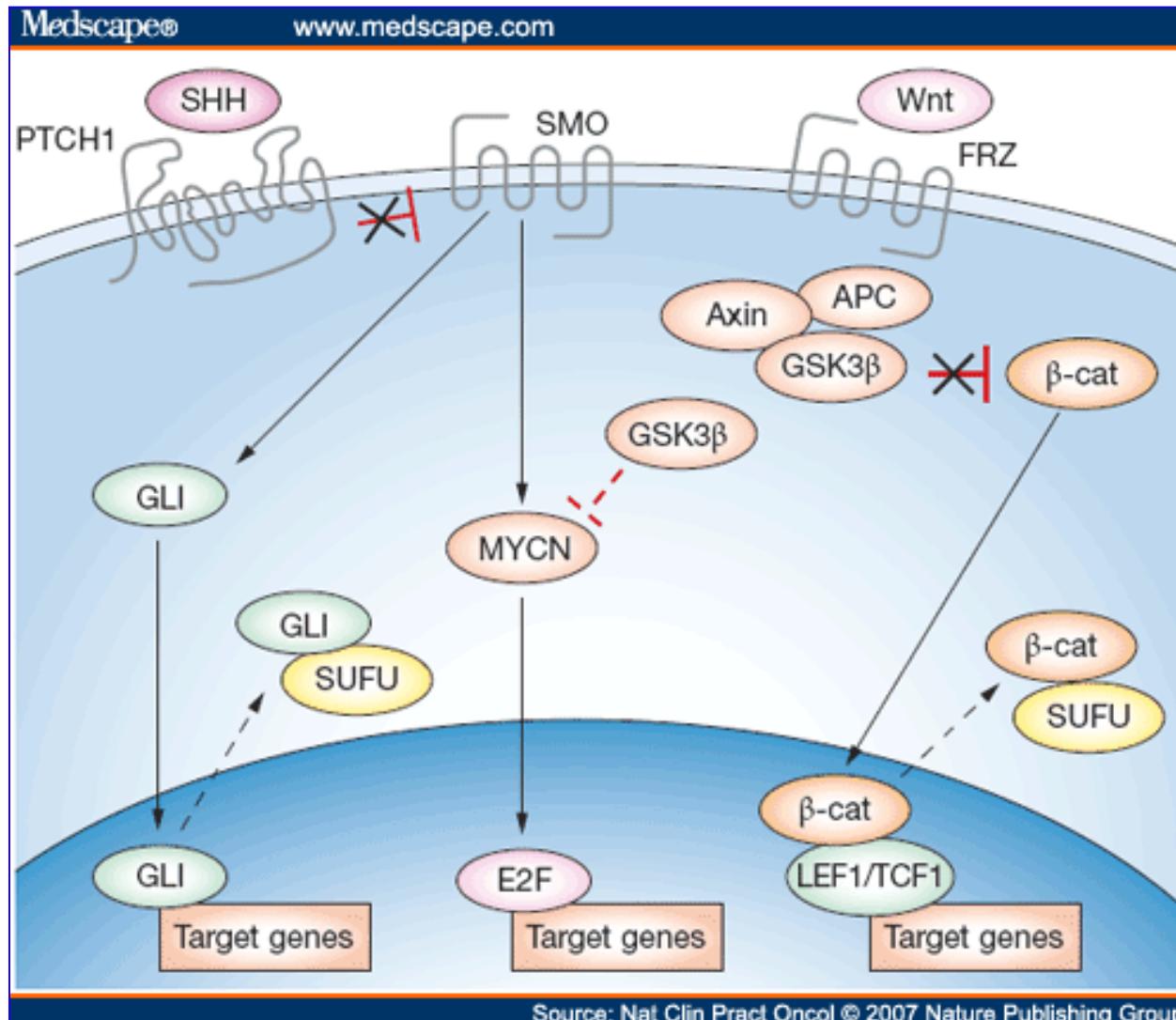
Genetic patterns in groups 3 & 4



Copy-umber driven?

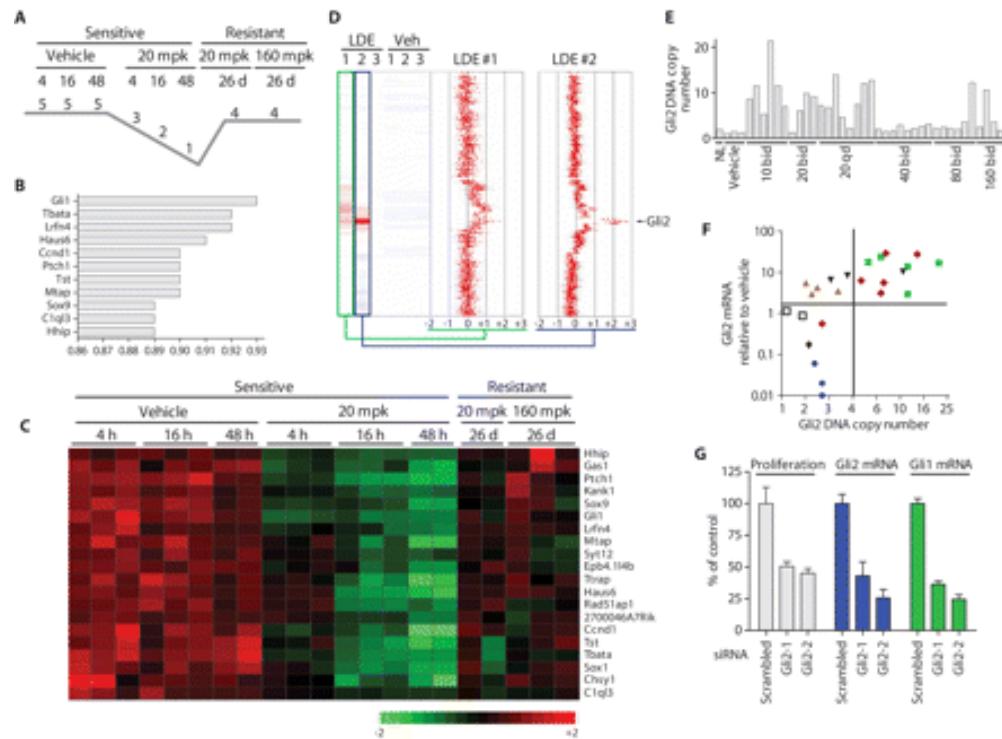
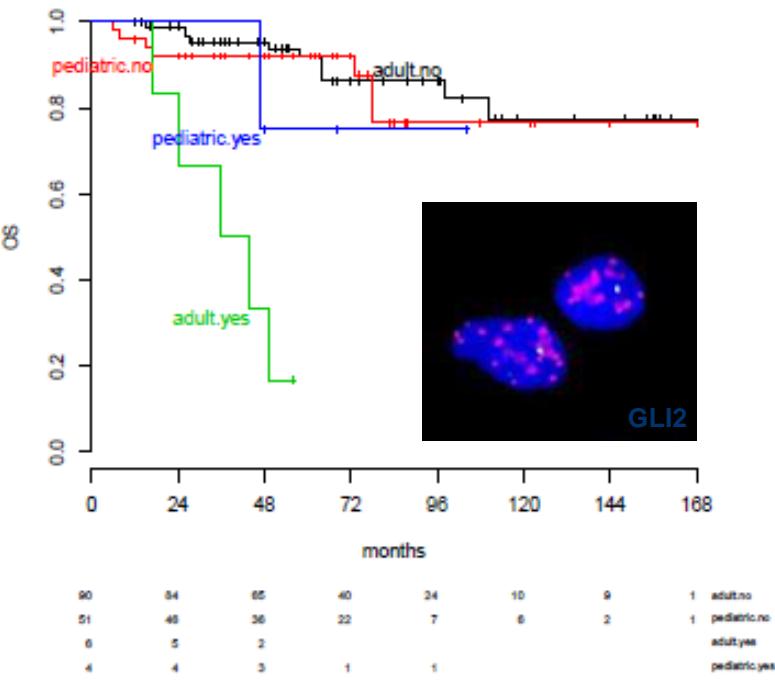
Copy-umber driven?

Outlook MB: SHH inhibitors in the clinic



Primary and secondary resistance

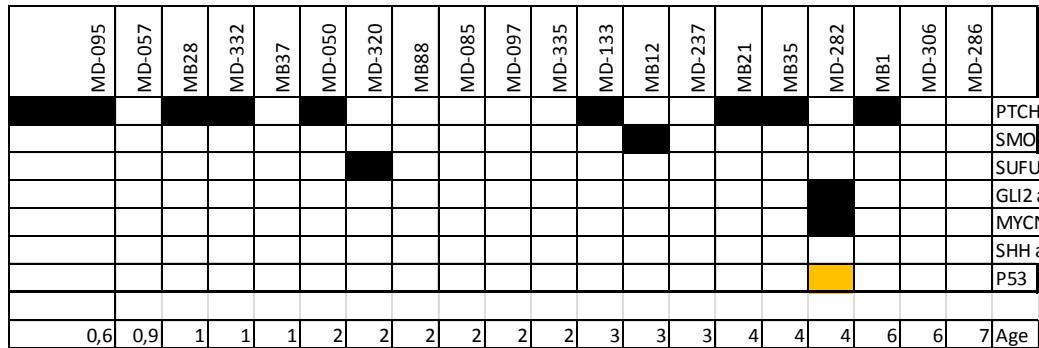
GLI2 Amplification



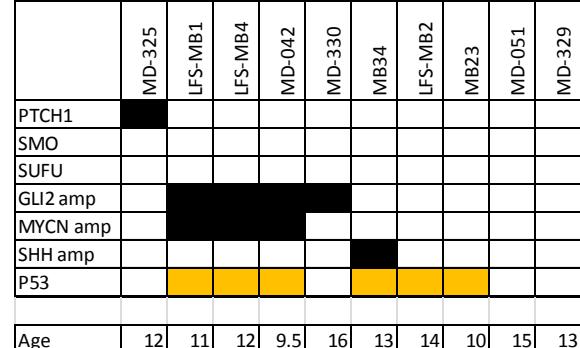
SMO inhibitors: Who is going to benefit?



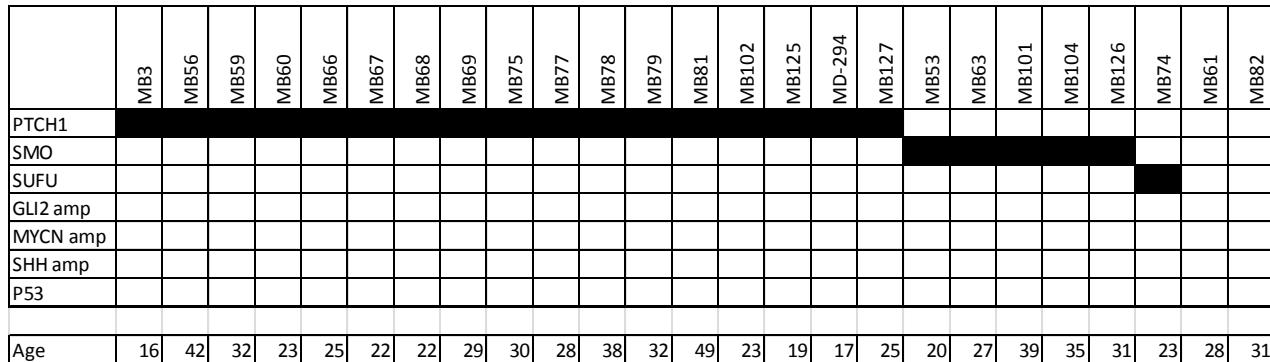
Young children (< 8)



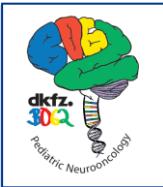
Older children (8 – 16)



Adults (>16)



Conclusions



- MB is not a single disease – even within subgroups it is very heterogeneous
- Germline predisposition (e.g., LFS) seems to play a much more important role than previously anticipated. LFS is a clinically actionable readout.
- Chromatin modifications and tetraploidy seem the most promising novel avenues for therapeutic interference
- Response to SHH inhibitors in the clinic may be predicted within the SHH subgroup

Outlook: CEFALO germline study



Gene	VAAST-Rank	P-Value	Score	# Samples	Subgroup
<i>PMS2</i>	9	2.61E-120	37.66	3	SHH
<i>MYOD1</i>	100	0.03	1.03	3	SHH
<i>SMARCA4</i>	117	0.04	1.62	1	SHH
<i>TP53</i>	240	0.12	10.36	3	SHH
<i>SUFU</i>	460	0.28	14.81	3	SHH
<i>MLH1</i>	599	0.38	0.26	1	SHH
<i>CHEK2</i>	729	0.55	1.52	1	SHH
<i>BCORL1</i>	748	0.58	2.42	2	SHH
<hr/>					
<i>PTCH1</i>	274	0.14	11.83	8	All subgroups
<i>APC</i>	435	0.26	5.82	5	All subgroups
<i>ATM</i>	145	0.06	20.69	7	All subgroups
<i>NBS1 (NBN)</i>	9139	1	0	3	SHH/Grp3/Grp4
<i>PALB2</i>	591	0.38	7.50	8	SHH/Grp3/Grp4
<i>CBP (CREBBP)</i>	1448	1	0	5	SHH/Grp3/Grp4
<i>RB1</i>	13160	1	0	1	Grp 3

*Data was derived from ICGC PedBrain and Broad sequencing projects (n~220)

Molecular Diagnostics of Pediatric Malignancies



DKTK: German Consortium for translational cancer research

Berlin	Whole exome sequencing of rec-ALL
Essen/Düsseldorf	Whole exome sequencing of retinoblastoma, AML and NHL
Frankfurt/Mainz	Whole exome sequencing of soft tissue sarcoma
Freiburg	Whole exome sequencing of MDS/JMML, Ewing Sarcoma
Heidelberg	Whole exome sequencing of T-ALL, brain tumors, neuroblastoma bioinformatic data analysis; data storage
München	Whole exome sequencing of osteosarcoma, hepatoblastoma
Tübingen	Whole exome sequencing of cALL

Principal investigators: Peter Lichter
Stefan Pfister

Pediatric Neurooncology



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Andreas E. Kulozik

Korbel Lab (EMBL)

Taylor Lab (Toronto)

Pomeroy/Cho/Meyerson (Boston)

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Jabado Lab (Montreal)

Collaborators

Andrey Korshunov (Heidelberg)

Andreas v. Deimling (Heidelberg)

Olaf Witt, Till Milde (CCU, Heidelberg)

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Pablo Landgraf & Arnd Borkhardt (Düsseldorf)

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Wolfgang Roggendorf (Würzburg)

Arend Koch (Berlin)

Volkmar Hans (Bielefeld)

Peter Collins (Cambridge)



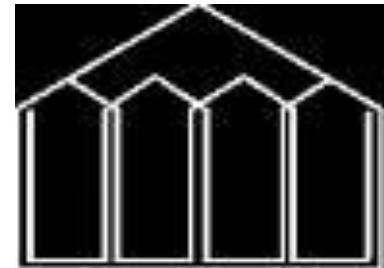
Funding



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der Wissenschaften



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Heidelberg/Mannheim



Deutsche Krebshilfe



Landesstiftung BW



BMBF