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Cardiovascular Toxicity Assessment in Oncology
Trials Workshop

Organized by FDA, AACR, ACC, AHA & ASCO



Session 3:
Nonclinical

Overview: What nonclinical studies are available to interrogate potential cardiovascular risk of an oncology product?

Hugo M. Vargas, PhD, DSP

Integrated Discovery & Safety Pharmacology

Comparative Biology and Safety Sciences

Amgen, Inc

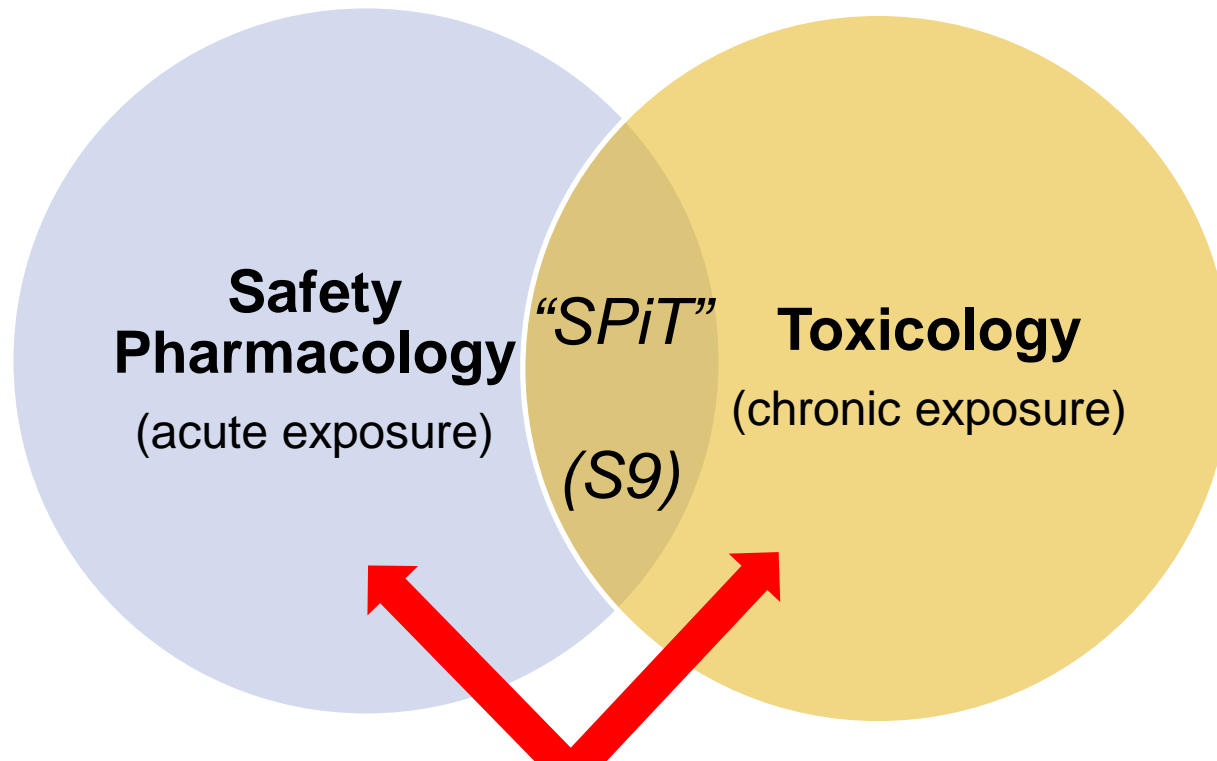
General nonclinical study requirements

	ICH M3 (Small molecules)	ICH S9 (Small/large molecule)	ICH S6 (Large molecules)
Repeat-dose studies	≤1 mo, supports P1	1 mo, supports P1 and P2	≤1 mo, supports P1
Repeat-dose chronic studies	6 mo (rodent) and 9 mo (non-rodent) as needed for clinical dosing duration, supports filing	3 mo, supports P3/filing	6 mo (1 sp) as needed for clinical dosing duration, supports filing
Safety Pharm	Dedicated Functional Studies (CV, CNS, Respiratory)	No dedicated studies unless warranted (SPiT approach)	No dedicated studies unless warranted (SPiT approach)
Genotoxicity	Supports P1	For marketing	Not warranted
Repro Tox	Fertility and embryo-fetal, supports P3; Peri-postnatal, supports marketing	Embryo-fetal, supports marketing	Embryo-fetal, supports P3; Peri-postnatal, supports marketing
Carcinogenicity	2 species, supports marketing	Not warranted	Not generally warranted
Misc	Local tolerance: conduct Phototoxicity, Impurities: as indicated by data	Phototoxicity, Local tolerance, Impurities: assessment and justification with filing	Phototoxicity: not warranted Tolerance: not warranted Impurities: as indicated by data



Major differences in regulatory requirements

CV Safety Pharmacology Assessment: Nonclinical Evaluation of Oncology Products



Acute *and* chronic functional effects
to be considered

SPiT: Safety Pharm endpoints In Toxicology studies (ICH S9)

Cardiovascular Risk Evaluation: “The Tool Box”

Safety Pharmacology Society Survey-2016 (85 respondents)

	Nonclinical Methods	Frequently Used (%)	Rarely Used (%)	Count (N)
In vitro - Ex vivo	hERG & non-hERG (Patch clamp)	89	11	66
	hERG trafficking	42	58	72
	Ion Channel Binding	57	43	68
	Human iPSC-cardiomyocytes	46	54	68
	Isolated heart (Langendorff)	38	62	63
	Cardiac wedge prep	18	82	51
	APD recordings (Purkinje fibers, etc)	51	49	61
In vivo	Anesthetized animal models	53	47	58
	Telemetry (implant; non-rodent)	88	12	67
	Telemetry (jacket; non-rodent)	62	38	63
	Pro-arrhythmia models	12	88	49
	Zebrafish models	4	96	47

>50% (majority)

Assessing CV Liability for an Oncology Drug:

The oncology drug target co-exists in the myocardium

Drugs Stage	Safety Activity	Comments
Discovery	Target liability review	Review of literature; human data of relevance; genetic info; competitor info.
Screening	hERG & non-hERG potency Isolated rabbit heart	Pro: evaluate off-target and target issues Con: solubility limitations?
Lead Optimization	Rat telemetry: BP & LVP	Pro: small animal; single or multi-day dose Con: Is rat relevant, e.g. target expression?
	Non-rodent telemetry: BP & LVP	Pro: single and multi-day dosing Con: PK/PD relationship to CV toxicity
Exploratory toxicity	Non-rodent: Toxicity: 7-14 day -Jacket Telemetry (ECG/HR) -Echocardiography -CV biomarkers	Pro: repeat-dosing; dedicated telemetry is an option Con: small groups (N≤3); timing of CV evaluation relative to PK
IND-enabling Safety Studies (GLP)	Rodent /Non-rodent Toxicity: 28 day -Jacket Telemetry (ECG/HR) -Echocardiography -CV biomarkers	Pro: larger group sizes; multi-day dosing Con: will CV toxicity emerge in 4 weeks; CV evaluation confounded by other effects (e.g., GI toxicity; dehydration, etc)?
FIH and beyond	BP, HR, ECG, vital signs -Echocardiography? -CV biomarkers?	Pro: monitoring of functional CV effects Con: what is a safety signal of concern? unanticipated events?

Cardiac Contractility & Dysfunction

An Emerging Safety Concern for Oncology Drugs

- **Several anti-cancer drugs cause contractile dysfunction*:**
 - Oncology SM: Doxorubicin, epirubicin, mitoxantrone, cyclophosphamide, 5-FU, capecitabine, sunitinib
 - Oncology LM: Trastuzumab
 - >30 kinases known to alter myocardial mechanics/function
- **Oncology patients may be susceptible to drug-induced cardiac dysfunction due to: age, co-morbidities, concomitant meds, prior chemotherapy**
- **Echocardiography: Essential tool for clinical LV function testing**
- **Nonclinical use in “SPiT” paradigm:**
 - Case-study with Doxorubicin in NHP

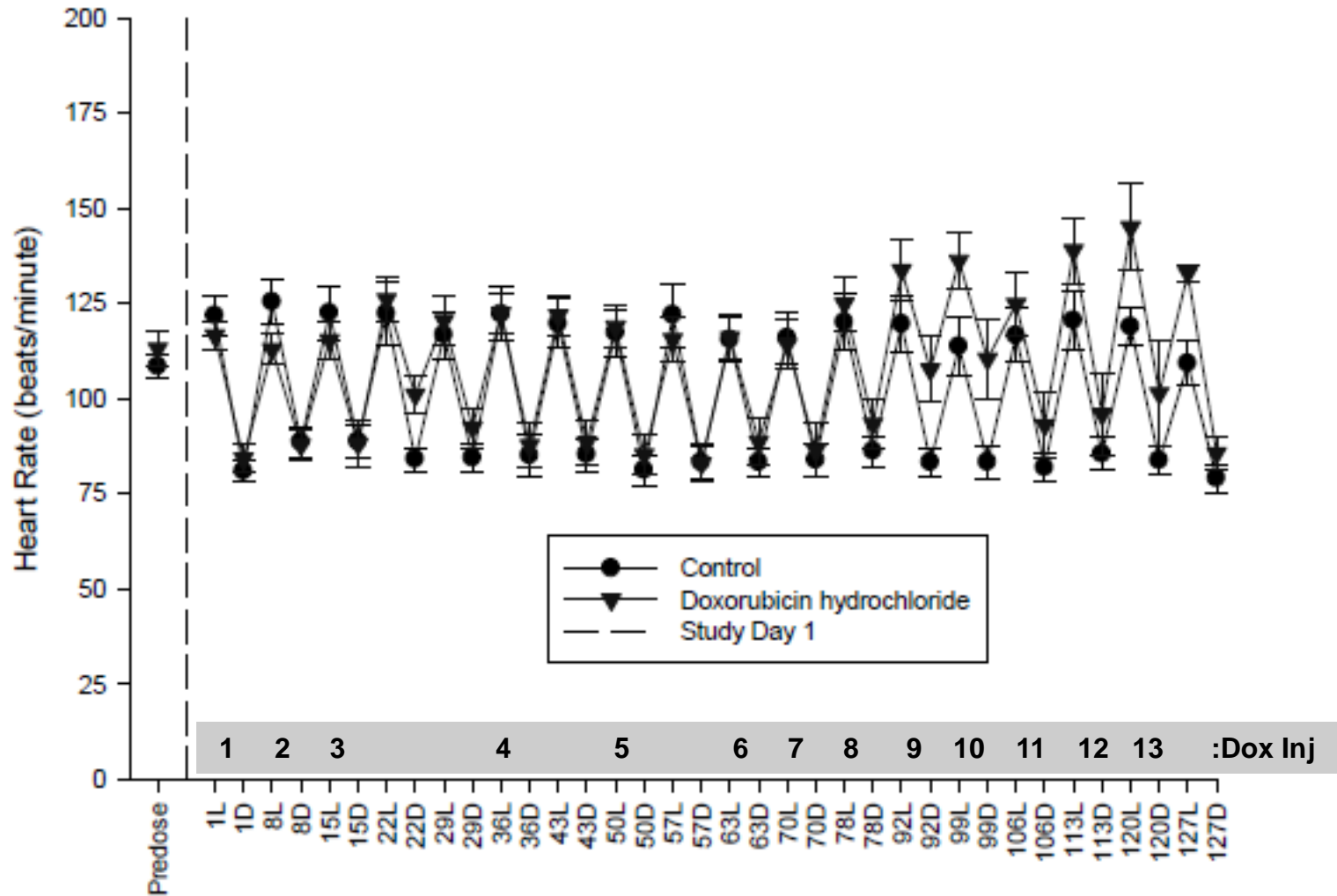
*: Slordal and Spigset, 2006; Force et al, 2006; Cheng & Force, 2010; Force and Kolaja, 2011; Eschenhagen et al (2011: Position paper from European Society of Cardiology)

DOX-induced LV Dysfunction in NHP:

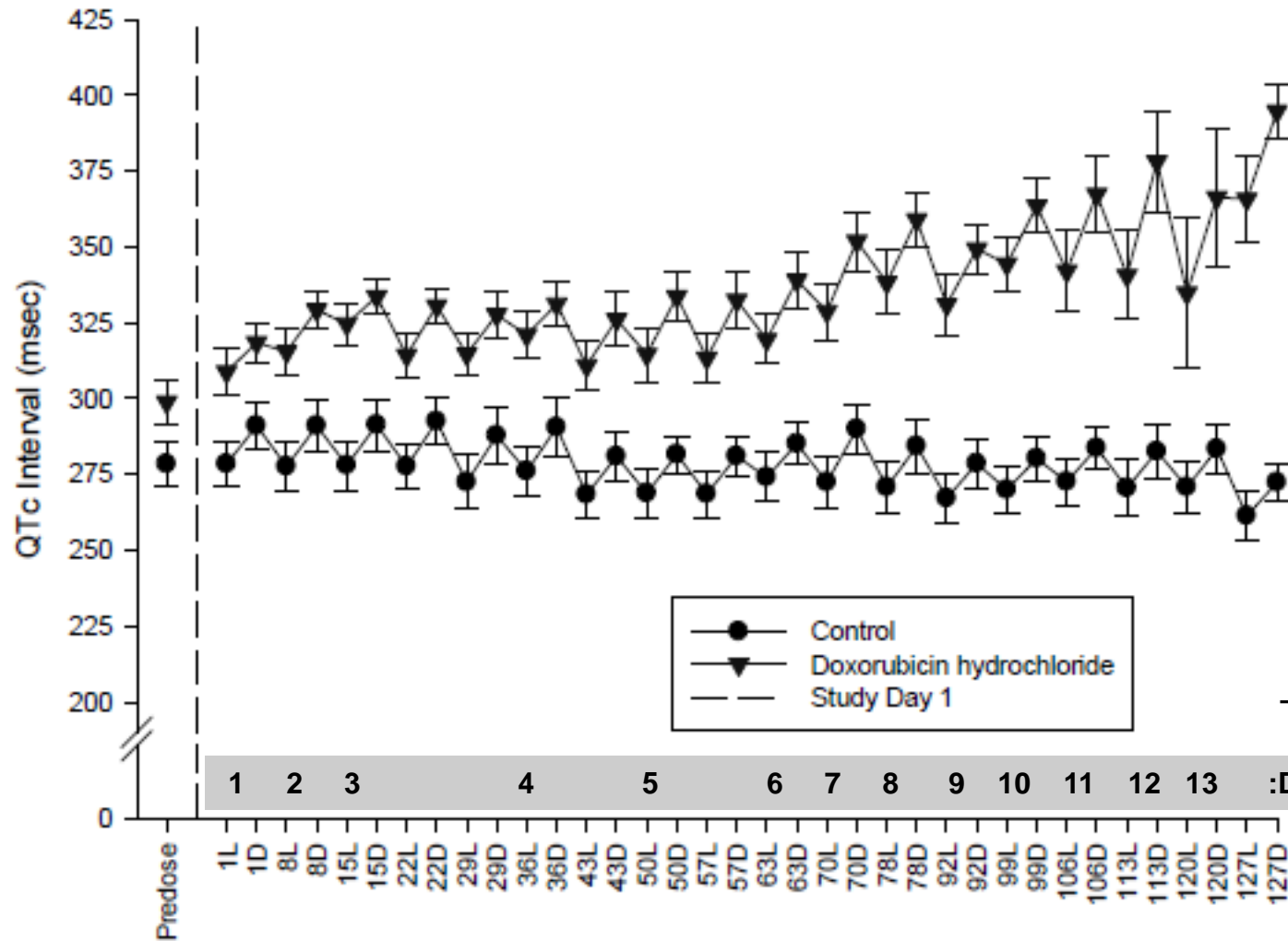
Study Design

- **Cynomolgus monkey (NHP): DOX (N=12) and Vehicle (N=12)**
 - Vehicle (IV): 0.9% NaCl for Injection (USP)
- **DOX: 2 mg/kg/wk x 3 wks, then by 1 mg/kg/wk every other week**
 - IV dose regimen selected to produce a gradual change in cardiac function and avoid overt bone marrow depression
 - Malik et. al., Proc Amer Assoc Cancer Res, Volume 45, 2004
 - Study Length: 4 mon
- **Cardiovascular Endpoints and Monitoring:**
 - **Implant Telemetry:** BP, LVP & ECG
 - Echocardiography
 - Cardiac Biomarkers: cTnl
- **Measurement Frequency:**
 - pre-study (baseline) & regularly (7-14 days) following first dose

Heart Rate

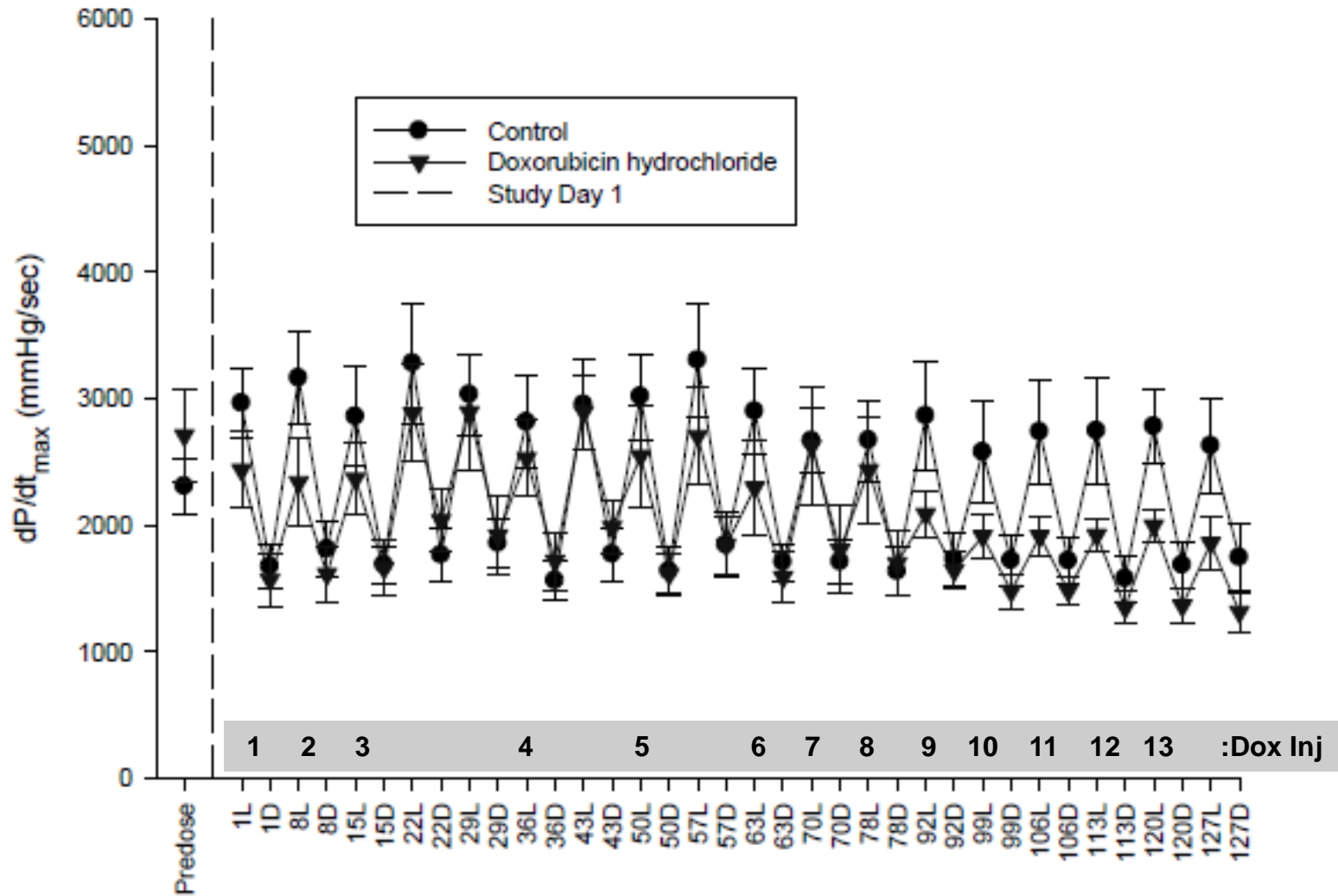


QTc

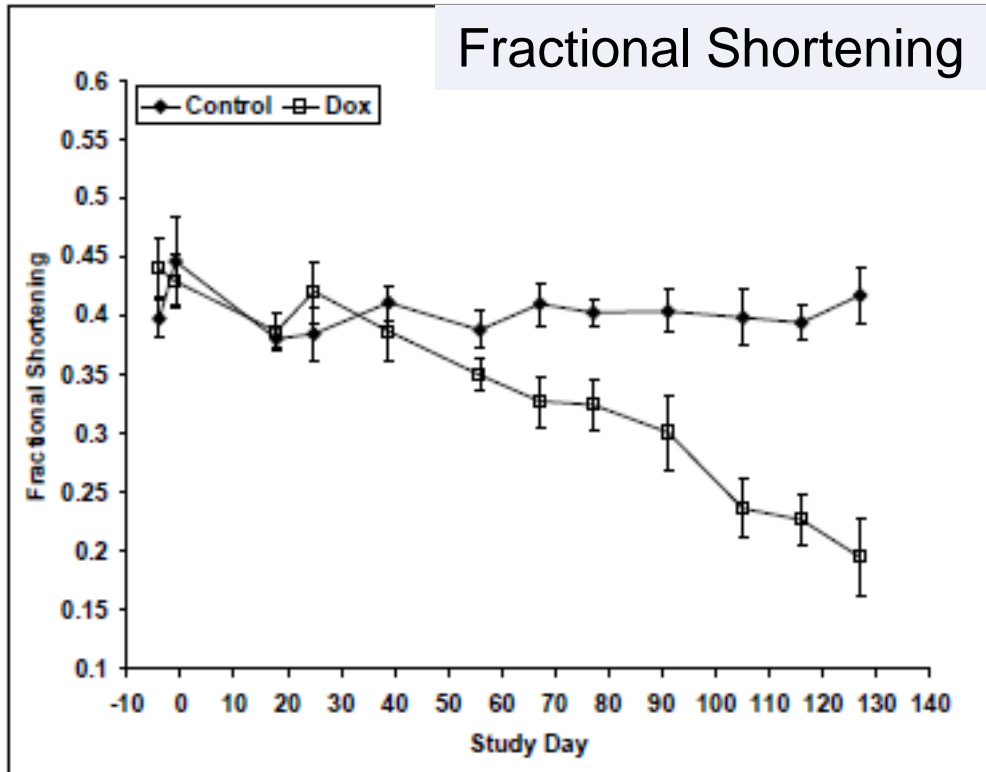


hERG-IC50:
 Direct: >30 μ M
 Trafficking: >30 μ M

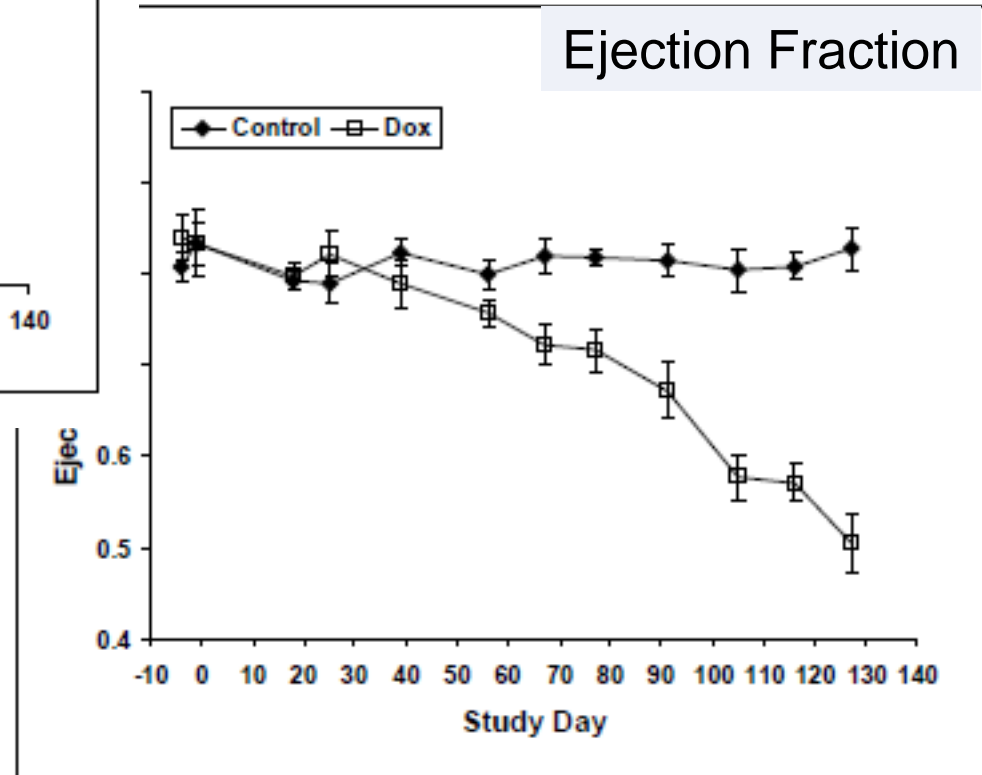
Index of LV Contractility: dP/dt max



Doxorubicin-Induced Heart Failure in NHP: Functional and Structural Changes over 4 months

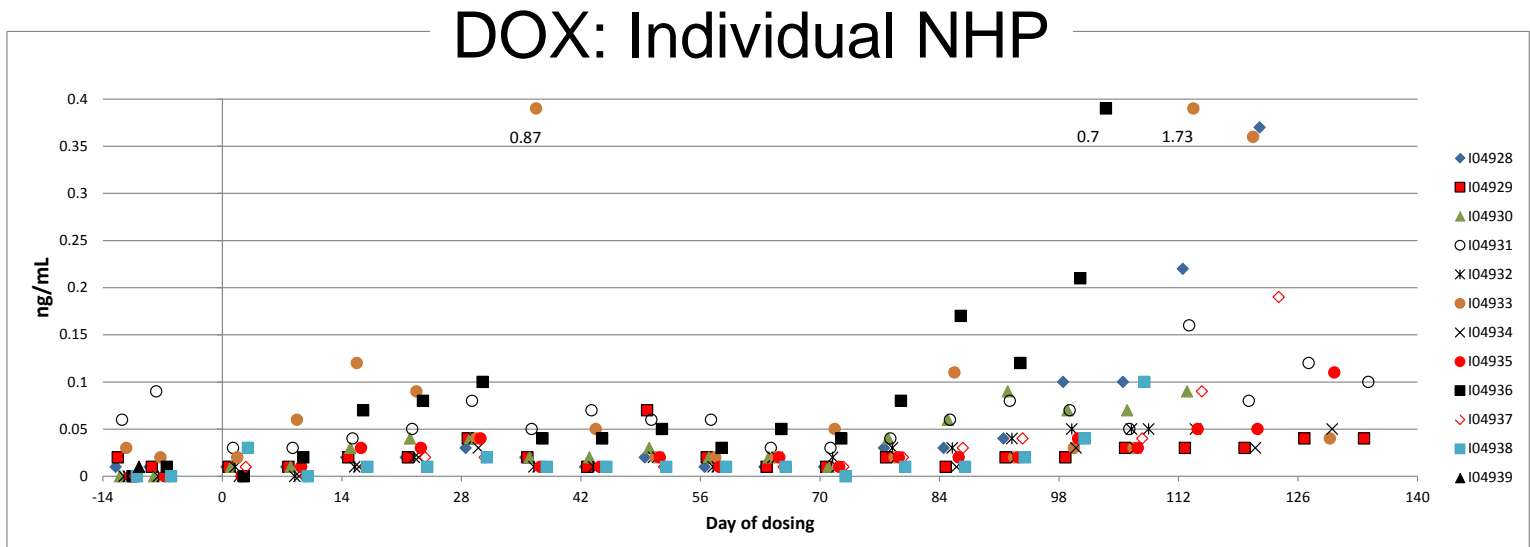
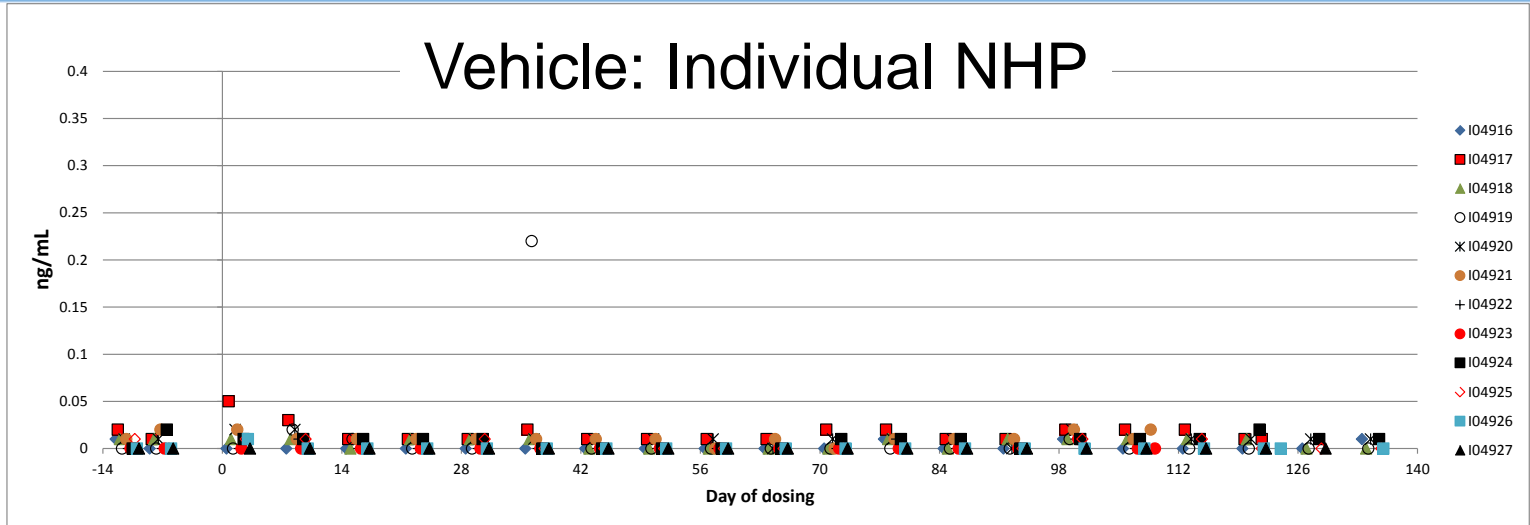


*Sonographer: Dr. Meg Sleeper
Echo collection under Ketamine anesthesia*



DOX: 2 or 1 mg/kg (weekly)

cTnI (Troponin) Levels



DOX Exposure-Response: NHP vs Human

Dose Injection	Dose (mg/m ²)	Cardiovascular Findings		Reference
		Cynomolgus	Human	
1	24	↑QTc	Ventricular arrhythmia	A
2	48	↑QTc	Ventricular arrhythmia	A
3	72	↑QTc		
4	84	↑QTc		
5	96	↑QTc; ↓EF/FS		
6	108	↑QTc; ↓EF/FS		
7	120	↑QTc; ↓EF/FS		
8	132	↑QTc; ↓EF/FS		
9	144	Above+ ↑HR; ↓dP/dtmax (min)		
10	156	Above+ ↑HR; ↓dP/dtmax (min)		
11	168	Above+ ↑HR; ↓dP/dtmax (min)		
12	180*	Above+ ↑HR; ↓dP/dtmax (min)		
13	192*	Above+ ↑HR; ↓dP/dtmax (min)		
14	204*	Above+ ↑HR; ↓dP/dtmax (min)	↓EF (<55%)	B
-	300	-	↓EF (<55%)	B
-	400	-	↓EF (CHF: ≤5%)	C
-	500	-	↓EF (CHF: 7-26%); ↑QTc	C, D
-	>700	-	↓EF (CHF: 18-48%)	C

*: not tolerated

Conclusions

- **CV functional assessment is needed → repeat-dose paradigms**
 - Oncology Products → balance regulatory & safety needs
 - Augment CV safety assessment in chronic toxicity studies
 - 3Rs: Minimize animal use, especially non-rodents
- **Methods are available for high quality functional data capture**
 - Non-invasive methods: ideal for CV toxicity application
 - Jacket telemetry & Minimally-invasive BP implants
 - Echocardiography: clinical translation
 - Invasive telemetry: best practice for chronic CV evaluations
- **“SPiT”: Opportunity for CV Hazard Identification & Risk Assmt.**
 - Pro: Evaluation multiple endpoints over long time periods
 - Con: Risk of false signals; timeframe for CV toxicity unknown; right species used; confidence in translation to human?

Acknowledgements

Amgen

- Michael Engwall, DVM, PhD
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- Nancy Everds, DVM, DACVP

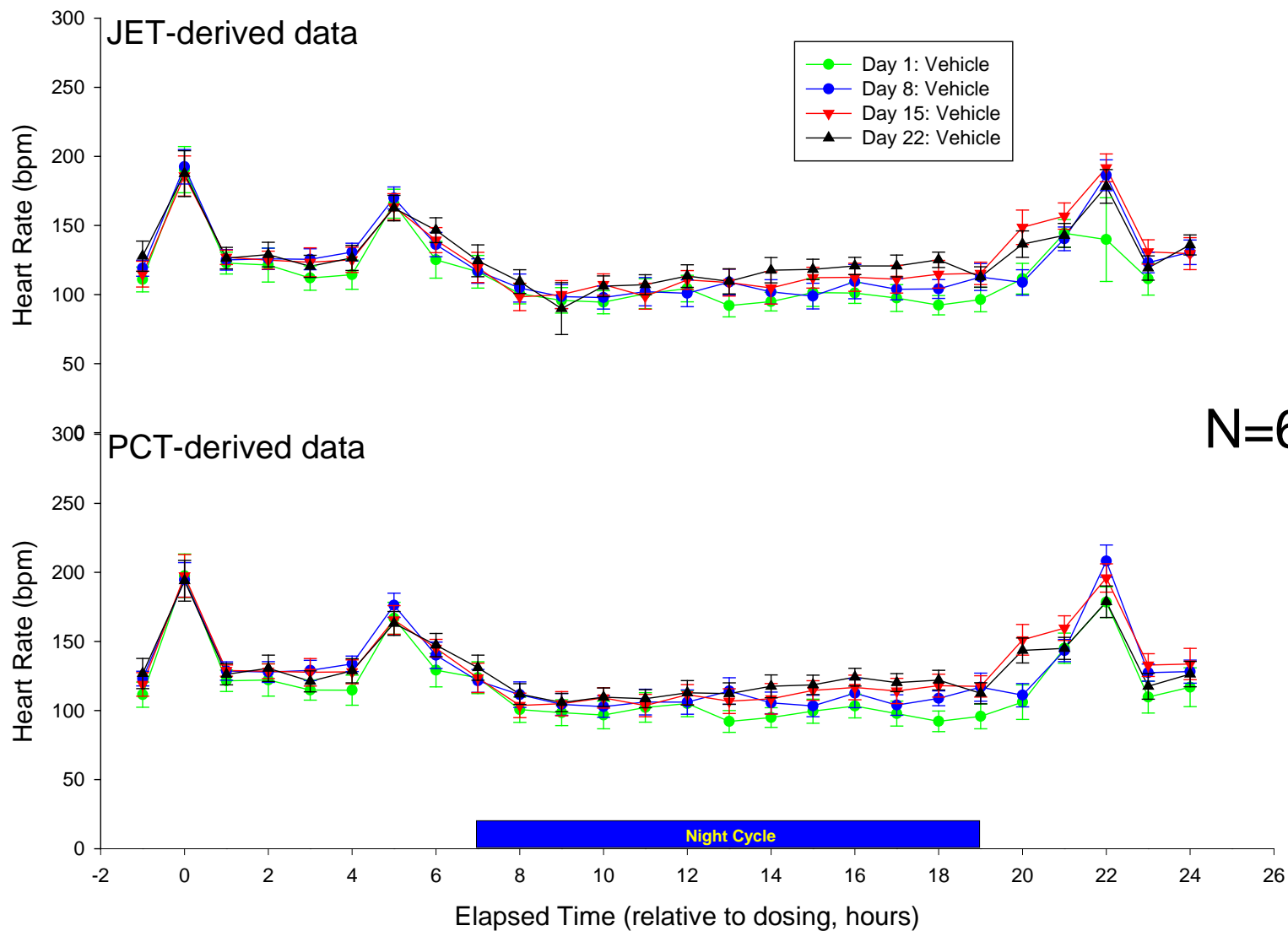
- Covance
 - Safety Pharmacology Group (Madison, WI)

Recent Publications on “SPiT”: Assess CV Safety after Repeated Dosing

- **Authier et al. (2013): Safety pharmacology investigations in toxicology studies: an industry survey.**
J. Pharmacol. Toxicol. Methods 68:44-51
 - Industry experience growing; regulatory acceptance of jacket telemetry
- **Redfern et al. (2013): Functional measurements in repeat-dose toxicity studies: the art of the possible.**
Toxicology Research DOI: 10.1039/c3tx20093k
 - Pros/cons of SPiT; how to do it right
- **Derakhchan et al. (2014): Detection of QTc interval prolongation using jacket telemetry in conscious non-human primates: comparison with implanted telemetry.**
British J. Pharmacol. 171:509-22
 - Amgen validation experience with a 1 month study design
 - Builds on prior jacket-based ECG collection in dogs (Chui et al., 2009)

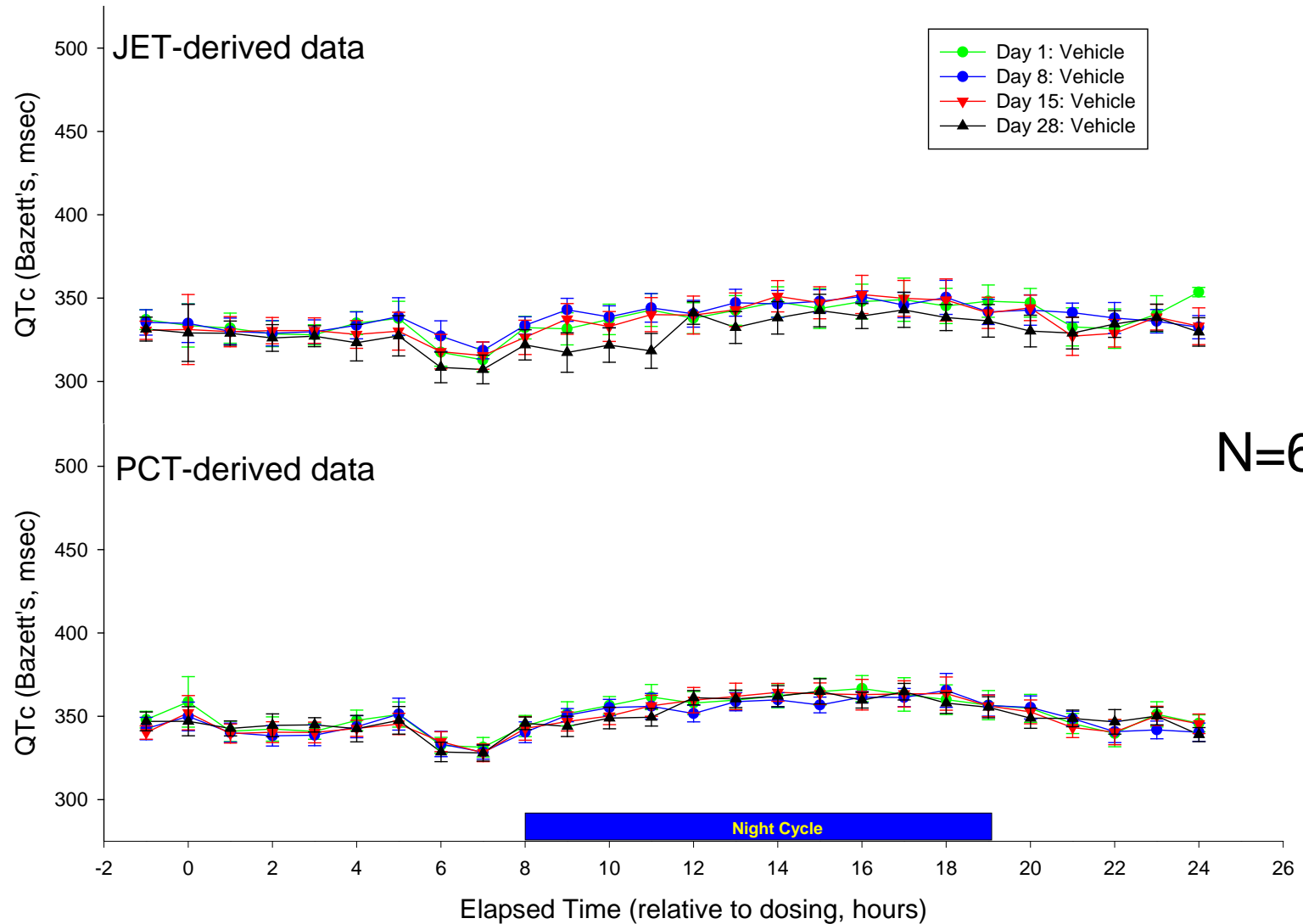
Jacket (JET) vs. Implant (PCT) Telemetry: NHP

Heart Rate following Vehicle (oral) over 4 weeks

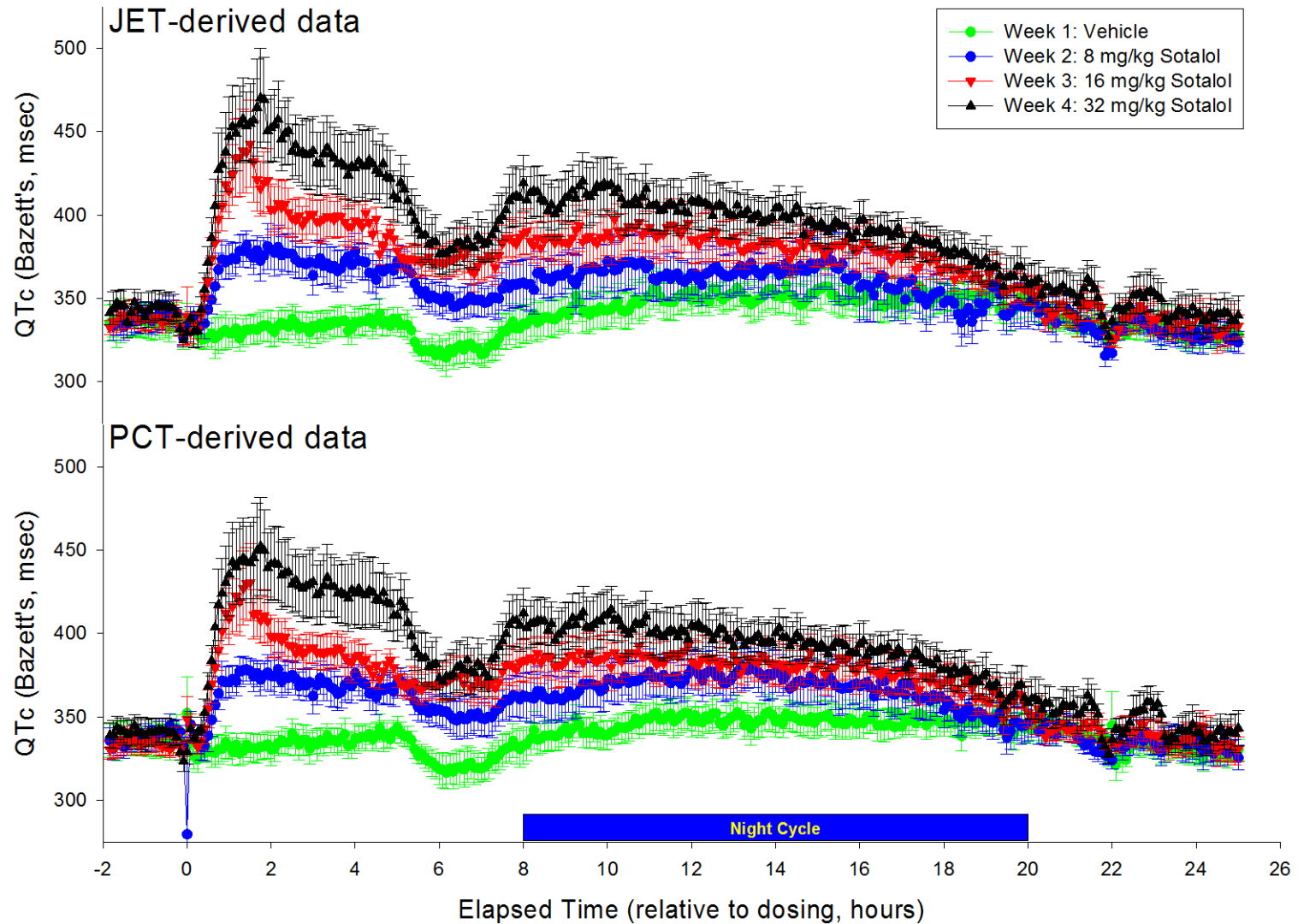


Jacket (JET) vs. Implant (PCT) Telemetry: NHP

QTc Interval following Vehicle (oral) over 4 weeks

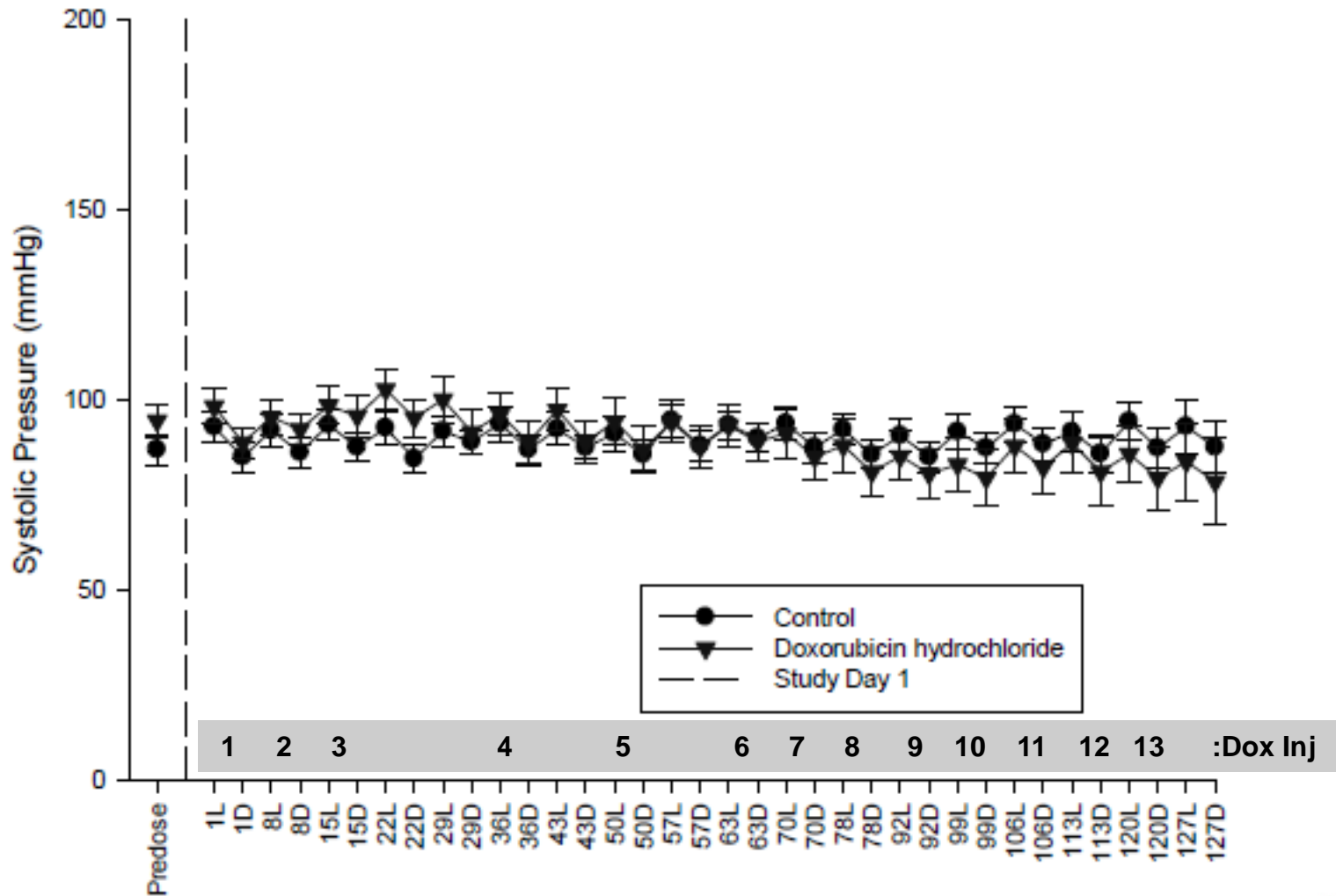


Jacket (JET) vs. Implant (PCT) Telemetry: NHP Sotalol-induced QTc Prolongation

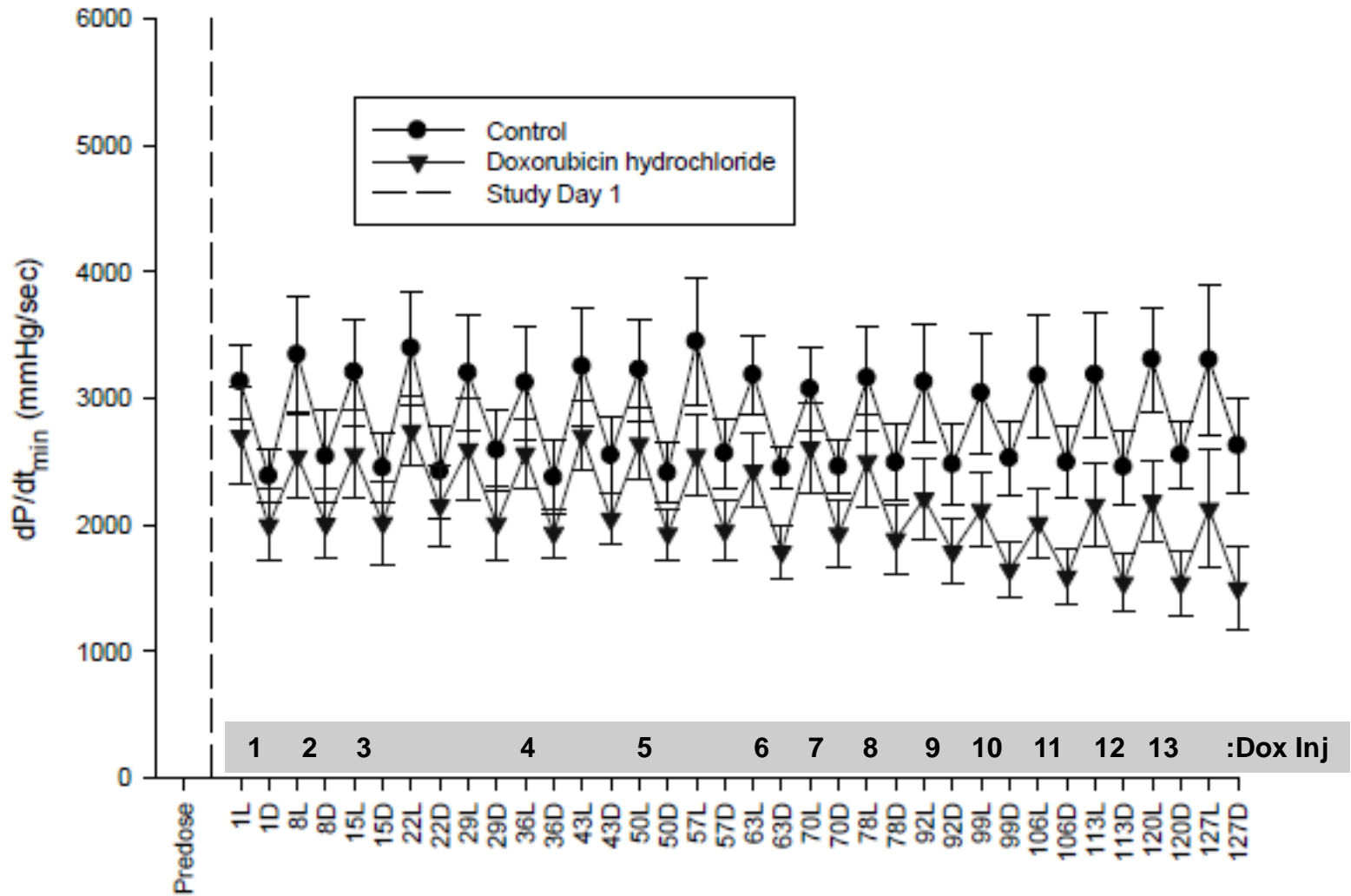


=6

Systolic Arterial Pressure



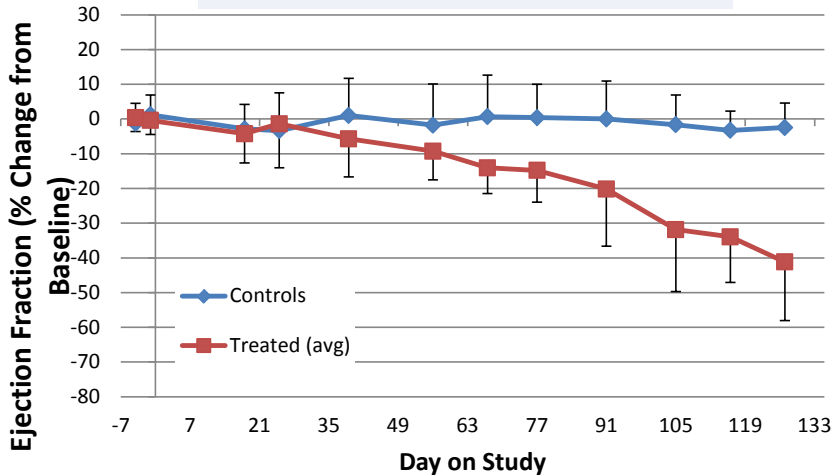
Index of LV Relaxation: dP/dt min



Doxorubicin-Induced Heart Failure in NHP

Functional and Structural Changes over 4 months

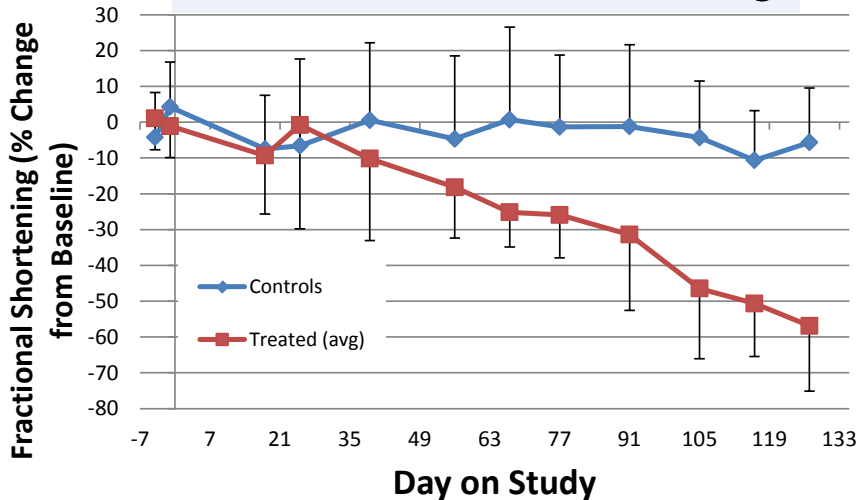
LV Ejection Fraction



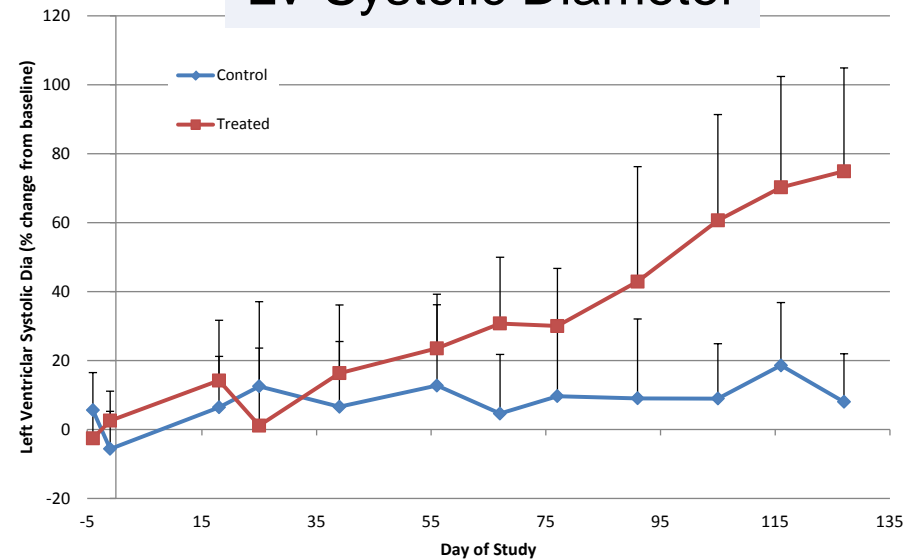
Sonographer: Dr. Meg Sleeper
Echo collection under Ketamine anesthesia

DOX: 2 or 1 mg/kg (weekly)

LV Fractional Shortening



LV Systolic Diameter



Histopathology

- **Minimal to mild degeneration of cardiomyocytes:**
 - DOX: 7 of 12 animals
 - VEH: 0 of 12 animals
- **Reduced immunoreactivity for cTnI and SOD2 in cardiomyocytes undergoing degeneration.**
- **Isolated instances of mild or moderate degeneration or necrosis in liver and mild or minimal fibrosis in the kidney**
 - considered secondary to hemodynamic changes

DOX CV TOX: Human References

A. J.S. Steinberg et al. *Cancer* 60:1213-1218 (1987)

- Single IV dosing (34 ± 12 mg/m²; 10 min) was associated with increased ventricular premature beats, ventricular couplets and supra-ventricular tachycardia over a 24 hr period (ambulatory ECG monitoring).

B. M.E. Caram et al. *Breast Cancer Res Treat* 152: 163-72 (2015)

- 11.5% (19/166) of dox-treated patients (240-359 mg/m²) had LVEF <55%

C. S.M. Swain et al. *Cancer* 97:2869-79 (2003)

- 26% of dox-treated patients experienced congestive heart failure (CHF) at 500 mg/m²
- Age (>65 yrs) was a risk factor for dox-induced CHF.
- >50% of dox-treated patients that developed CHF had LVEF <30% on study

D. T. Nousiainen et al. *J. Internal Med.* 245:359-363 (1999)

- QTc increased at 500 mg/m²; non-significant QTc prolongation at 200 and 400 mg/m²

CV Testing Strategy for Oncology Drug: Amgen Examples

- **Small Molecules: exploratory evaluation during lead optimization**
 - hERG blockade:
 - assess SAR and off-target potency
 - Isolated rabbit heart:
 - assess direct effects on electrophysiology & contractility
 - Rat Telemetry: profile drug for BP/HR & contractility effects
 - Single ascending doses or repeat (e.g., 4-day)
 - QTc/ECG evaluation integrated into repeat dose toxicology (non-rodent)
 - 14 day (exploratory; non-GLP) and/or IND-enabling studies (GLP)
 - Other approaches: case-by-case
 - target-based liability
 - non-rodent telemetry
 - echocardiography
- **Biologicals: case-by-case approach**
 - CV risk: low for direct hERG-mediated effects and off-target CV liabilities
 - QTc/ECG evaluation integrated into repeat dose toxicology (non-rodent)
 - LV function assessment: echocardiography
 - Assess: case by case based on target-based biology