IOM Workshop on Comparative Oncology

Pharmacokinetics in Companion Animal Cancer Studies

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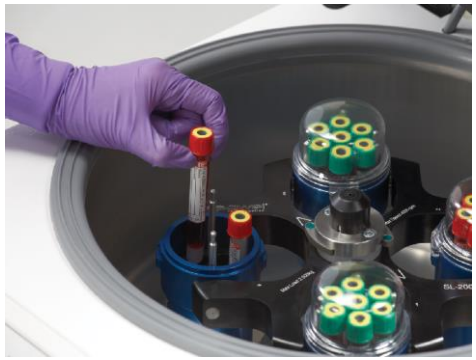
Topics to be Covered

1. **Carrying out PK studies in companion animals**
    a. Variability in PK data
    b. Utilization of PK data
2. **Dose finding studies inform PK and toxicity (PD)**
    a. Dose escalation studies
    b. Drug delivery and toxicity assessment in a manner consistent with human use
3. **Impact of tissue sampling on PK/PD measures**
    a. PK/PD correlations
    b. Assessment of clinical applicability

Carrying Out PK Studies



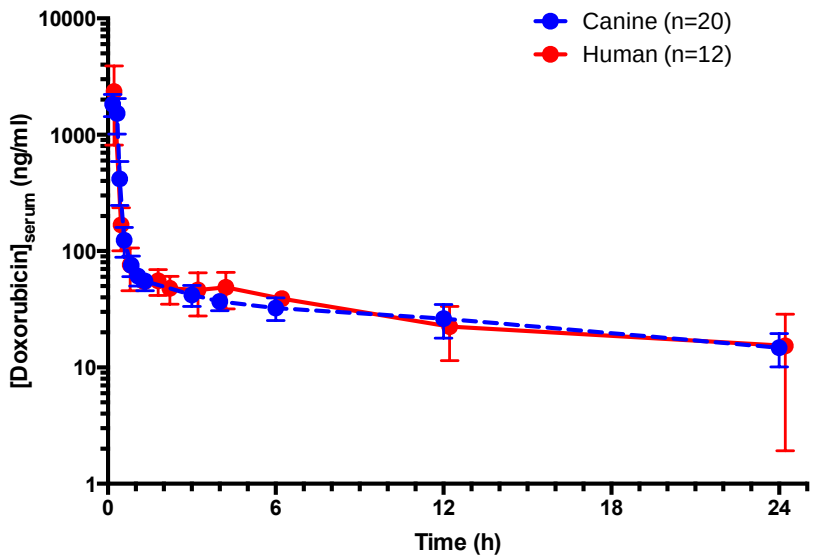
- ✓Time course sampling in individual animals
- ✓Drug dispensing done in a manner consistent with human medicine
- ✓Sample collection done by a professional staff
- ✓Defined SOPs for sample processing

Variability in Pharmacokinetics

Doxorubicin in Humans and Dogs

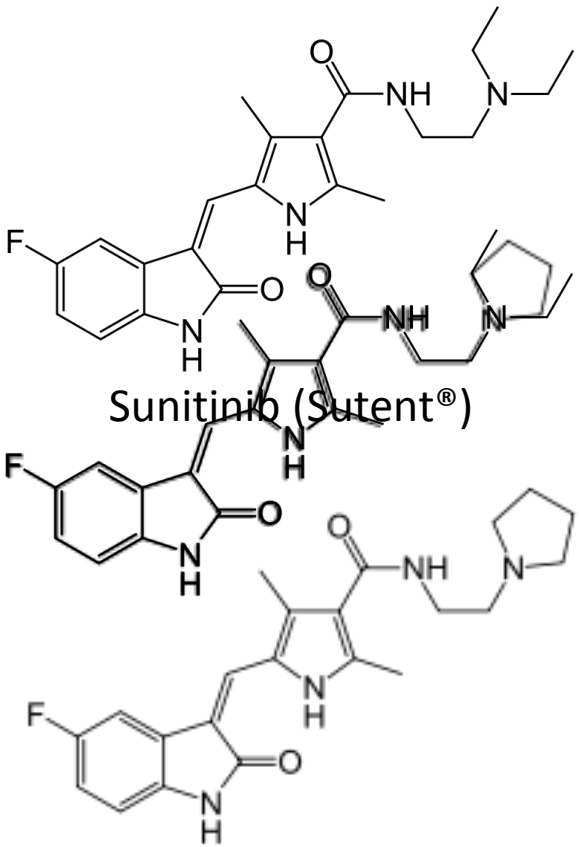
Exposure following a therapeutic dose of 60 mg/m^2 (humans) or 30 mg/m^2 (dogs)

Species	AUC (ng/ml) x hr	SD	CV
Human (n=12)	1536	422	27.5%
Dog (n=20)	1591	327	20.5%



- Similar drug exposure at the MTD-based dose
- Similar variability is observed
- Time-course of drug exposure is similar

Variability in Pharmacokinetics



Sunitinib (Sutent®)

Toceranib (Palladia®)

PK Parameter	Human[1] (50 mg qD)	Dog[2] (3.25 mg/kg EOD)
C_{max} (ng/ml)	27.7 (51%)	79.0 (32%)
C_{min} (ng/ml)	44.0 (59%)	19.1 (62%)
AUC_{0-t} (ng/ml) x hr	420 (50%)	1870 (35%)

1- Faivre *et al. J Clin Oncol* 24:25-35, 2006.
2- Yancey *et al. J Vet Pharmacol Therap* 33:162-171, 2009.

For Dose Comparison:

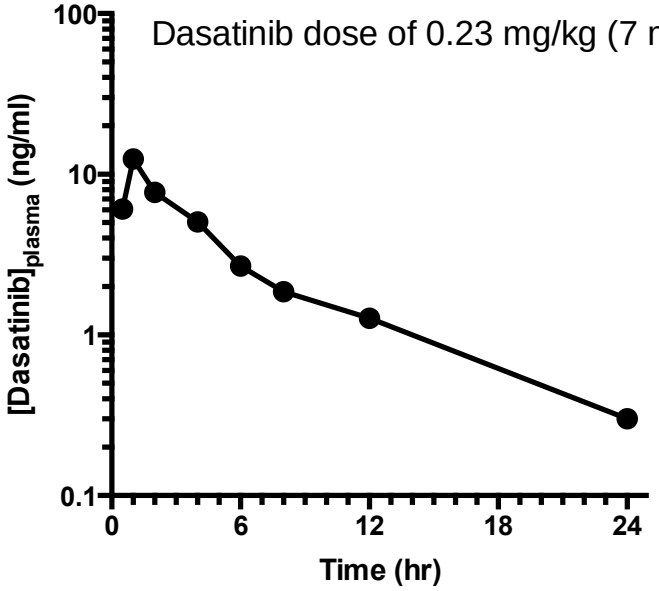
Human 50 mg is approximately = 30 mg/m^2 or 0.7 mg/kg

Dog 3.25 mg/kg is approximately = 100 mg/m^2

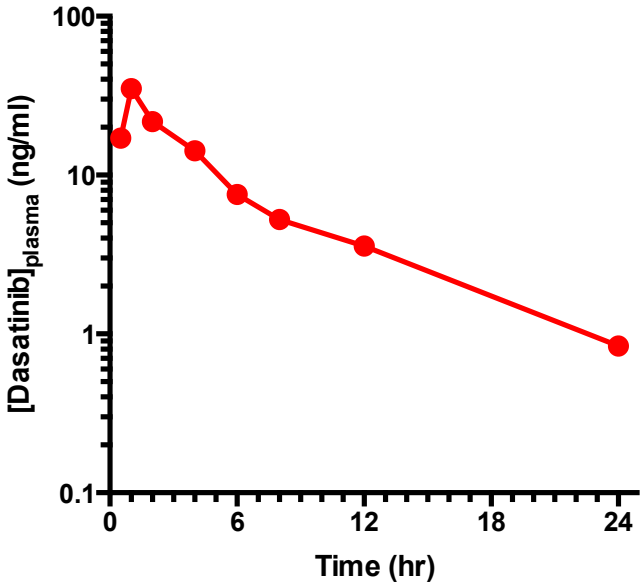
Utilization of PK Data

(Microdosing and TDM)

Microdosing with dasatinib to determine daily dose to achieve a C_{max} of 35 nM, a putative active plasma concentration of drug.



Predicted drug levels at 0.65 mg/kg dasatinib dose



- Test efficacy and toxicity at target dose
- Determine feasibility and accuracy of PK-directed, microdosing-based therapeutic drug monitoring

Current ongoing study, PI: Dr. Seguin

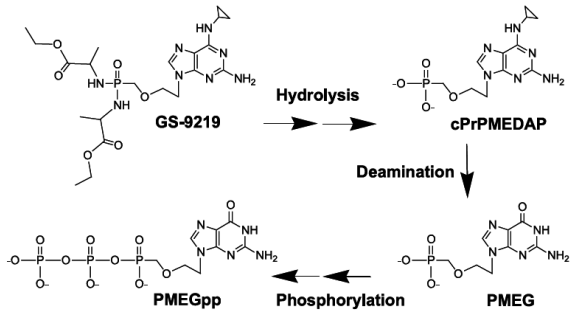
Dose Escalation Studies

(Dog Phase I Trials)

Table 1. GS-9219 scheduled treatment regimens evaluated

Cohort	GS-9219 dose (mg/kg)*	Frequency†	Dogs treated	Dose-limiting adverse events	Best response observed
Cohort 1-low	0.20	Daily for 5 d, every 21 d	7‡	1	5 CR/2 PR
Cohort 1-high	0.29	Daily for 5 d, every 21 d	6§	3	5 CR/1 PR
Cohort 2-low	0.66	Once every 7 d	3	0	2 CR/0 PR
Cohort 2-high	0.82	Once every 7 d	6§	2	3 CR/2 PR
Cohort 3-low	0.66	Once every 14 d	3	0	1 CR/0 PR
Cohort 3-high	0.82	Once every 14 d	6§	1	3 CR/2 PR
Cohort 4-low	0.66	Once every 21 d	3	0	2 CR/0 PR
Cohort 4-high	0.82	Once every 21 d	4‖	0	2 CR/0 PR

*Based on GS-9219 free base.
†Each regimen was to be repeated for five total treatment cycles.
‡One dog was diagnosed with a second malignancy after entry and a seventh dog was added to cohort.
§Cohort expanded to 6 dogs due to a single dose-limiting adverse event in first 3 dogs.
‖One dog died of tumor-related complications before response evaluation and an additional dog was added to the cohort.



- Dose escalation PK and Tox performed
- Exposure parameters defined in plasma and PBMCs for parent and active metabolites
- More optimal schedules defined in terms of efficacy and toxicity

Table 3. Pharmacokinetic parameters following 30 min intravenous infusion of GS-9219 at 0.82 mg/kg

	Plasma				PBMC
	$t_{1/2}$ (h)	C_{max} (nmol/L)	T_{max} (h)	$AUC_{(0-\infty)}$ (nmol/L h)	Total C_{24} (nmol/L)*
PMEG	ND†	<12.3	ND	ND	4,020 ± 4,400
cPrPMEDAP	4.02 ± 0.30	404 ± 91	2.00 ± 0	3,700 ± 1,140	456 ± 736
GS-9219	0.33 ± 0.20	4,280 ± 1,550	0.32 ± 0.16	2,910 ± 830	NP

NOTE: Plasma and PBMC pharmacokinetic parameters are the mean ± SD based on n = 8 and 6 dogs, respectively. NP, not performed.
*Total concentration represents intracellular concentrations of parent nucleotide and all of its phosphorylated species measured following dephosphorylation.
†ND, not determined, as plasma levels of PMEG were below level of detection.

Assessment of GS-9219 in a Pet Dog Model of Non-Hodgkin's Lymphoma

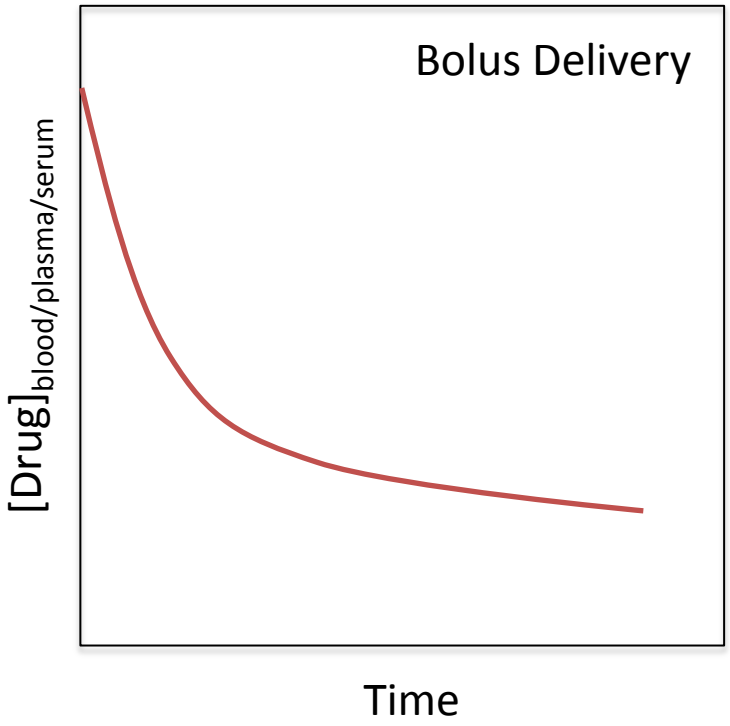
David M. Vail,[1] Douglas H. Thamm,[2] Hans Reiser,[3] Adrian S. Ray,[3] Grushenka H.I. Wolfgang,[3] William J. Watkins,[3] Darius Babusis,[3] Ilana N. Henne,[3] Michael J. Hawkins,[3] Ilene D. Kurzman,[1] Robert Jeraj,[1] Matt Vanderhoek,[1] Susan Plaza,[2] Christie Anderson,[2] Mackenzie A. Wessel,[1] Cecilia Robat,[1] Jessica Lawrence,[1] and Daniel B. Tumas[3]

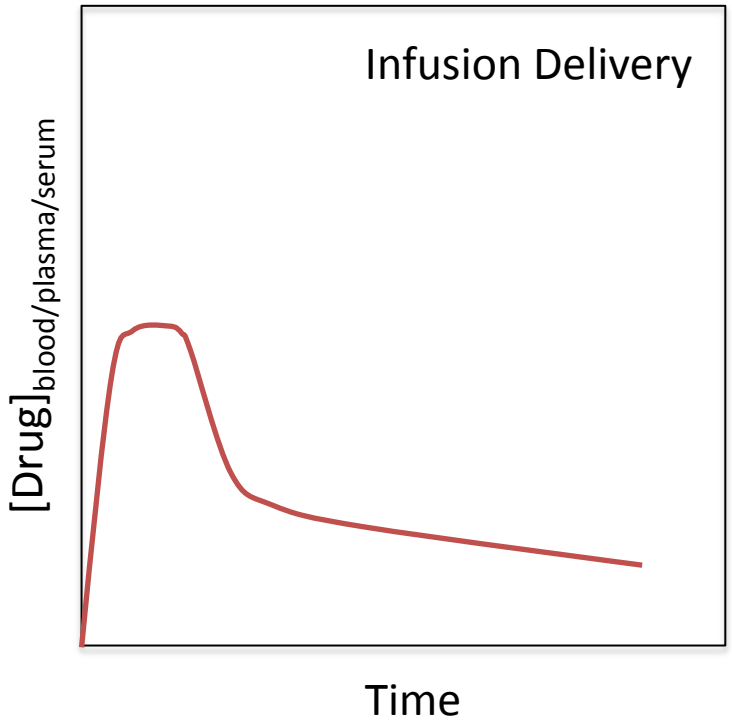
Clin Cancer Res 2009;15(10) May 15, 2009

Drug Delivery and Toxicity Assessment

Infusion vs. Bolus Delivery

For companion animal trials, drug is delivered in an appropriate vehicle using drug delivery schemas consistent with what is done in human trials





Toxicity Assessment

Graded and Standardized

Table 3. Sunitinib Grade 3 to 4 Toxicity per Patient and Cycle

Flat Dose (mg/m²)	50 mg/d (30 mg/m²)	75-100 mg/d (42 mg/m²)	100-150 mg/d (59 mg/m²)
No. of patients	9	12	3
No. of cycles	25	27	7
General toxicity			
Asthenia			
Per patient	2	6	2
Per cycle	2	9	2
Nausea/vomiting			
Per patient	—	1	—
Per cycle	—	3	—
Hypertension			
Per patient	—	1	1
Per cycle	—	1	1
Phlebitis			
Per patient	—	1	—
Per cycle	—	1	—
Pulmonary embolism			
Per patient	—	—	1
Per cycle	—	—	1
Neutropenia			
Per patient	3	2	—
Per cycle	10	4	—
Thrombocytopenia			
Per patient	2	2	1
Per cycle	3	2	1
Skin/mucous toxicity			
Palmar-plantar erythrodysesthesia syndrome			
Per patient	—	1	—
Per cycle	—	1	—
Edema peripheral			
Per patient	1	1	—
Per cycle	1	1	—
Esophagitis			
Per patient	—	1	—
Per cycle	—	1	—

Sunitinib in humans

Faivre *et al*. *J Clin Oncol* 24:25-35, 2006.

Table 3 Enrollment and toxicity by dose/regimen

Dose group	Regimen	Dose (mg/kg)	No. of dogs	Toxicities					
				Diarrhea	Anorexia	Vomiting	Fatigue	Hind limb weakness	Neutropenia
1	QD	1.25	5	2 (40%)	4 (80%)	2 (40%)	0 (0%)	2 (40%)	1 (20%)
2	QD	2.50	5	3 (60%)	4 (80%)	2 (40%)	0 (0%)	2 (40%)	0 (0%)
3	EOD	1.25	2	1 (50%)	1 (50%)	0 (0%)	0 (0%)	0 (0%)	1 (50%)
4	EOD	2.50	16	6 (38%)	5 (31%)	2 (13%)	1 (6%)	6 (38%)	1 (6%)
5	EOD	3.25	20	10 (50%)	8 (40%)	1 (5%)	4 (20%)	3 (15%)	0 (0%)
6	EOD	3.75	3	0 (0%)	1 (33%)	1 (33%)	0 (0%)	1 (33%)	0 (0%)
7	QDx7; EOD	2.50	5	2 (40%)	2 (40%)	1 (20%)	0 (0%)	3 (60%)	1 (20%)
8	QDx7; EOD	3.75	1	0 (0%)	1 (100%)	0 (0%)	1 (100%)	1 (100%)	0 (0%)

Toceranib in dogs

London *et al. Clin Cancer Res* 9:2755, 2003

Similar toxicities observed as well as some that were more species dependent:

Similar

- Weakness
- Vomiting
- Neutropenia

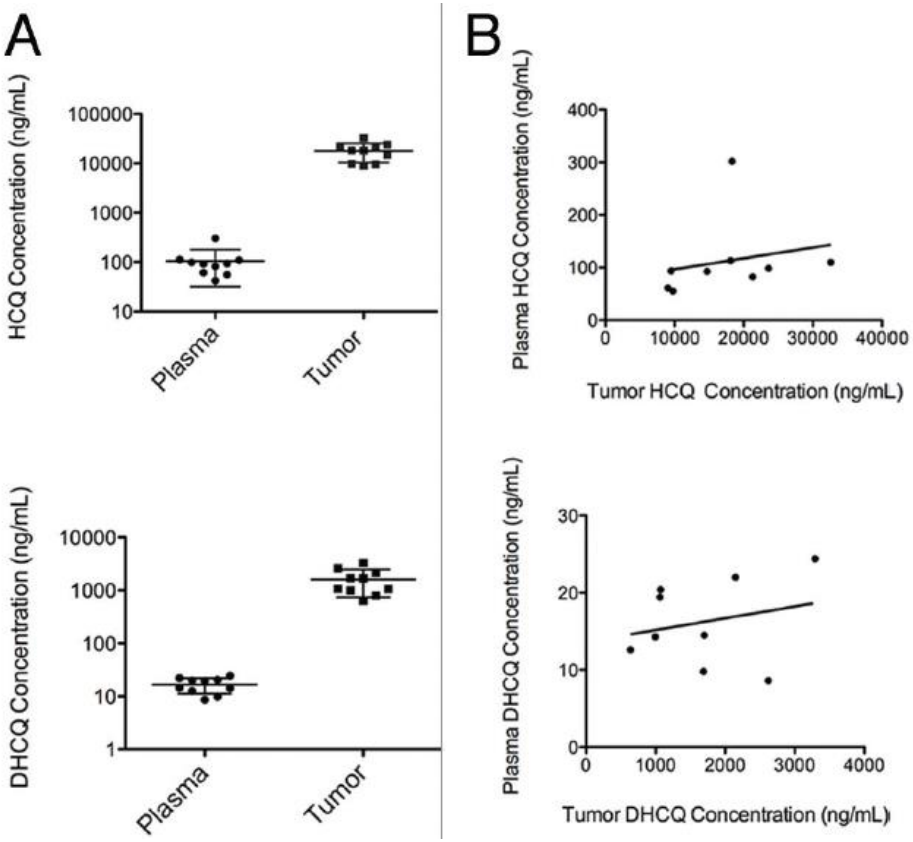
Species Dependent

- Diarrhea
- Hypertension
- Skin toxicity

Tissue Sampling

PK/PD Correlations

Plasma and tumor drug levels of hydroxychloroquine (HCQ) and the primary active metabolite N-desethylyHCQ (DHCQ) for the inhibition of autophagy.



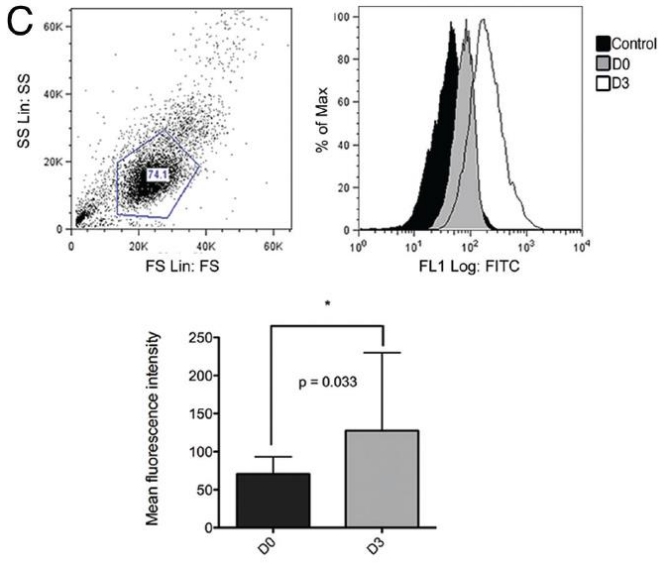
- HCQ and DHCQ accumulate approximately 100-fold in tumor tissue as compared to plasma levels.
- There is no correlation between plasma and tumor drug levels.

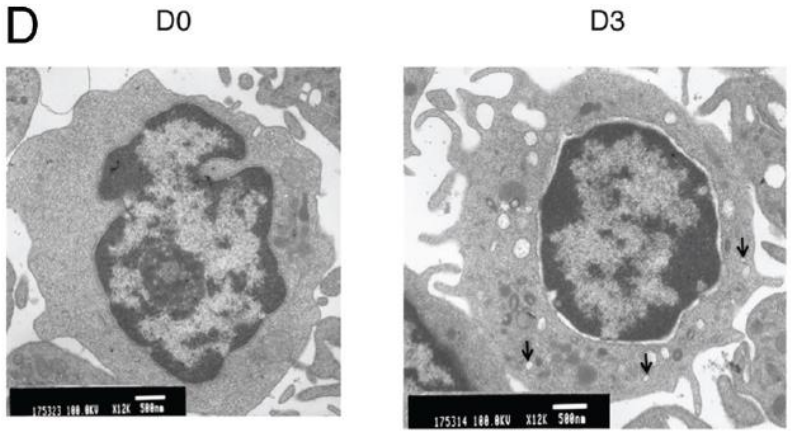
Thus, using plasma drug levels as a measure of drug exposure for potential dose modification is not indicated by these findings.

Barnard *et al. Autophagy* 10:1415, 2014

Tissue Sampling

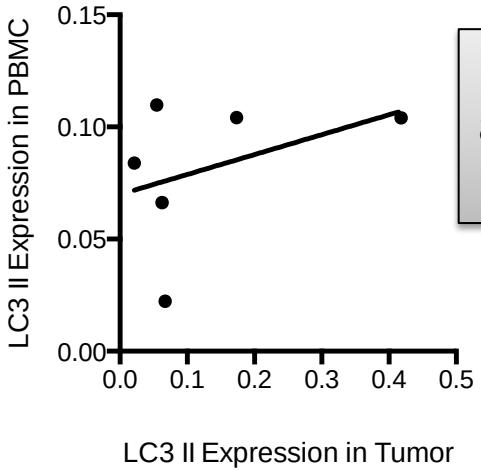
PD/PD Correlations





PBMCs from dogs treated with HCQ showed changes consistent with inhibition of autophagy by HCQ.

- Increase in LC3 positive cells
- Increase in autophagic vesicles as measured by EM



No correlation between autophagy inhibition in PBMCs and tumors

Barnard *et al. Autophagy* 10:1415, 2014

Clinical Trials of HCQ in Cancer

CLINICAL RESEARCH PAPER

Autophagy 10:8, 1403–1414; August 2014; © 2014 Landes Bioscience

Combined autophagy and HDAC inhibition

A phase I safety, tolerability, pharmacokinetic, and pharmacodynamic analysis of hydroxychloroquine in combination with the HDAC inhibitor vorinostat in patients with advanced solid tumors

Devalingam Mahalingam,[1,†,*] Monica Mita,[1,2,†] John Sarantopoulos,[1] Leslie Wood,[1] Ravi Amaravadi,[3] Lisa Davis,[4] Alain Mita,[1,2] Tyler J Curiel,[1] Claudia M Espitia,[1] Steffan T Nawrocki,[1] Francis J Giles,[1,5] and Jennifer S Carew[1,6,*]

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Autophagy 10:8, 1–10; August 2014; © 2014 Landes Bioscience

A phase I/II trial of hydroxychloroquine in conjunction with radiation therapy and concurrent and adjuvant temozolomide in patients with newly diagnosed glioblastoma multiforme

Myrna R Rosenfeld,[1,2] Xiaobu Ye,[1,3] Jeffrey G Supko,[1,4] Serena Desideri,[1,3] Stuart A Grossman,[1,3] Steven Brem,[1,5] Tom Mikkelson,[1,6] Daniel Wang,[7] Yunyoung C Chang,[7] Janice Hu,[7] Quentin McAfee,[7] Joy Fisher,[1,3] Andrea Troxel,[8] Shengfu Piao,[7] Daniel F Heitjan,[8] Kay See Tan,[8] Laura Pontiggia,[9] Peter J O'Dwyer,[7,10] Lisa E Davis,[7,11] Ravi K Amaravadi[7,10,*]

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Autophagy 10:8, 1–12; August 2014; © 2014 Landes Bioscience

Combined MTOR and autophagy inhibition

Phase I trial of hydroxychloroquine and temsirolimus in patients with advanced solid tumors and melanoma

Reshma Rangwala,[1,†] Yunyoung C Chang,[1,‡] Janice Hu,[1] Kenneth Algazy,[1] Tracey Evans,[1] Leslie Fecher,[1,§] Lynn Schuchter,[1] Drew A Torigian,[2] Jeffrey Panosian,[2] Andrea Troxel,[3] Kay-See Tan,[3] Daniel F Heitjan,[3] Angela Demichele,[1] David Vaughn,[1] Maryann Redlinger,[1] Abass Alavi,[2] Jonathon Kaiser,[4] Laura Pontiggia,[5] Lisa E Davis,[1,4] Peter J O'Dwyer,[1] and Ravi K Amaravadi[1,*]

CLINICAL RESEARCH PAPER

Autophagy 10:8, 1380–1390; August 2014; © 2014 Landes Bioscience

Combined autophagy and proteasome inhibition

A phase 1 trial of hydroxychloroquine and bortezomib in patients with relapsed/refractory myeloma

Dan T Vogl,[1,*] Edward A Stadtmauer,[1] Kay See Tan,[2] Daniel F Heitjan,[2] Lisa E Davis,[3] Laura Pontiggia,[4] Reshma Rangwala,[1,†] Shengfu Piao,[1] Yunyoung C Chang,[1,‡] Emma C Scott,[1] Thomas M Paul,[1] Charles W Nichols,[1] David L Porter,[1] Janeen Kaplan,[1] Gayle Mallon,[1] James E Bradner,[5] and Ravi K Amaravadi[1]

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Autophagy 10:8, 1–11; August 2014; © 2014 Landes Bioscience

Phase I trial of hydroxychloroquine with dose-intense temozolomide in patients with advanced solid tumors and melanoma

Reshma Rangwala,[1,†] Robert Leone,[1,‡] Yunyoung C Chang,[1,§] Leslie Fecher,[1,‖] Lynn M Schuchter,[1] Amy Kramer,[1] Kay-See Tan,[2] Daniel F Heitjan,[2] Glenda Rodgers,[1] Maryann Gallagher,[1] Shengfu Piao,[1] Andrea B Troxel,[2] Tracey Evans,[1] Angela DeMichele,[1] Katherine L Nathanson,[1] Peter J O'Dwyer,[1] Jonathon Kaiser,[3] Laura Pontiggia,[4] Lisa E Davis,[1,3] and Ravi K Amaravadi[1,*]

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Phase I clinical trial and pharmacodynamic evaluation of combination hydroxychloroquine and doxorubicin treatment in pet dogs treated for spontaneously occurring lymphoma

Rebecca A Barnard,[1] Luke A Wittenburg,[1] Ravi K Amaravadi,[2] Daniel L Gustafson,[1] Andrew Thorburn,[3] and Douglas H Thamm[1,*]

Canine trial published as part of a series with human clinical trials of HCQ