XenTech patient-derived xenograft (PDX) panels: a clinically relevant platform for drug efficacy , target validation and predictive biomarker discovery studies.

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INTRODUCTION

- Despite considerable efforts in understanding the biology and genetics of cancer, most currently available treatments fail to achieve tumor eradication in the majority of patients. Key to more effective therapies is adequate disease classification and subsequent patient stratification. In addition, it is important to understand the mechanisms of drug-response or resistance and identify novel targets amenable to therapeutic intervention. It is increasingly recognized that at the preclinical stage, testing therapeutic strategies and validating target relevance in more predictive models closely mimicking clinical disease such as patient derived xenografts (PDXs), may translate into improved clinical efficacy and lower rate of drug attrition.

We therefore developed a large collection of PDX models capturing the genetic and drug response diversity of clinical disease.

STRATEGY AND AIMS

- XenTech maintains a collection of over 120 running PDX models that are deeply characterized and readily available for in vivo preclinical assays. PDX models were established by grafting post-surgery human tumor fragments in the interscapular region of immunodeficient mice. Our preclinical platform has been developed to provide a reliable surrogate of patient cohorts and address several aims:

1. Evaluate tumor response to treatment. PDXs can be subjected to parallel evaluation of tumor response to various treatment protocols. Drug-response profile is linked to tumor histotype and molecular features in order to identify predictive markers of drug response to assist treatment choice.
2. Development of bioluminescent metastatic models to study the mechanisms of tumor invasion and to test anti-metastatic therapy.
3. Assess treatment-driven residual tumor eradication. The ability of a treatment to induce complete tumor response is assessed by monitoring tumor regression over a long period. Most tumors, despite complete macroscopic regression, are still present as latent microscopic nodular islands that may give rise to tumor recurrence. Molecular characterization of tumor foci responsible for tumor relapse may be performed to identify genes/pathways involved in residual tumor cell survival, which may provide new diagnostic and/or therapeutic targets for designing novel adjuvant treatment strategies.
4. Constitution of preclinical panels of rare malignancies to obtain phase II-like tumor cohorts. Development of new therapies for rare tumors is rendered difficult by the unavailability of patient cohorts wide enough to set up robust clinical trials. To assist the clinical need, these panels would allow the evaluation of new and more efficient therapies.
5. Development of a mid-throughput in vitro assay system for antitumor drug activity profiling from PDXs. This ex vivo model system offers a useful platform for drug activity profiling, complementary to classical screening on tumor cell lines. Moreover, this primary tumor cell culture system is useful for rapid screening of tumor drug response levels and selection of specific PDXs for in vivo assays.

RESULTS

Xentech PDX collection

CANCER TYPE	SUBTYPES	QUALIFIED MODELS	MODELS IN DEVELOPMENT	CODING GENE EXPRESSION MICROARRAYY (Affymetrix U133 2.0 plus)	aCGH (Affymetrix SNP 6.0)	miRNA ARRAY(Exiqon)	EXON SEQUENCING (74 genes frequently mutated in cancer)
BREAST	TNBC (70%),ER+, PR+, HER2+	37	2	37	37	28	35
COLORECTAL		21		21	21	21	21
LUNG (NSCLC)		15	7	15	15	-	11
LUNG (SCLC)		7		7	7	-	7
PROSTATE	Hormone-dependent + castration-resistant variant	2		2	2	-	2
BRAIN	Glioblastoma	7		7	7	-	7
PANCREAS		3		3	3	-	3
SKIN	Melanoma, Merkel cell carcinoma	6		6	6	-	2
OVARY		3		3	3	-	3
ENDOMETRIUM		4		4	4	-	3
KIDNEY	RCC (clear cells, papillary)	7	1	7	7	-	7
LIPOSARCOMA		1		1	1	-	1
PEDIATRIC LIVER TUMORS	Hepatoblastoma, Rhabdoid tumor		14	5	5	-	5

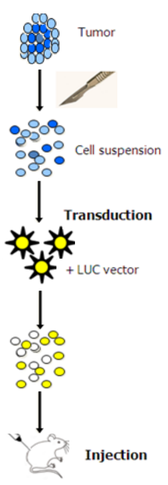
1. Tumor response to treatment

PDX code	PDX drug response								*In vitro* primary culture assay
	Capecitabine	Bevacizumab	Capecitabine + Bevacizumab	Cetuximab	Capecitabine + Oxaliplatine	Capecitabine + Oxaliplatine + Bevacizumab	Capecitabine + Oxaliplatine + Cetuximab	Capecitabine + Irinotecan	
TC01	NR	NR	NR	NR	NR	NR	NR	NR	+
TC07-FAL	R	NR	HR	R	R	HR	HR	HR	-
TC116-DES	R	R	HR	NR	HR	HR	HR	HR	-
TC118	R	NR	R	R	R	R	R	R	-
TC122A	R	R	HR	NR	R	HR	HR	HR	+
TC124B	NR	NR	R	NR	NR	R	NR	NR	-
TC302	R	NR	HR	NR	R	HR	HR	HR	-
TC303	NR	NR	R	NR	NR	R	R	R	-
TC305-BAU	R	R	HR	NR	HR	HR	HR	HR	-
TC306-BAU	HR	NR	HR	NR	HR	HR	HR	HR	-
TC307-BAU	HR	NR	HR	NR	HR	HR	HR	HR	-
TC308-BAU	NR	R	HR	R	R	HR	HR	HR	-
TC314-LEV	HR	NR	HR	R	R	HR	HR	HR	-
TC316-LBO	NR	NR	R	NR	NR	R	R	R	-
TC320-LBO	NR	NR	NR	NR	NR	NR	NR	NR	-
TC329-HUT	NR	NR	NR	NR	NR	NR	NR	R	-
TC336-SAR	NR	NR	R	NR	R	R	HR	HR	-
TC37	NR	NR	R	NR	NR	R	R	R	-
TC71	HR	NR	HR	NR	HR	HR	HR	HR	+
TC82	R	NR	HR	NR	R	HR	R	R	-
TCM001-HK	R	NR	HR	NR	R	HR	HR	HR	-

Legend:
- HR — High responder - T/C%<10%
- R — Responder 42%> T/C% >10%
- NR — Non-responder T/C% ≥ 42%

Eight-arm preclinical phase 2-like assay in a panel of 21 colon cancer PDXs with 5 different standards of care used as single agent or in combination.

2. Development of bioluminescent metastatic models

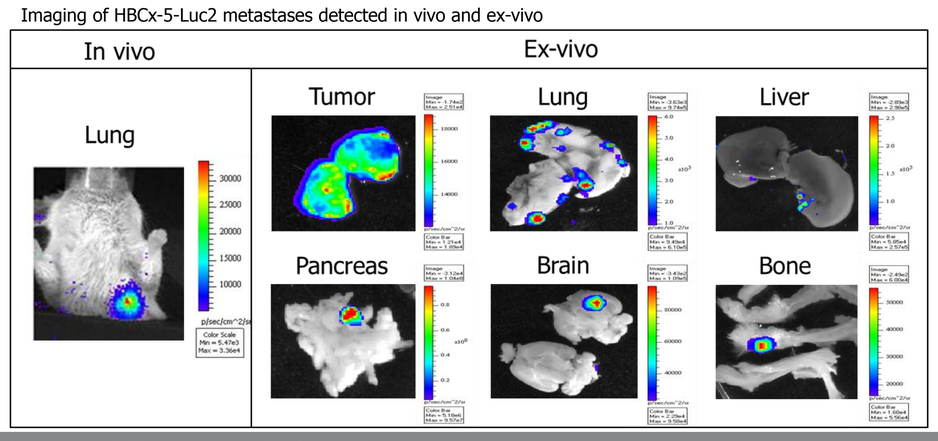


High efficiency lentiviral transduction was used to obtain near 100% luciferase-expressing tumor cells without in vitro selection.
This approach ensures that luciferase expressing models are phenotypically identical to the parent tumor.

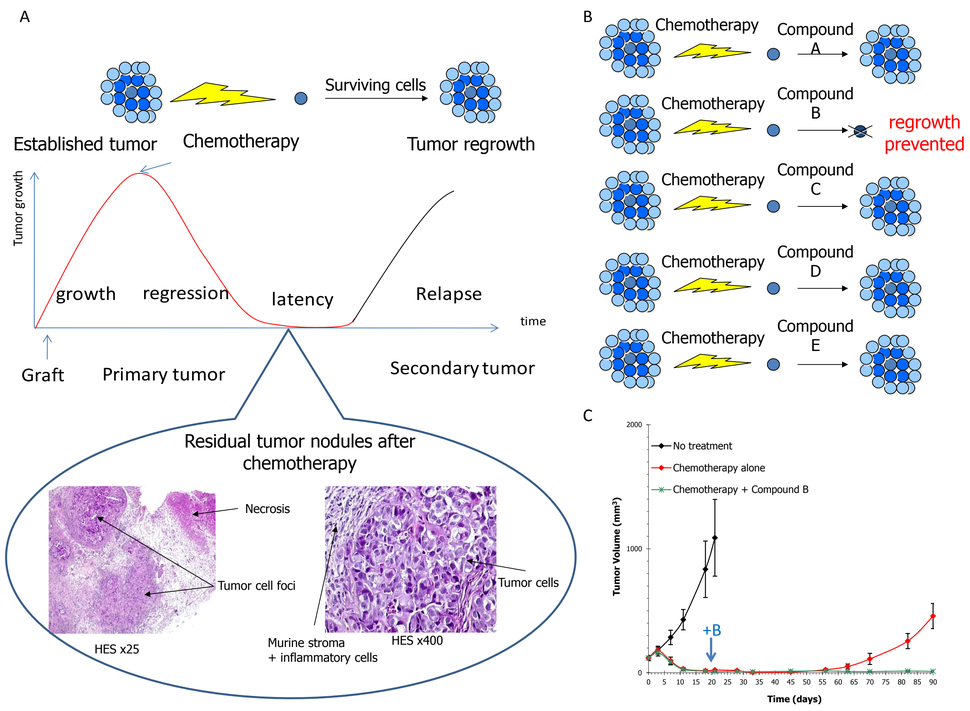
List of bioluminescent PDXs analyzed for metastatic spreading

PDX Model	Primary tumor resection	N of mice	Luciferase-positive metastasis at endpoint						
			mean latence (days)	In vivo	ex-vivo				
				Lung	Lung	Liver	Spleen	Pancreas	Brain	Bones
HBCx-12B-Luc3	No	10	50+/-12	10/10	4/4	1/4	0/4	0/4	2/4	0/4
HBCx-5-Luc2	No	17	83+/-40	16/17	17/17	6/12	1/13	8/13	11/13	10/13
HBCx-14-Luc1	Yes	16	75+/-8	8/16	10/16	NA	NA	NA	NA	NA
PAC120-Luc	No	6	65	1/5	4/5	NA	NA	NA	NA	NA
PANx-005-Luc1	No	11	42	6/11	11/11	0/11	1/11	0/11	0/11	1/11

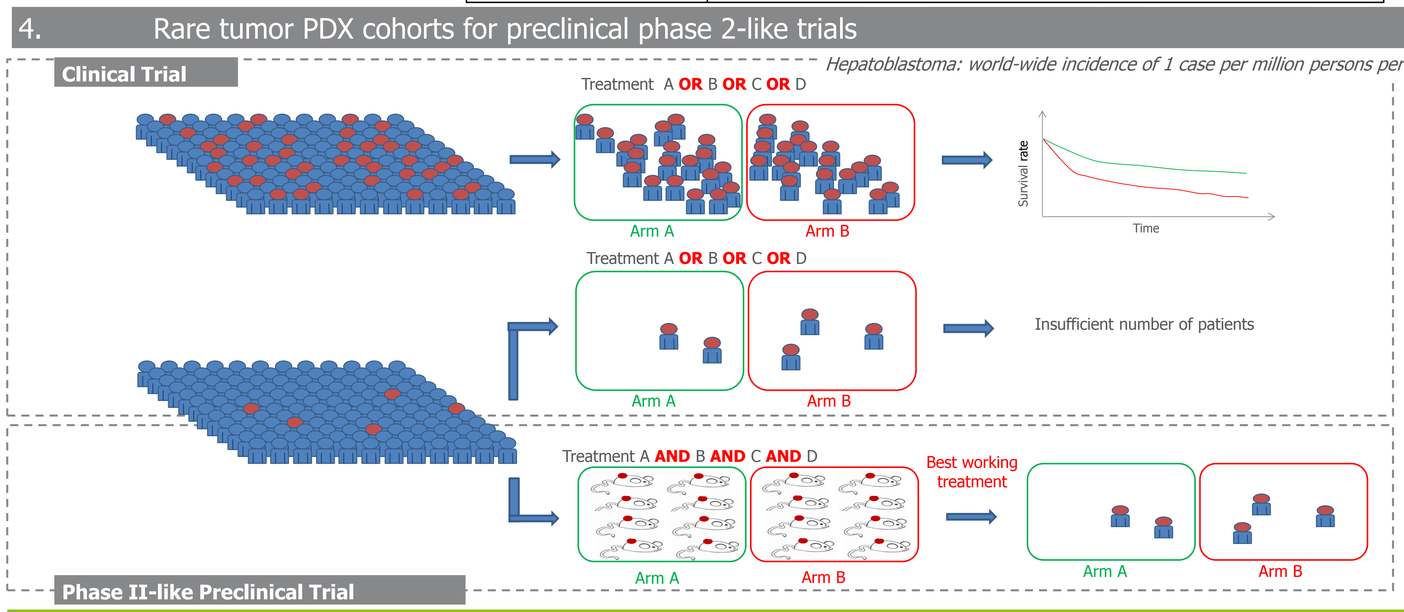
Imaging of HBCx-5-Luc2 metastases detected in vivo and ex-vivo



3. Treatment-driven residual tumor eradication



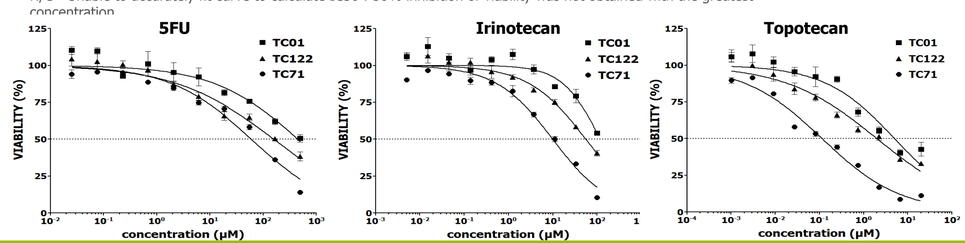
4. Rare tumor PDX cohorts for preclinical phase 2-like trials



5. PDX-derived mid-throughput in vitro cytotoxicity assay

		Drug and Concentration Range				
		5-FU 0.033 to 500 µM	Adriamycin 5.10-4 to 10 µM	Irinotecan 0.005 to 100 µM	Etoposide 0.005 to 100 µM	Topotecan 0.001 to 20 µM
PDX code	Cancer type	IC50 (µM)				
HBCx-3	Breast	N/C	0.65	N/T	N/C	4.5
HBCx-19	Breast	N/C	0.2	N/T	33.09	4.13
HBCx-23	Breast	N/C	0.5	N/T	90.05	N/C
MCM001-AV	Melanoma	218.82	0.4	N/T	61.55	0.42
MCM002-FJ	Melanoma	N/C	0.17	N/T	N/C	13.23
ODA14-RAV	Brain	175.08	0.74	N/T	36.1	0.76
OVA2-BUR	Ovary	124.44	0.75	N/T	58.79	0.33
GBM14-CHA	Brain	198.74	0.33	N/T	15.61	0.42
SC6	Lung	147.33	0.09	N/T	3.6	0.44
TC01	Colorectal	320.13	1.74	N/C	N/C	3.03
TC71	Colorectal	35.39	0.22	9.96	1.43	0.03
TC122A	Colorectal	140.6	2.4	55.41	N/C	1.62

N/C - Unable to accurately fit curve to calculate IC50 : 50% inhibition of viability was not obtained with the greatest concentration



PERSPECTIVES

- Our PDX collection is based on several large panels representing major solid tumor types (breast, colorectal and lung), as well as on a variety of models for other important cancers.
- This preclinical platform represents a powerful tool to identify optimal therapeutic options for patients by exploring and improving anti-cancer therapeutic strategies.
- PDX offer today's most clinically relevant models for target validation
- Extensive SOC response annotations for model choice based on drug resistance/sensitivity
- Deep molecular characterization of our PDX panels allows for identification of predictive drug response markers and companion test early development programs
- XenTech highly experienced staff deliver high quality services in preclinical pharmacology:
- drug tolerability and efficacy assays; blood and tissue sampling for PK/PD studies
- XenTech R&D department continuously provides new and improved services, such as:
  - Metastatic models with in vivo imaging metastases detection
  - In vitro cytotoxicity assays with PDX-derived primary cell cultures
  - Post-chemotherapy residual disease models for new target and CSC exploratory studies