P-glycoprotein activity is dependent on sex, circadian timing and feeding conditions – Implications for pharmacokinetics modeling

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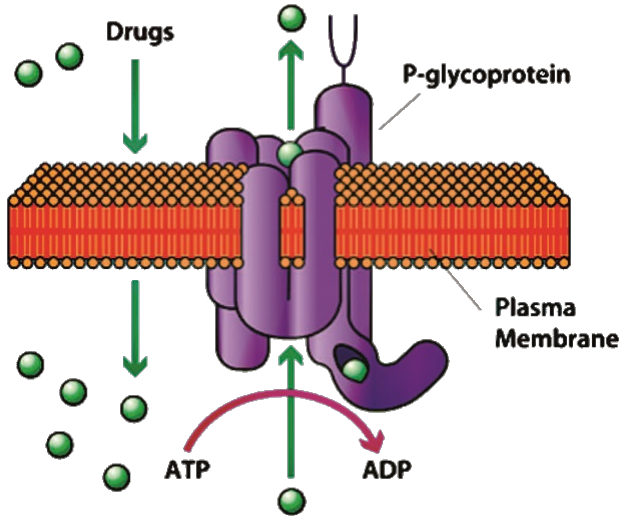
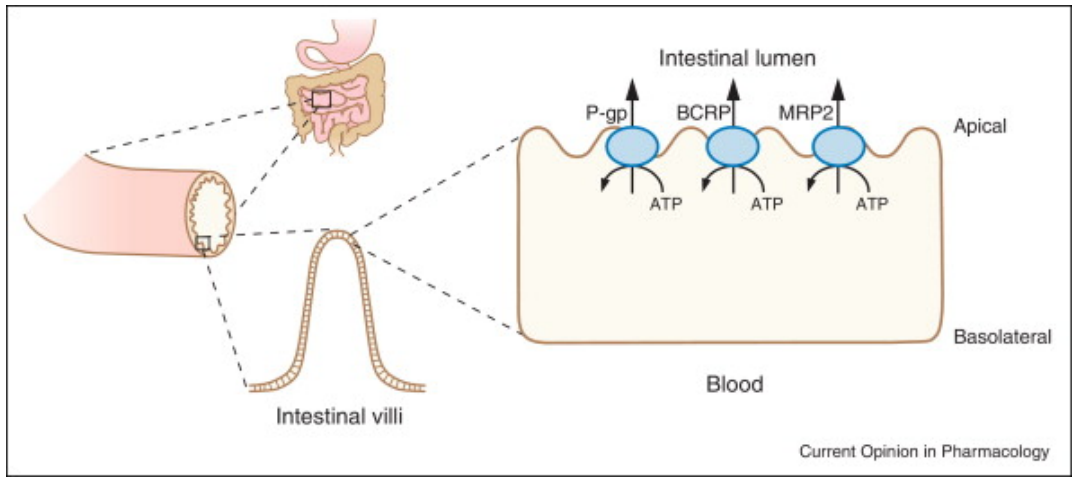
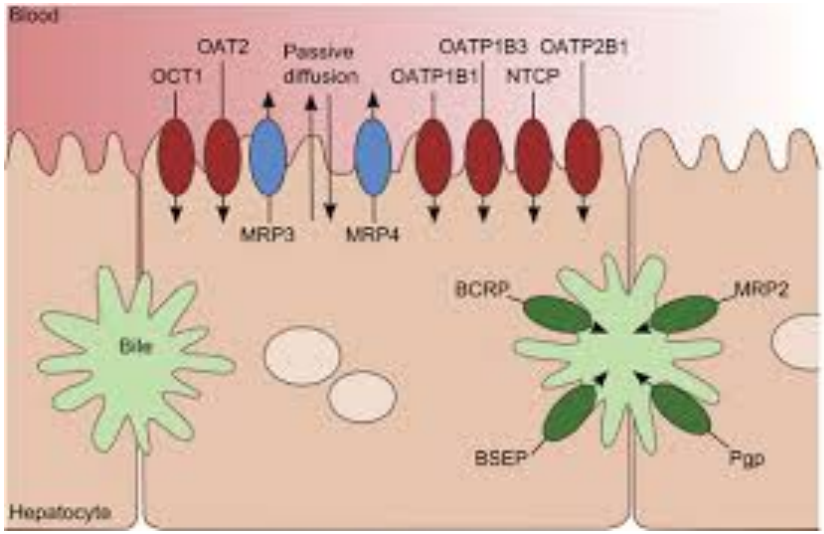
P-glycoprotein (P-gp)

- P-glycoprotein is an **efflux pump** of the ABC (ATP-Binding Cassette) family.



- In the gastro-intestinal system: highly expressed in the intestine and liver





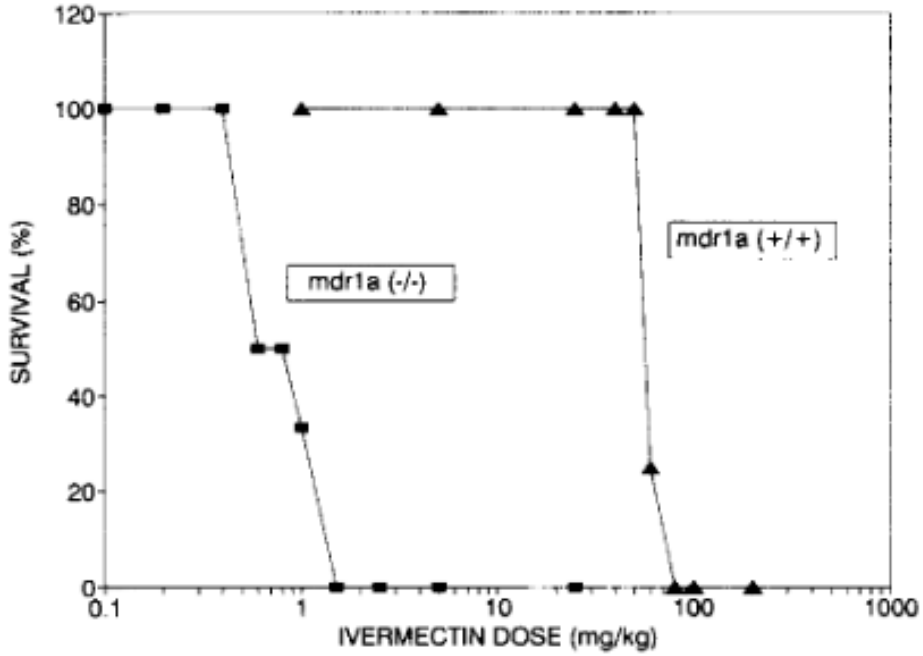
https://www.absorption.com/kc/efflux-transporters/

P-glycoprotein (P-gp)

- P-gp highly influence the pharmacokinetics of numerous xenobiotics.
- Example: neurotoxic pesticide Ivermectin

Table 1. Tissue Concentrations of Ivermectin in *mdr1a* (+/+) and (−/−) Mice 24 hr after Oral Injection of a Dose of 0.2 mg/kg

Tissue	*mdr1a* (+/+)	*mdr1a* (−/−)	Ratio (−/−):(+/+)
Brain	1.5 ± 1.2	131 ± 16	87
Muscle	9.6 ± 3.3	48 ± 3	5.0
Heart	25 ± 10	100 ± 23	4.0
Kidney	47 ± 14	141 ± 27	3.0
Liver	130 ± 45	497 ± 74	3.8
Gall bladder	147 ± 17	1376 ± 804	9.4
Lung	23 ± 6	91 ± 24	4.0
Stomach	63 ± 60	107 ± 46	1.7
Small intestine	31 ± 13	121 ± 30	3.9
Colon	31 ± 12	108 ± 30	3.5
Fat (neck)	188 ± 62	486 ± 78	2.6
Fat (organ)	126 ± 77	152 ± 41	1.2
Testis	9.4 ± 4.2	70 ± 7	7.4
Epididymis	59 ± 20	164 ± 7	2.8
Spleen	13 ± 4	48 ± 10	3.7
Thymus	43 ± 13	121 ± 49	2.8
Plasma	16 ± 6	52 ± 8	3.3



Schinkel et al., Cell, 1994

Systems pharmacology to study P-gp activity

Aim: characterize P-gp activity in the gastrointestinal system according to the animal **sex, feeding condition and circadian status**.

Methods: B6D2F1 male and female mice, fed or fasted for 12h, synchronized to LD12:12 (ZT0 defines light onset).

Outline

1. Study of P-gp expression and activity (talinolol as P-gp substrate)
2. Implications for studying the PK of the anticancer drug irinotecan

Sex-specific P-gp expression

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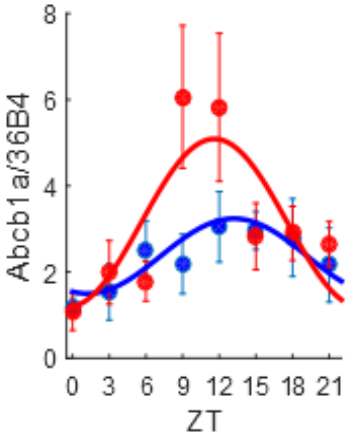

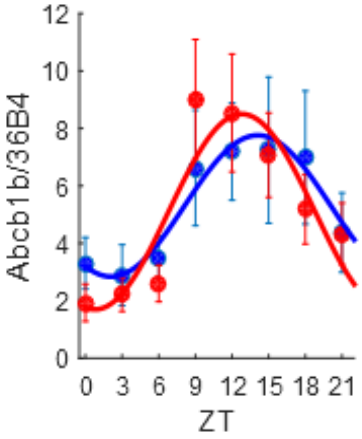

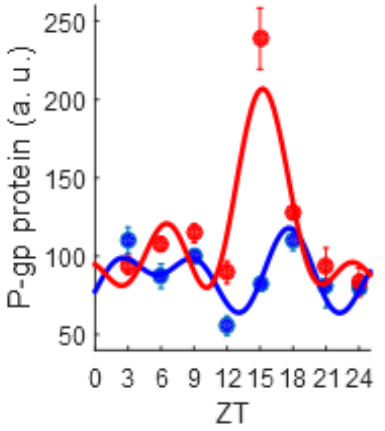

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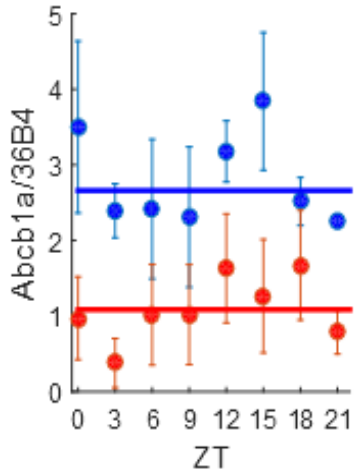
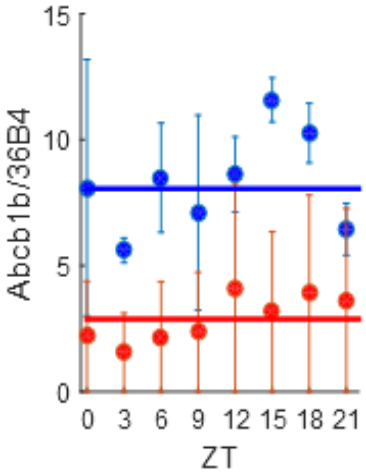
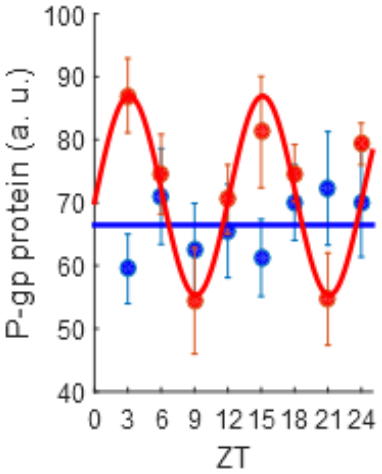
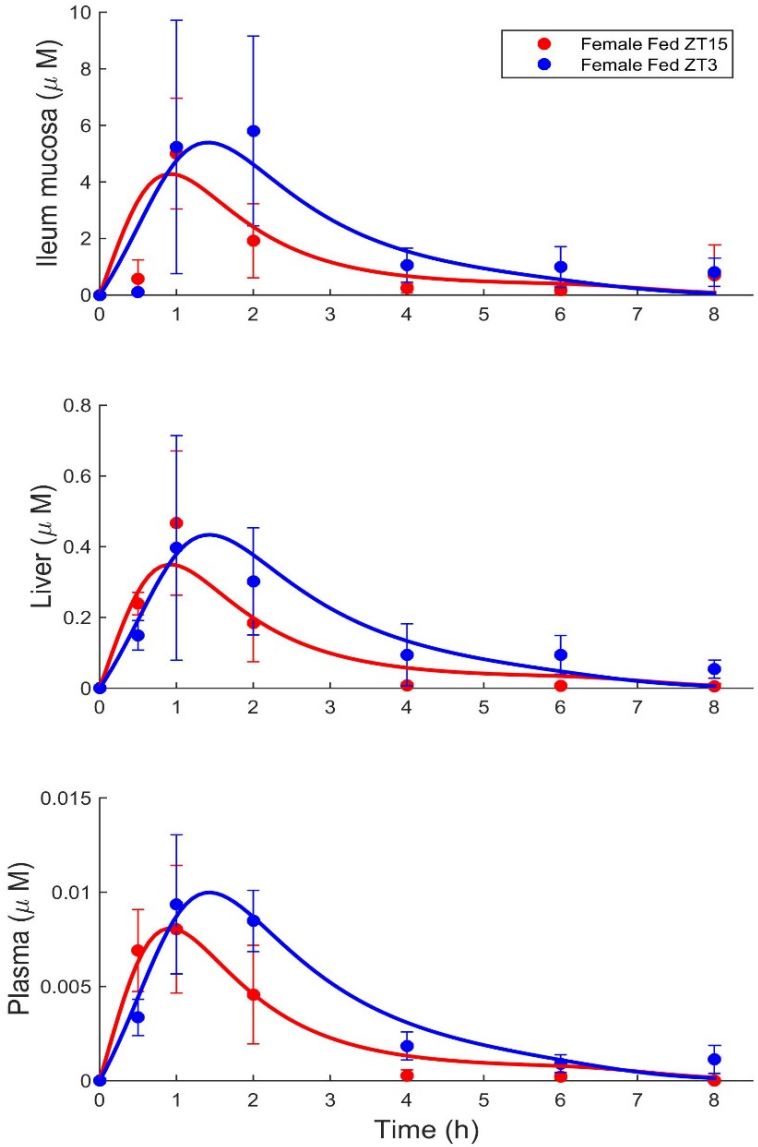
Colon

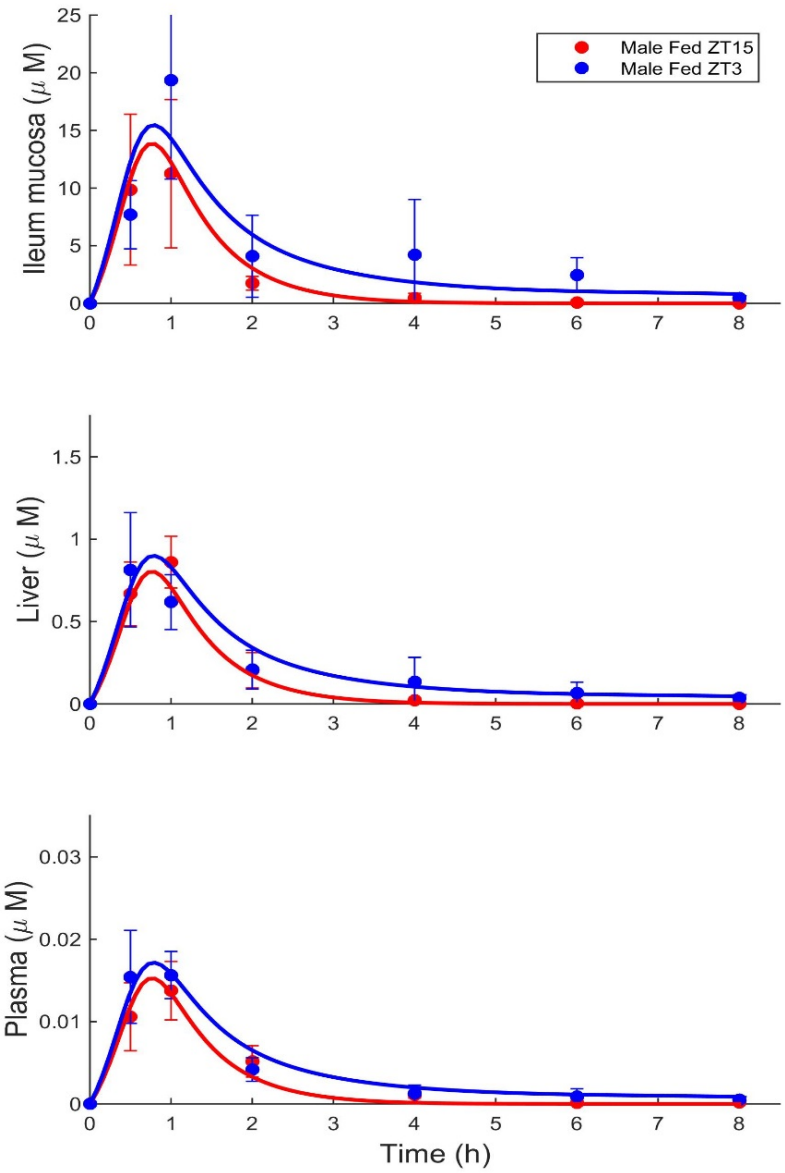




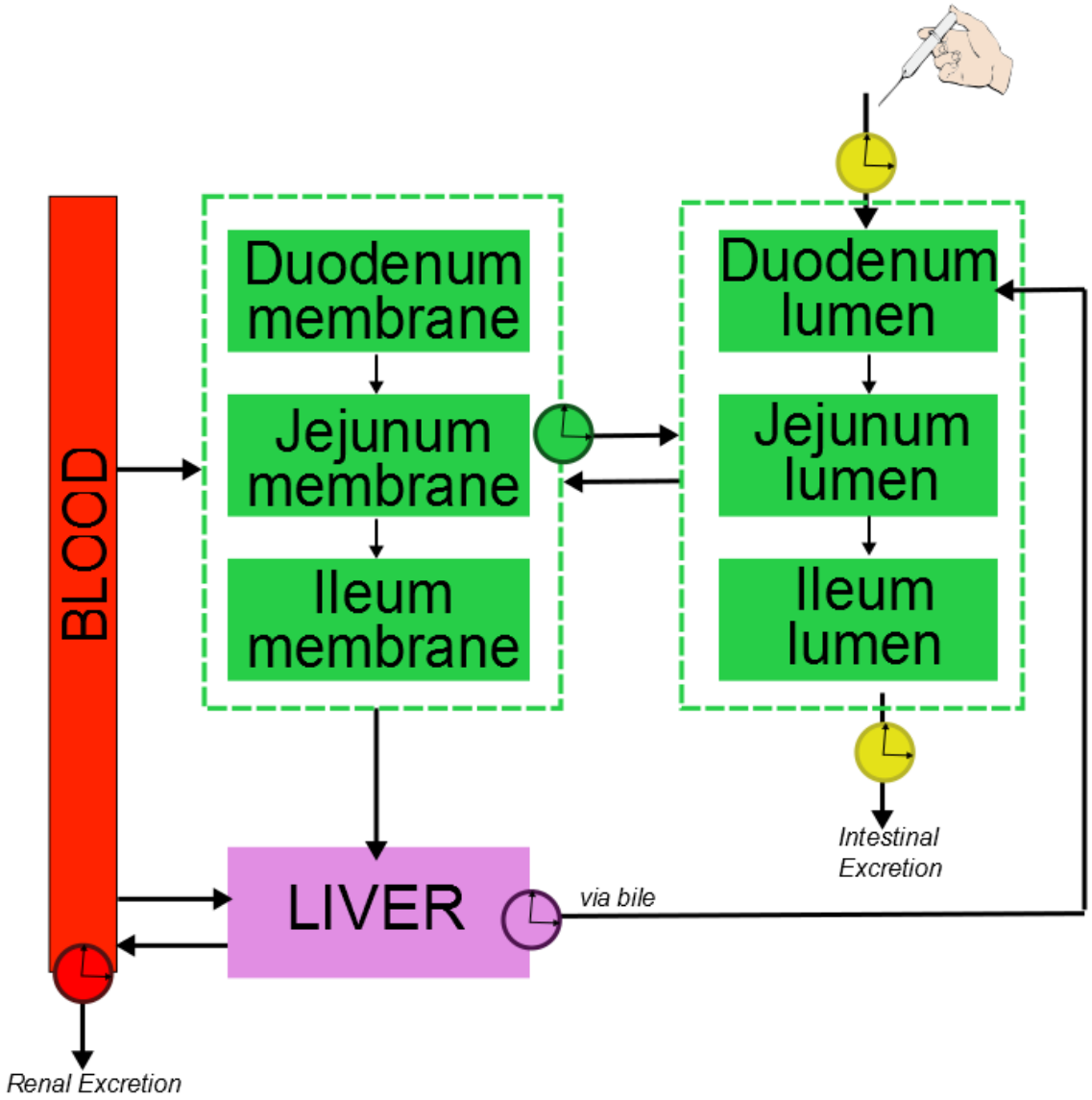


ChronoPK of talinolol, a pure P-gp substrate



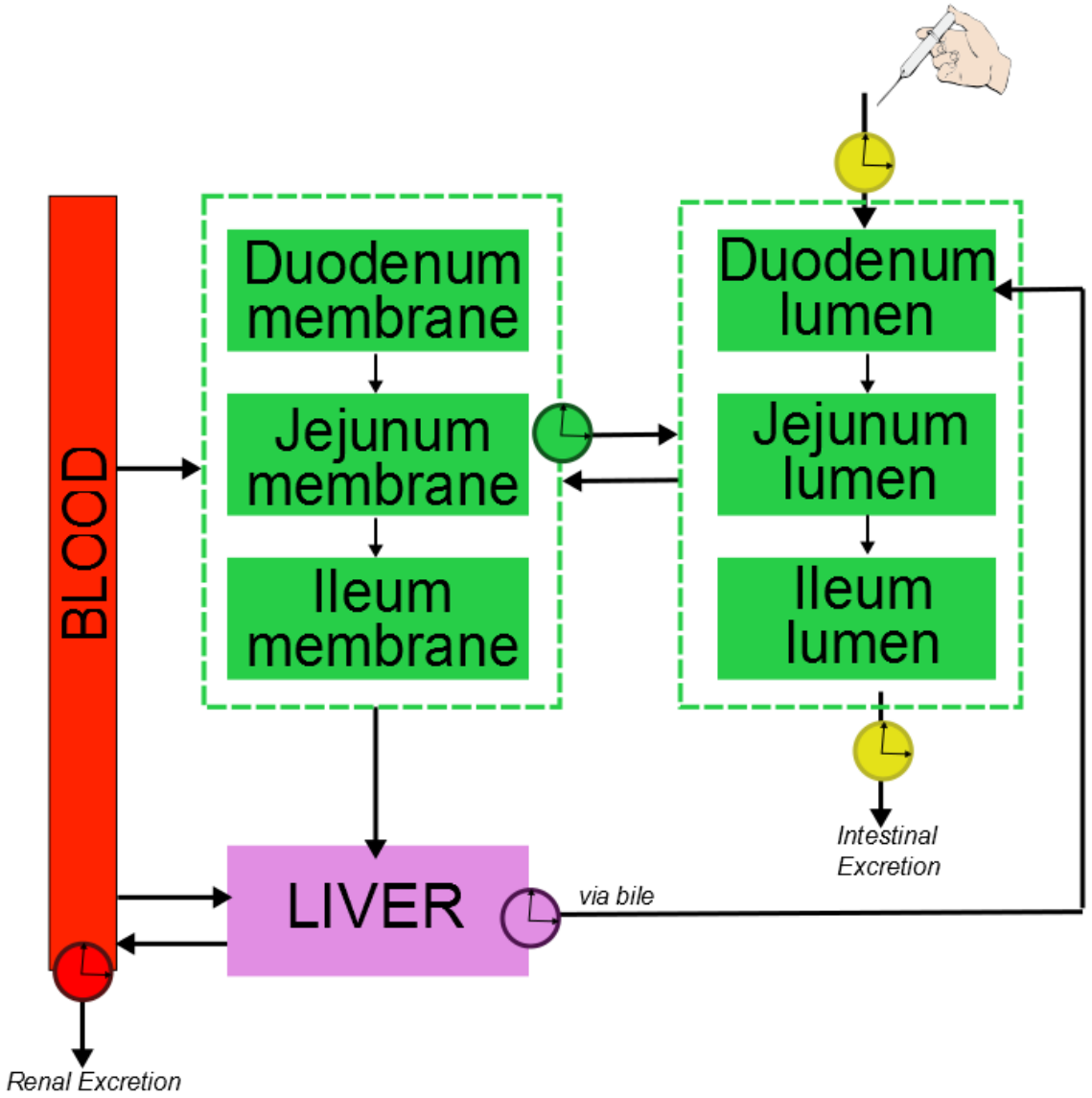


Model of talinolol chronoPK



- ODE-based model,
- calibrated for each mouse category: male/female, fed/fasted.

Model of talinolol chronoPK



- Renal and intestinal drug elimination, intestinal and hepatic P-gp activity assumed to display circadian rhythms
- Model:

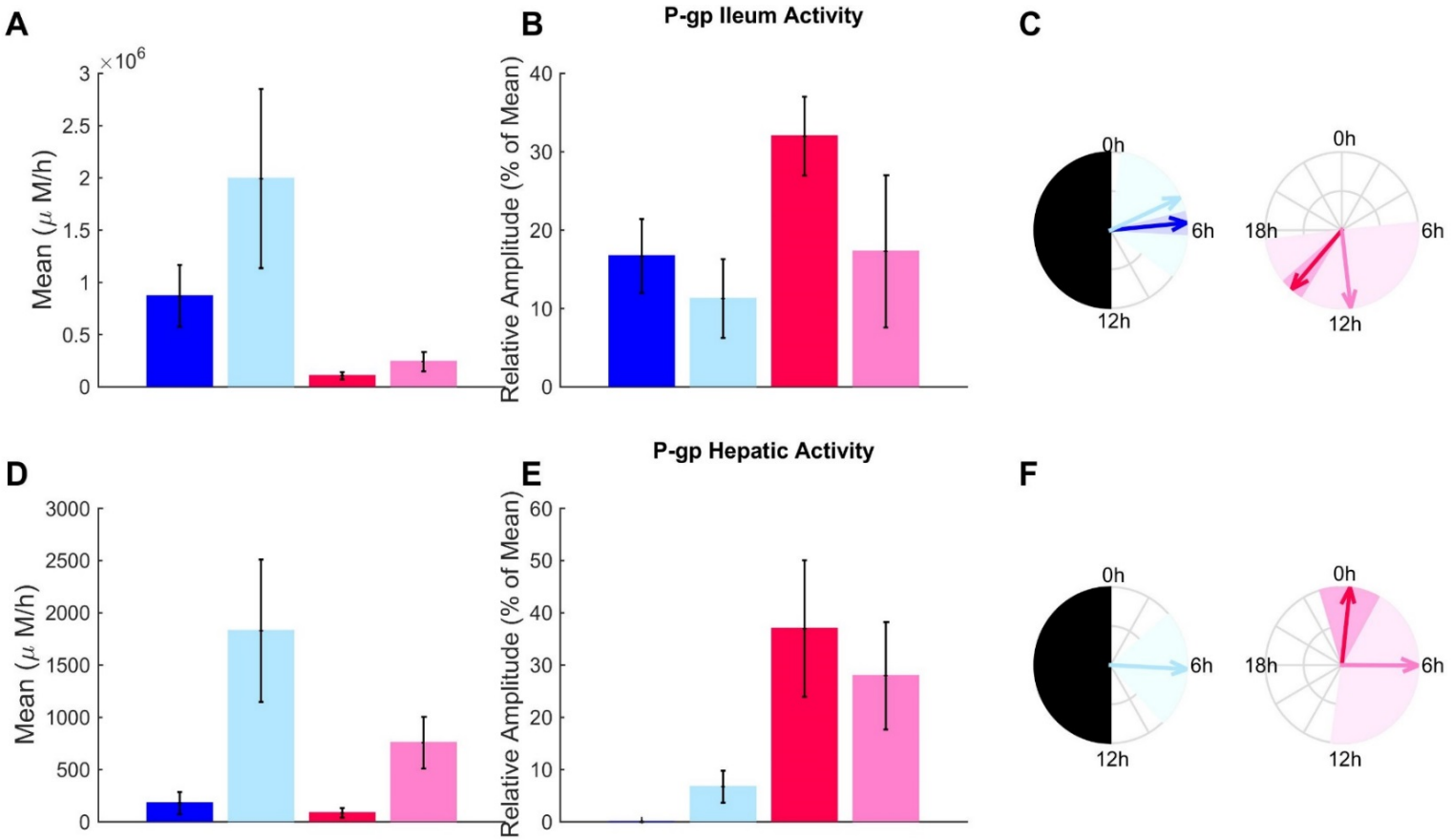
$$M+A\cos(w(t-\varphi))$$

with

M circadian mean
A circadian amplitude
Φ circadian phase

Model-estimated P-gp activity

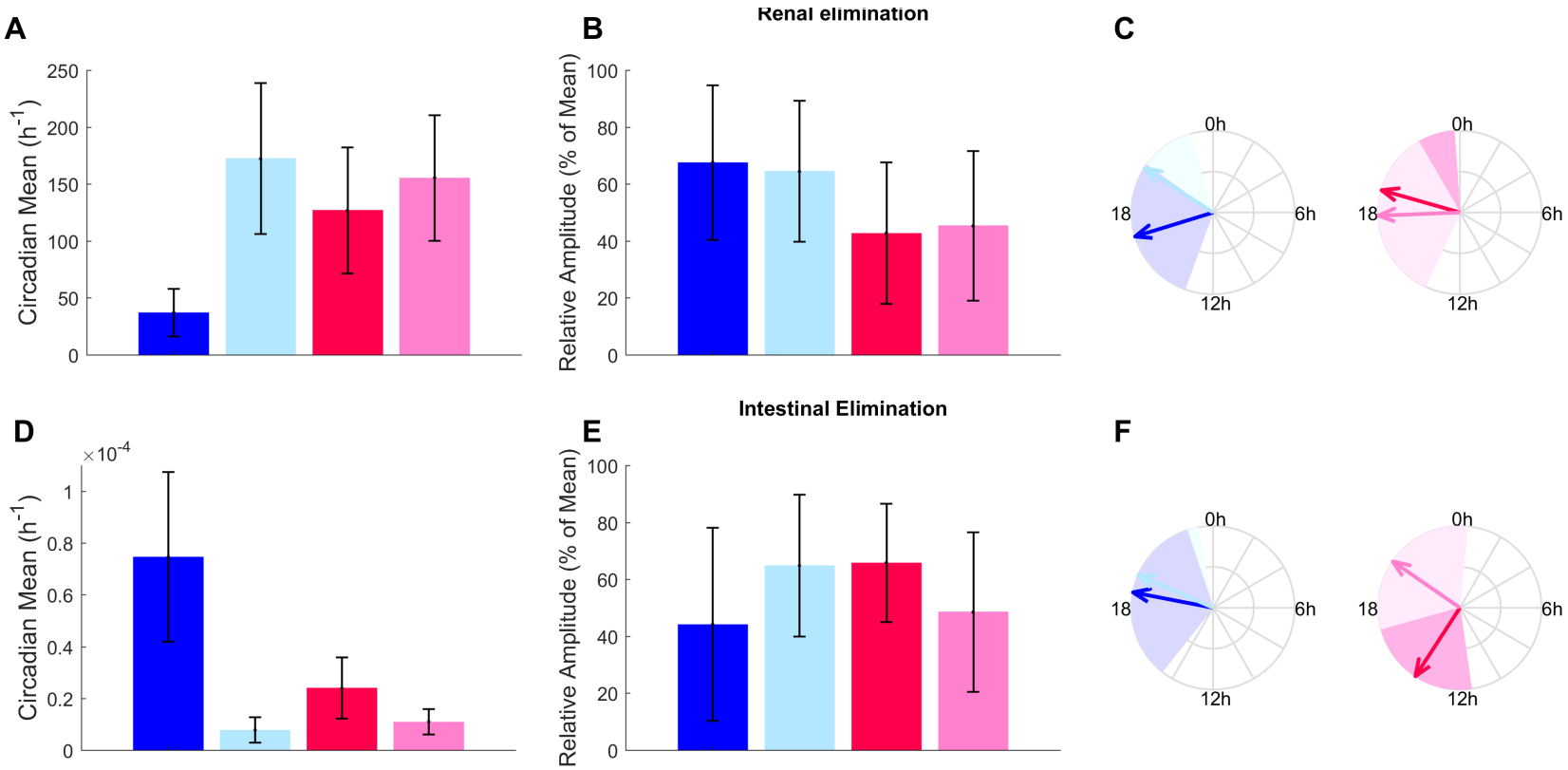
Fed Male
Fasted Male
Fed Female
Fasted Female



- Circadian mean higher in males compared to females.
- Circadian amplitudes consistently higher in females than in males.
- Circadian phases varied by up to 10h with respect to sex.

Model-estimated Renal and intestinal clearance

Fed Male
Fasted Male
Fed Female
Fasted Female



- High Circadian amplitudes for both elimination routes.
- Circadian phases in the activity phase (dark).

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La science pour la santé
From science to health

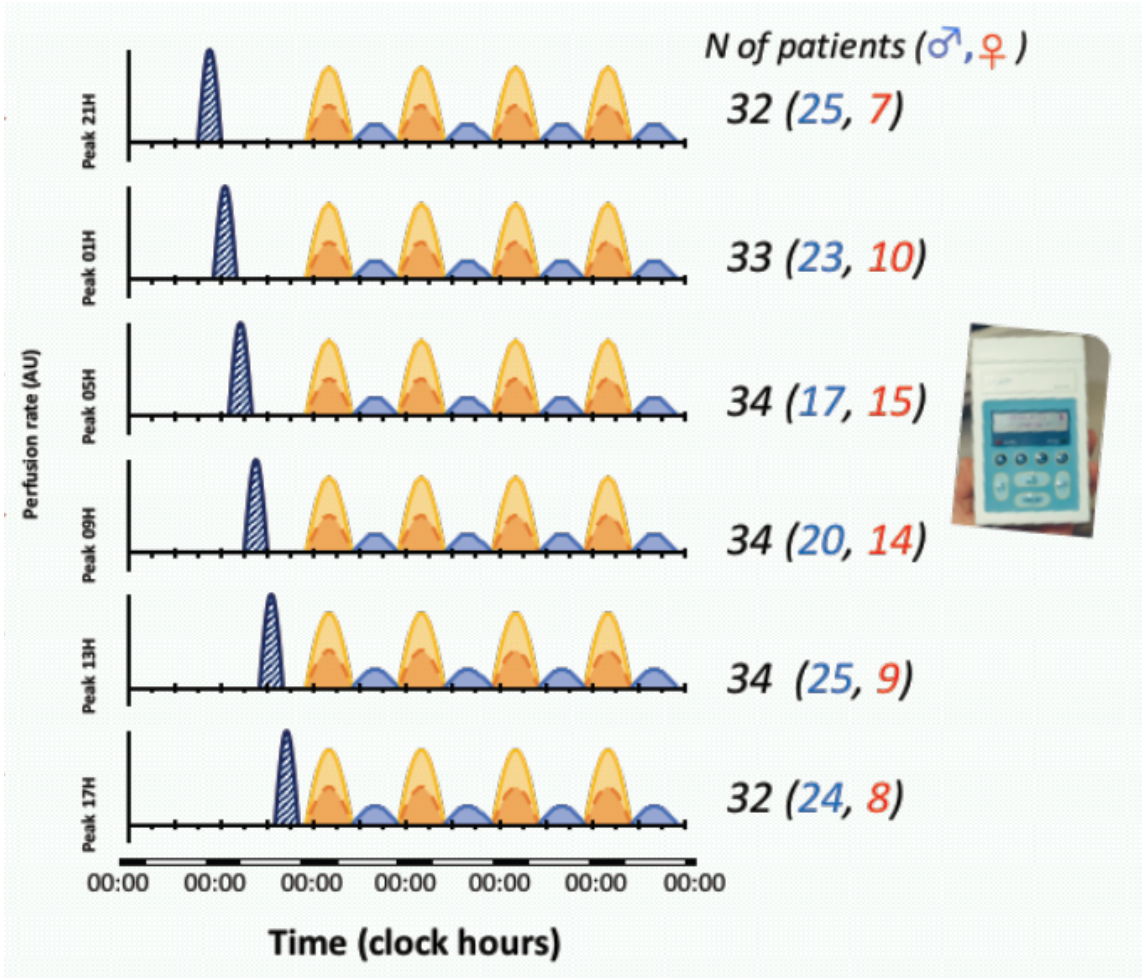
Implications for Irinotecan pharmacokinetics

Irinotecan

- Irinotecan= 1 of the 3 cytotoxic drugs of gold-standard chemotherapies against digestive cancers.
- Responsible for dose-limiting toxicities
- Irinotecan pharmacokinetics and toxicities depend on sex and circadian timing in mice and in cancer patients

Gender-specific chronotoxicity of irinotecan in patients

- 199 metastatic colorectal patients received 5-fluorouracil, oxaliplatin and irinotecan as 1st or 2nd line.
- 6 peak admin times for irinotecan: 1am, 5am, 9am, 1pm, 5pm, 9pm.
- Peak times set for 5-fluorouracil (4am) and oxaliplatin (4pm); over 4 days.
- Chronomodulated drug delivery through programmable pumps.



Innominato et al., in preparation

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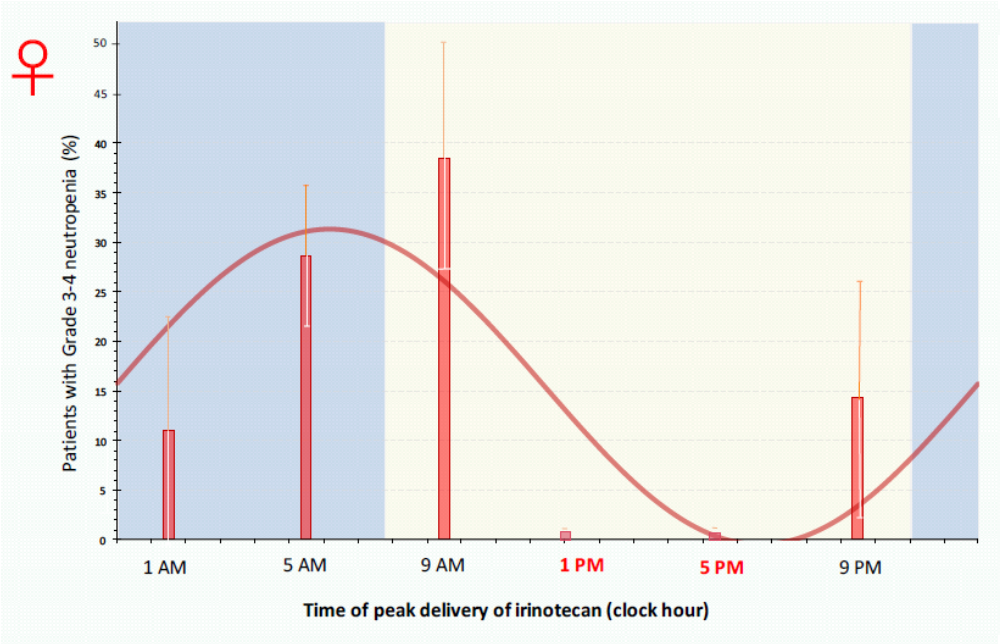
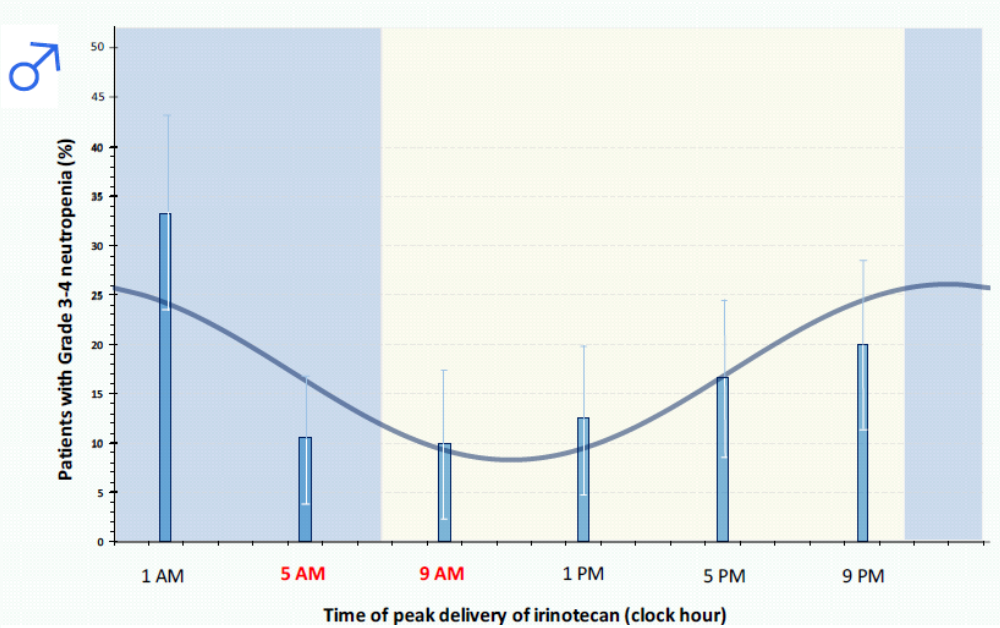
Gender-specific chronotoxicity of irinotecan in patients





Large amplitude

Optimal timing in the afternoon

Moderate amplitude

Optimal timing in early morning

Innominato et al., in preparation

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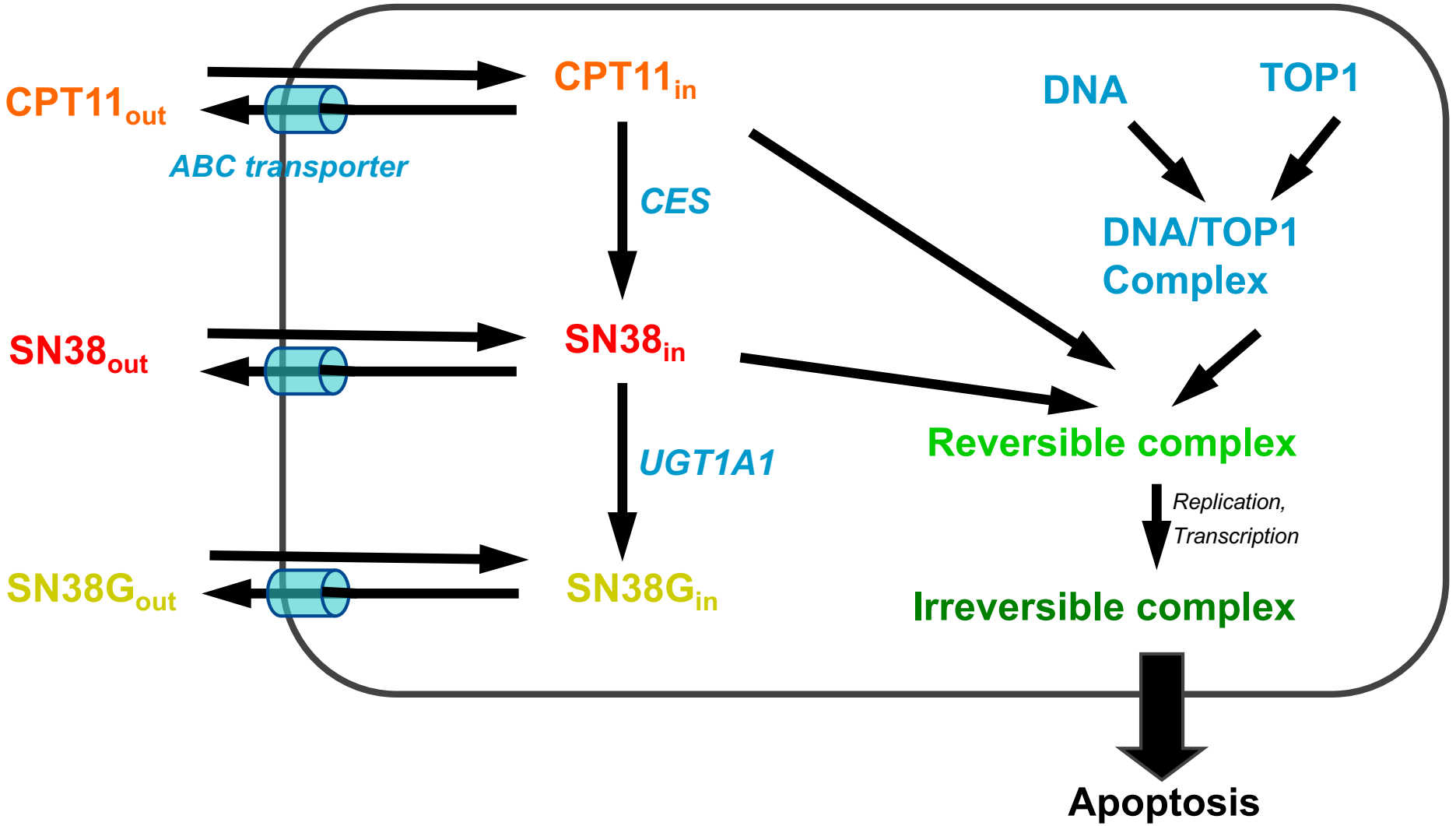
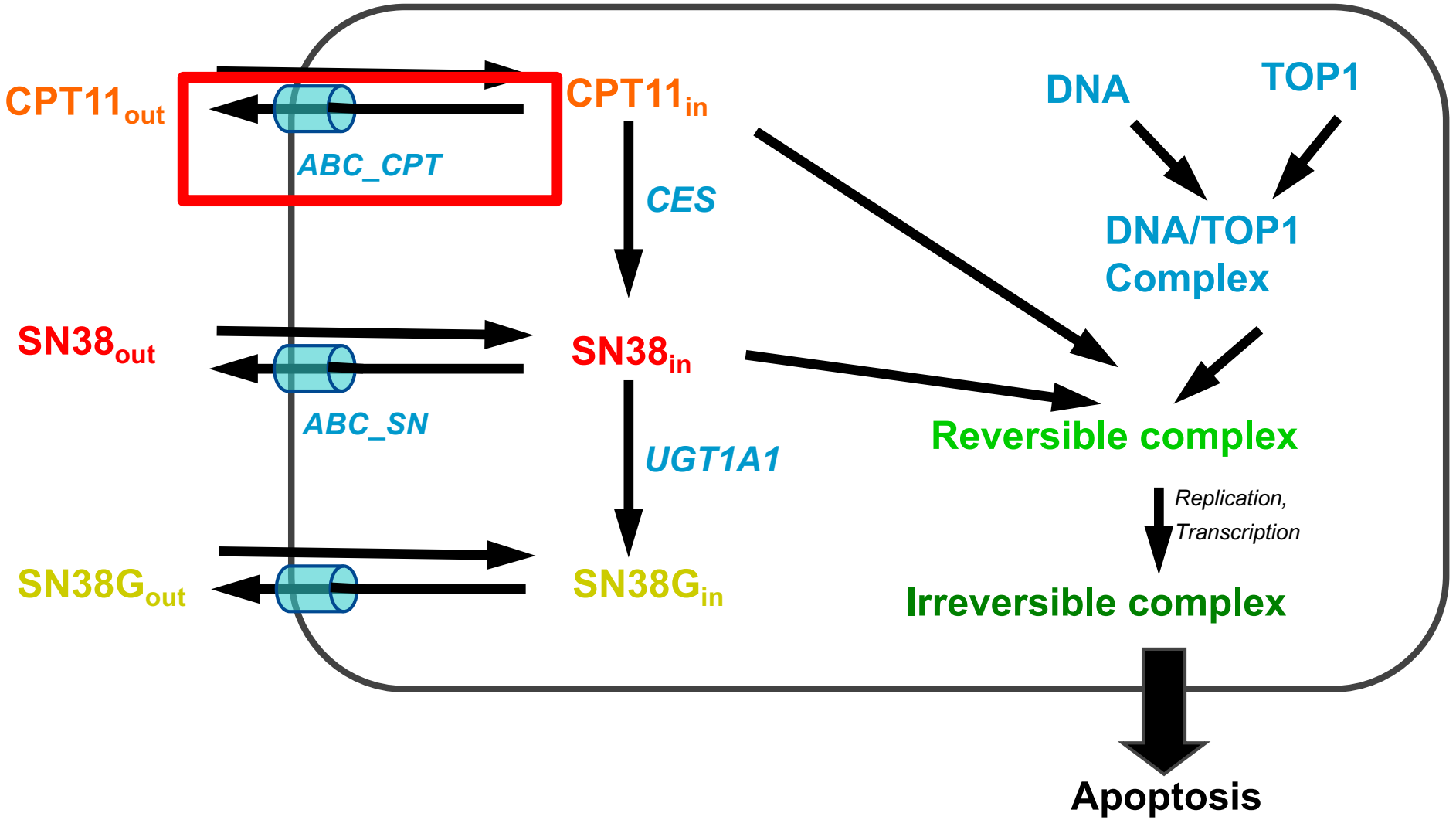
Irinotecan cellular chronoPK-PD model

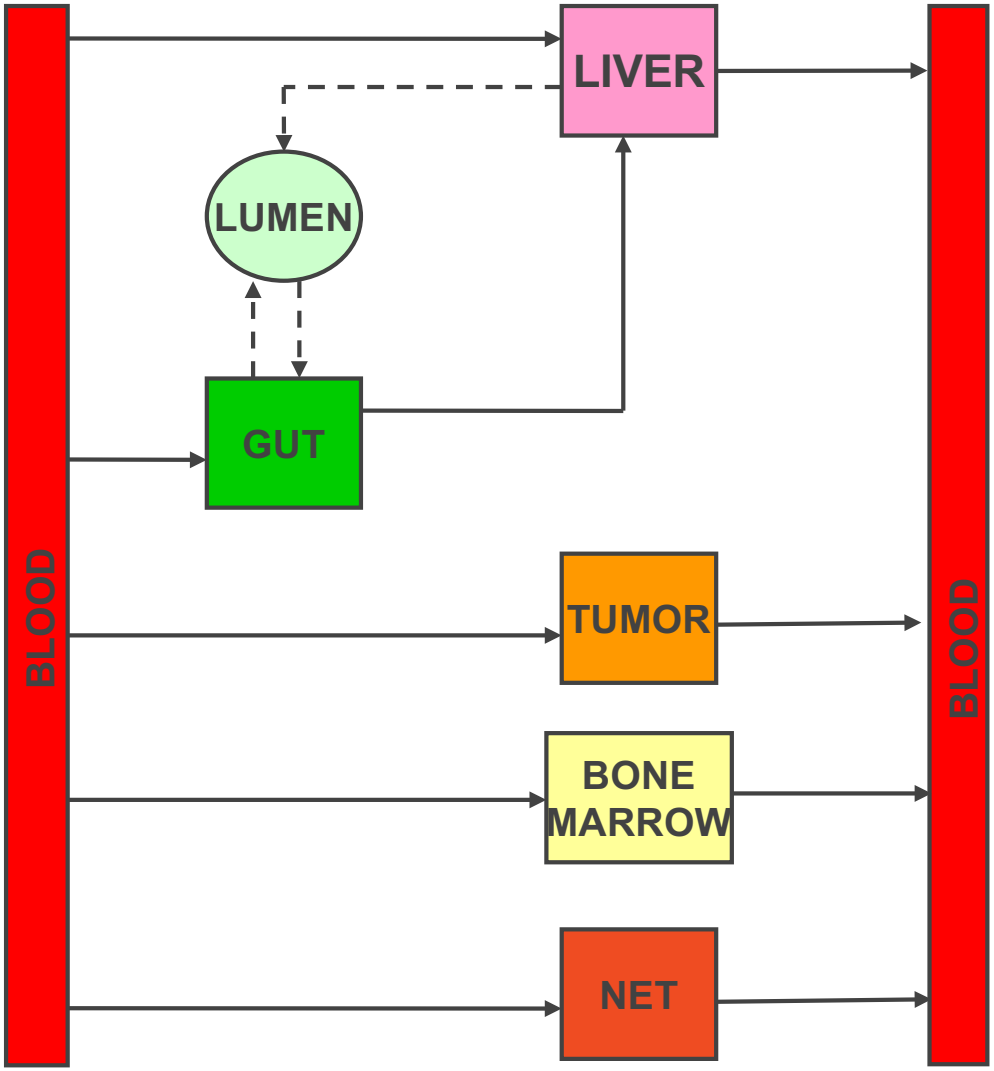


Irinotecan= CPT11

Focus on P-glycoprotein (P-gp)



Physiologically-based model of irinotecan PK-PD

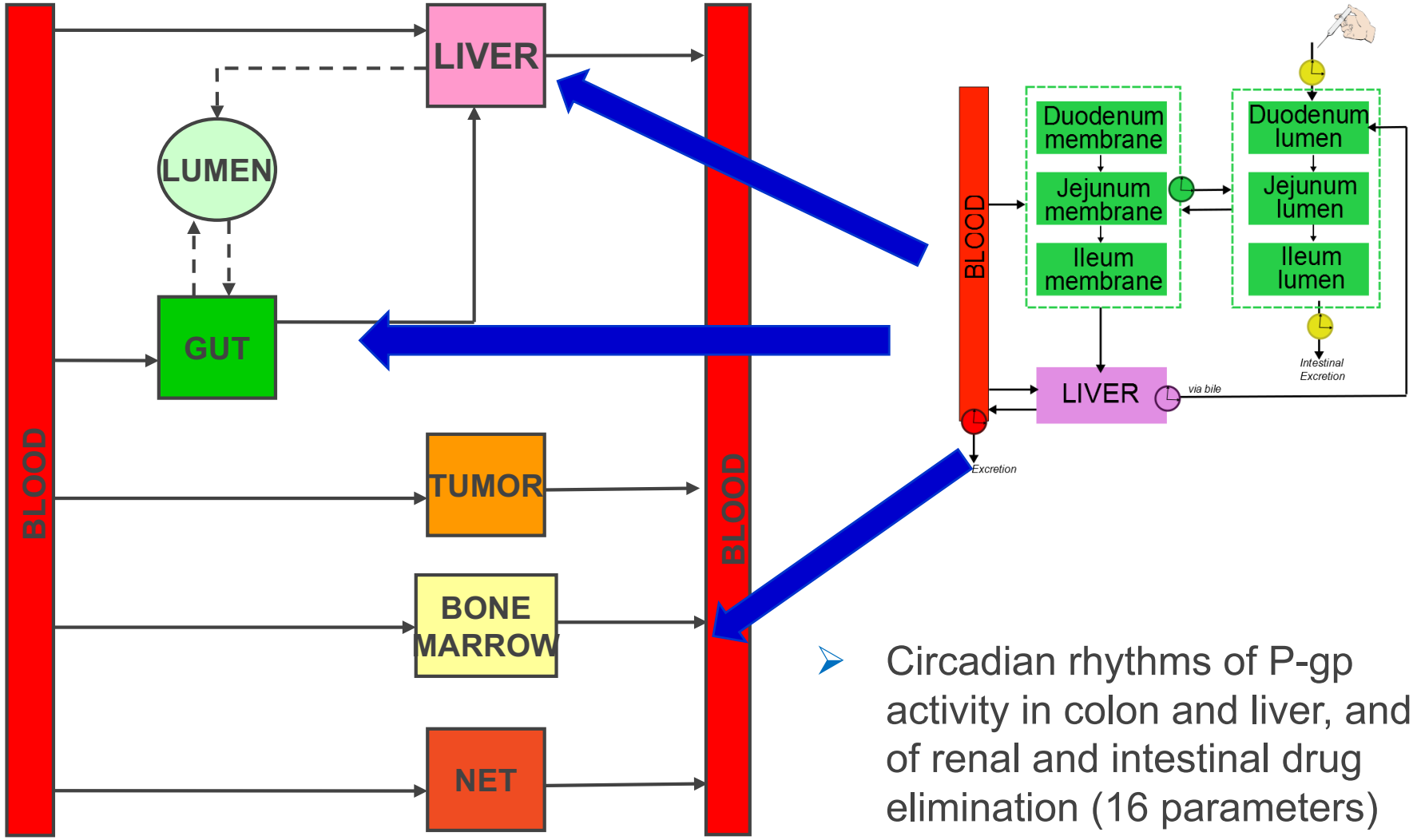


Aim: whole-body model of irinotecan PK-PD and their circadian control, sex-specific

For: investigating molecular determinants of drug chronotoxicity; designing optimal circadian infusion schemes.

How: Ordinary differential equations based on law of mass action, Michaelies-Menten kinetics, First Fick's law for passive transport.

Irinotecan sex-specific chronoPK-PD in mice

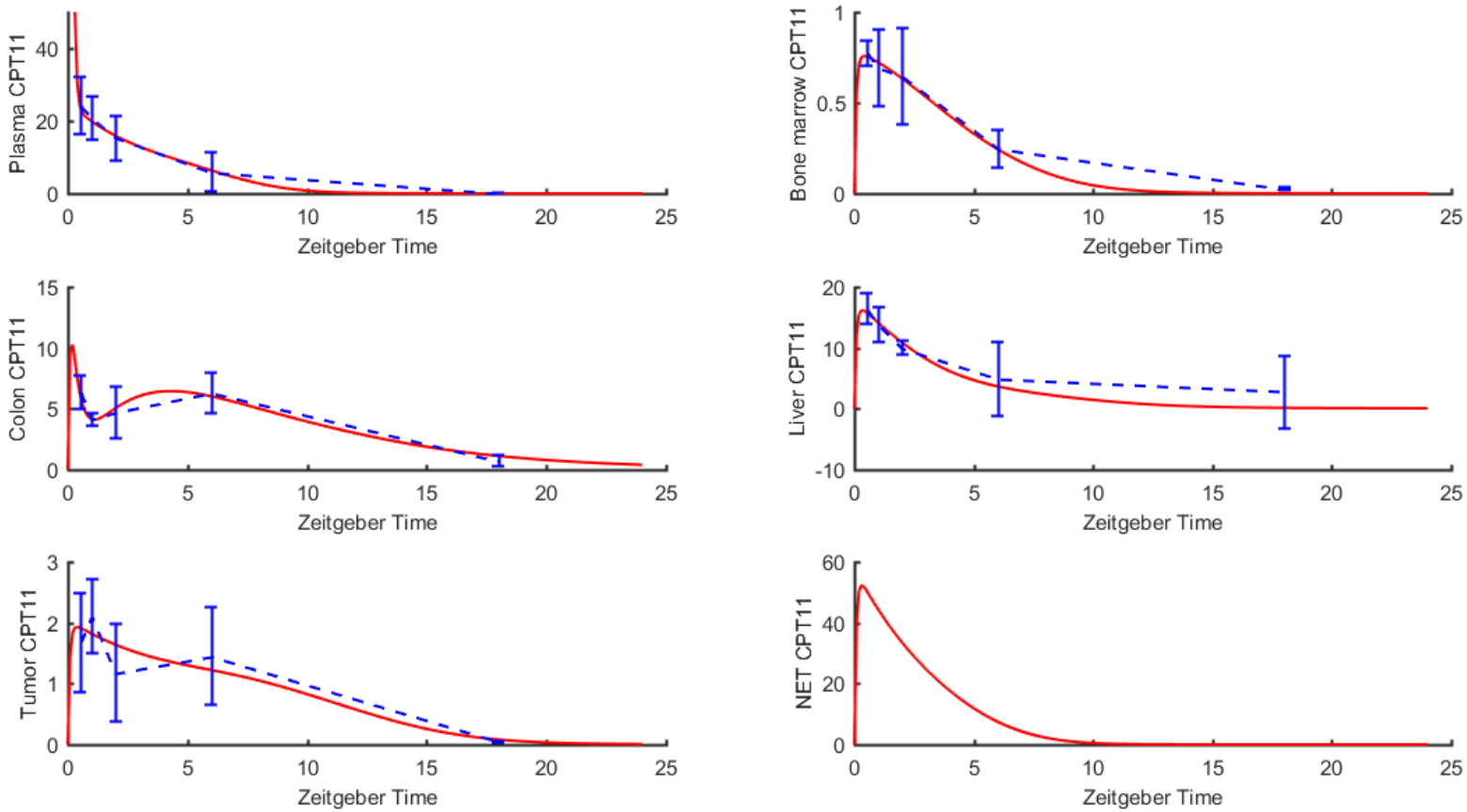


- Circadian rhythms of P-gp activity in colon and liver, and of renal and intestinal drug elimination (16 parameters)

Ballesta et al. Springer, 2012

Irinotecan sex-specific chronoPK-PD in mice

- Ongoing: Fit irinotecan model to chronoPK data and circadian protein levels for male/female B6D2F1 mice



Irinotecan chronoPK in B6D2F1 male mice, *during CPT11 exposure at Best time of tolerability (ZT11).*

Conclusions

- P-gp expression and activity **highly depends on sex, organ, circadian timing and feeding conditions**
- P-gp systems pharmacology study allow quantification of physiological parameters for each mouse category and organ
- **Critical information for physiologically-based modeling ->** here used for irinotecan
- Future: using irinotecan chronoPK-PD model to investigate molecular determinants of chronotoxicity rhythms

Collaborations

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Narin Ozturk

Zeliha Pala

North Wales Cancer Treatment Centre, Department of Oncology, Bodelwyddan, UK

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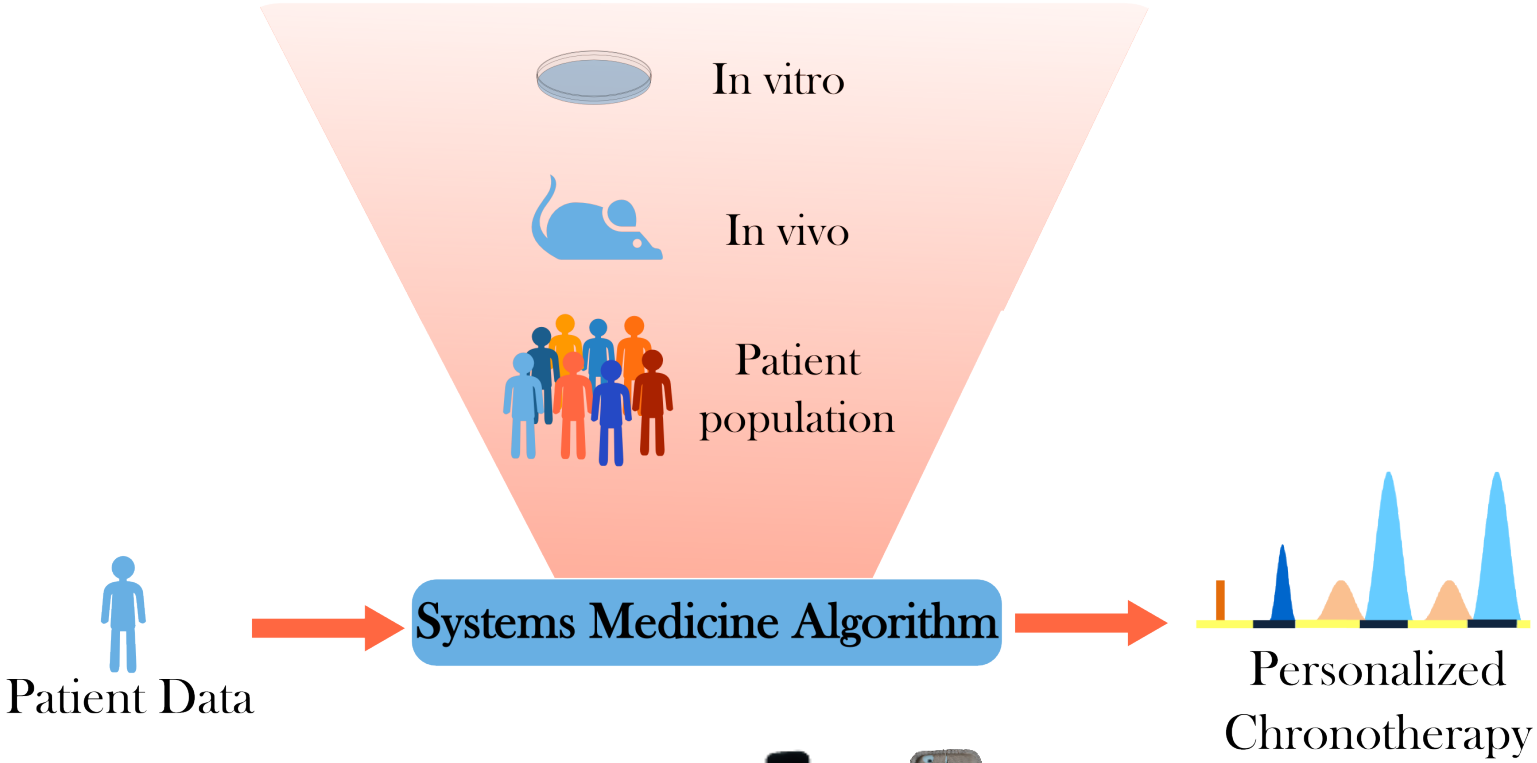
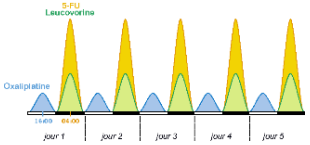
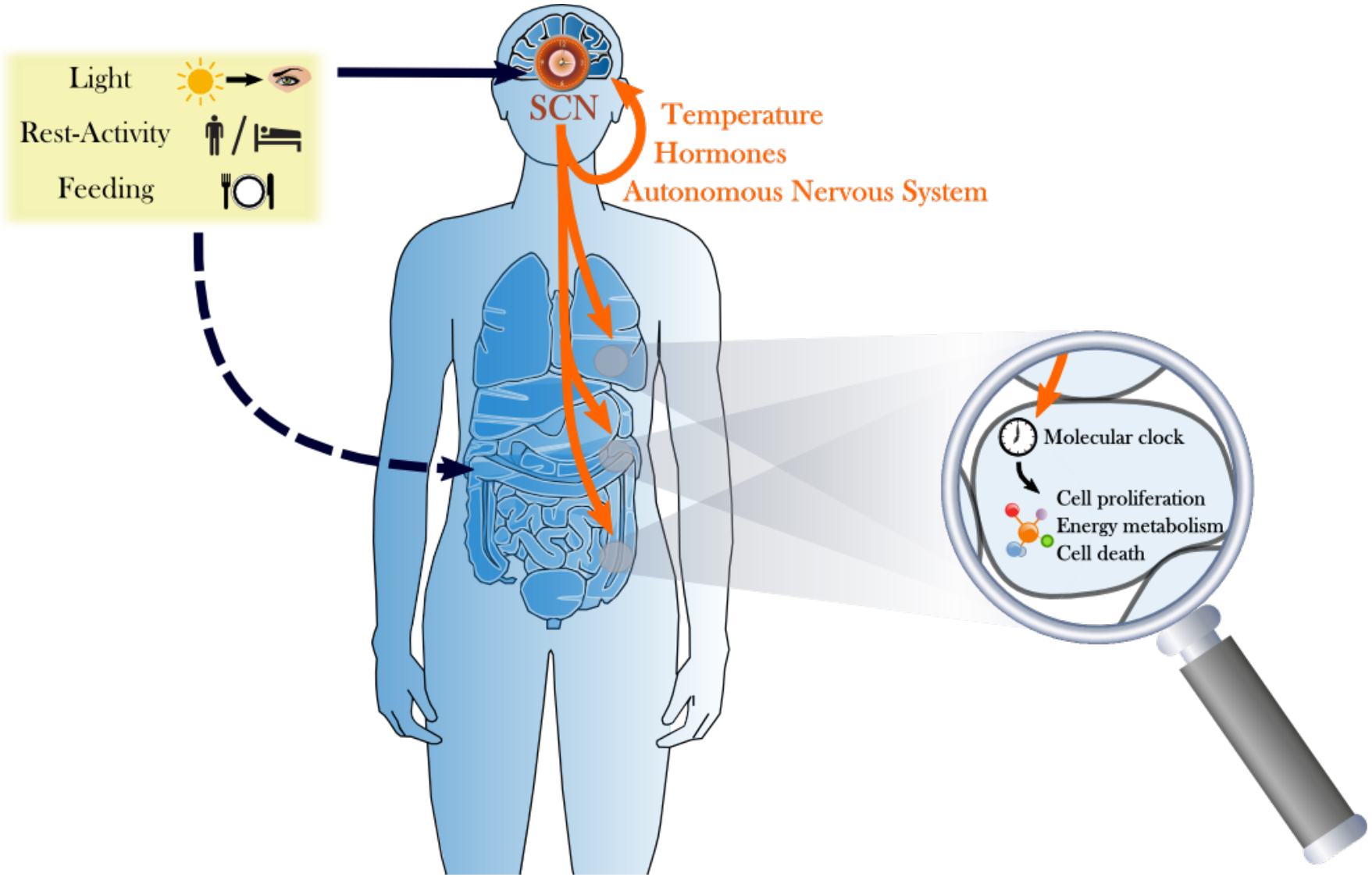
Future



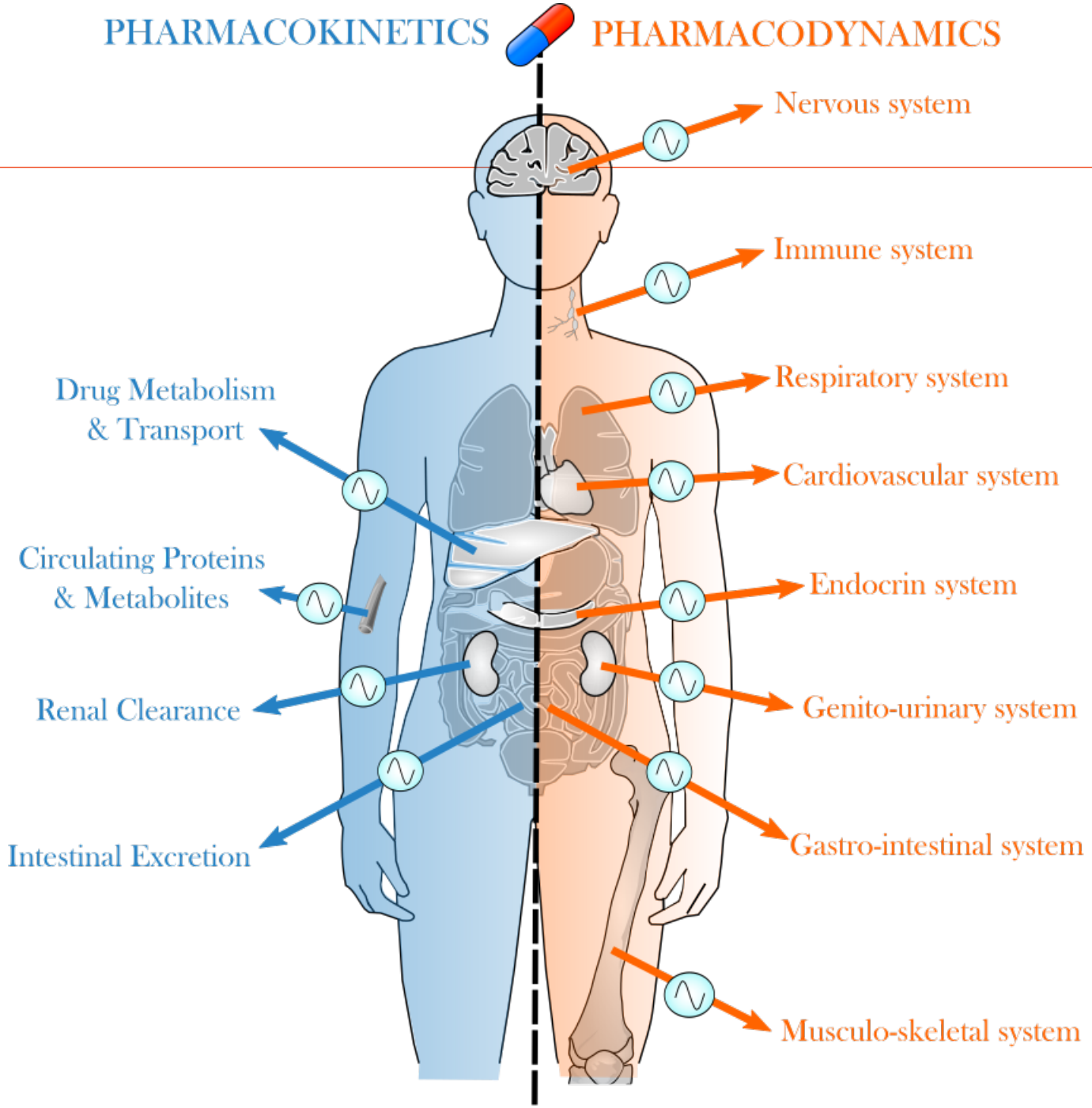
- Non-invasive sensors: rest-activity, skin surface temperature, position
- Salivary, urinary, blood sampling: cortisol, melatonin, multi-omics.
- Covariates: sex, age, gene polymorphisms



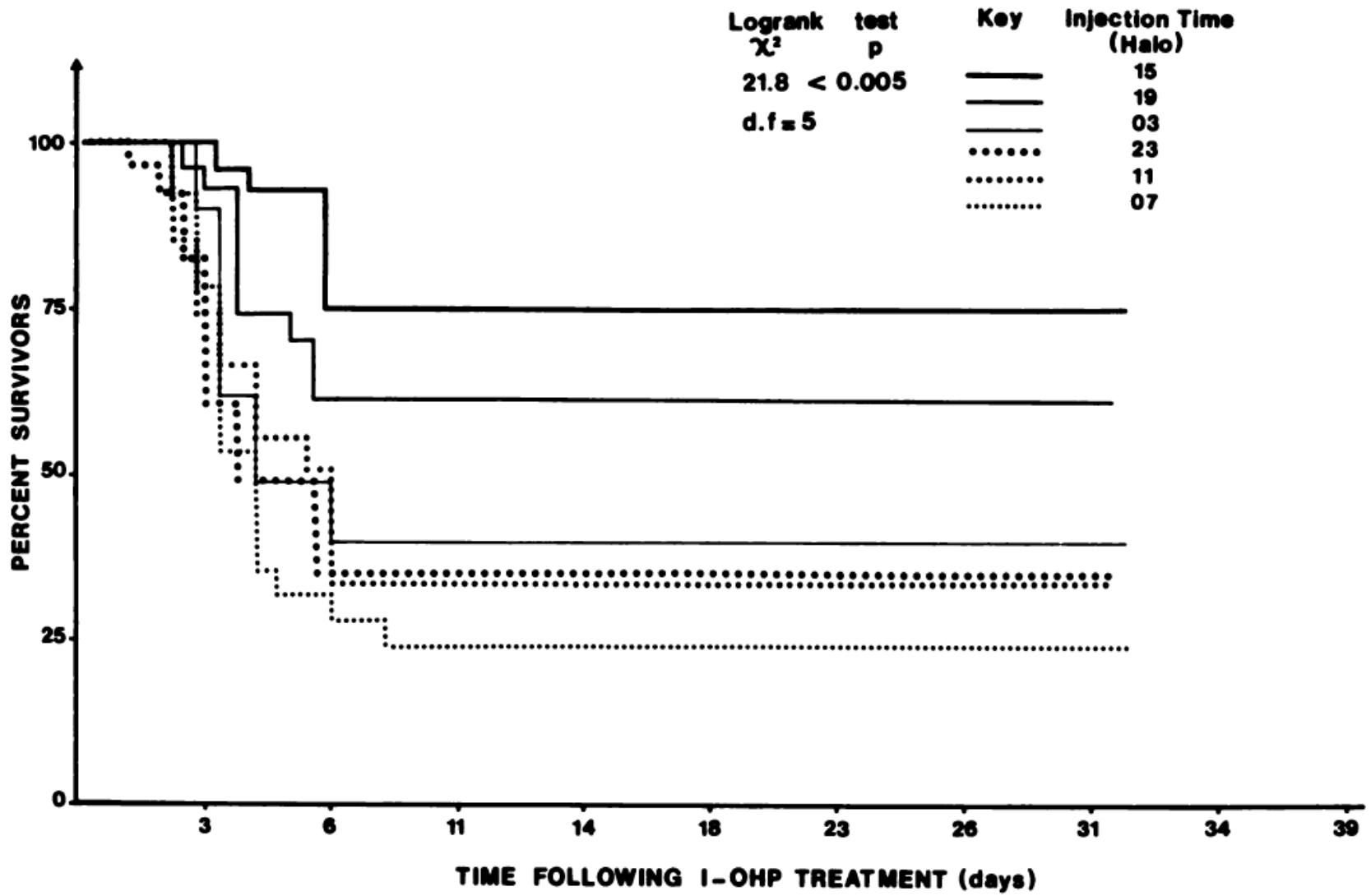
The Circadian Timing System



Ballesta et al., Pharmacological Reviews, 2017


PHARMACOKINETICS
PHARMACODYNAMICS
Nervous system
Immune system
Respiratory system
Cardiovascular system
Endocrin system
Genito-urinary system
Gastro-intestinal system
Musculo-skeletal system
Drug Metabolism & Transport
Circulating Proteins & Metabolites
Renal Clearance
Intestinal Excretion

Oxaliplatin chronotoxicity in mice



Anticancer drug chronotoxicity in preclinical models

Antimetabolites
- Gemcitabine
- L-alanosine

Topoisomerase inhibitors
- Irinotecan
- Mitoxantrone
- Etoposide

Intercalating agents
- Theprubicin

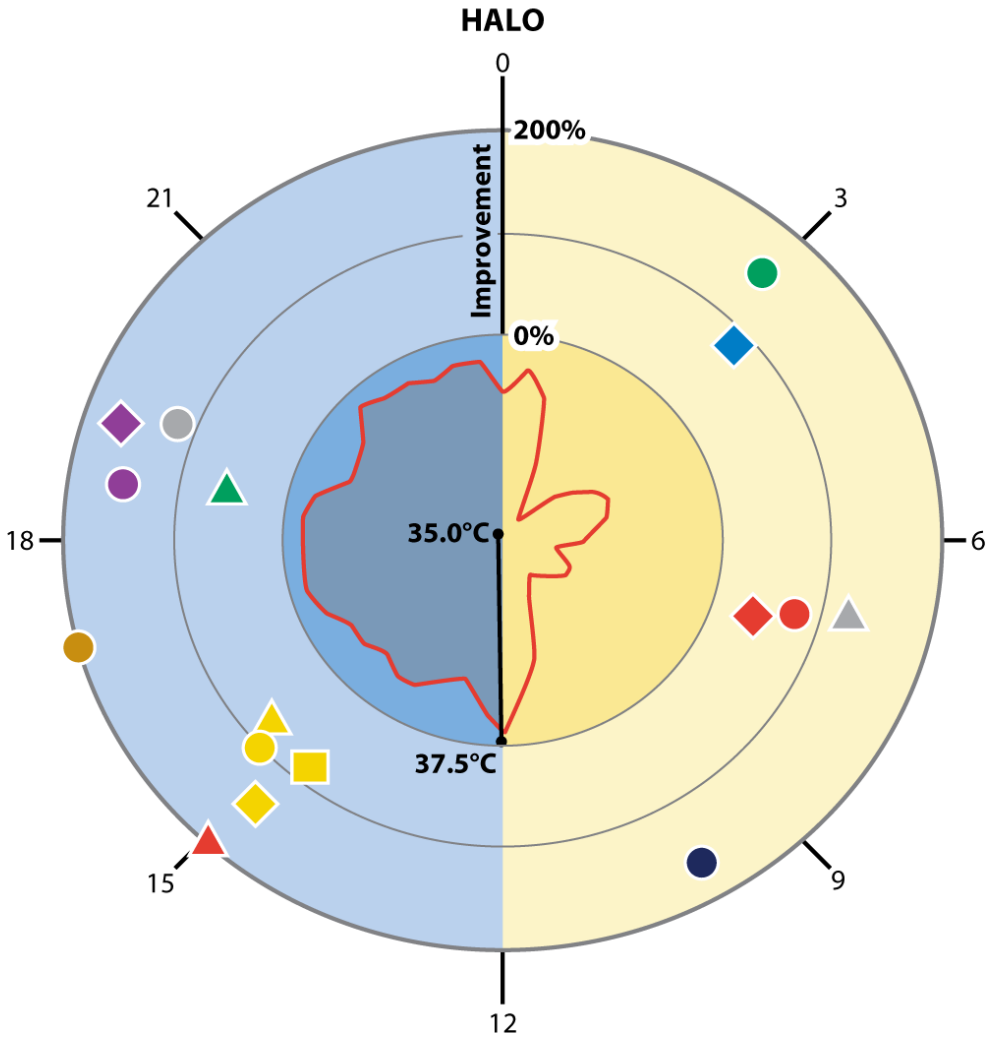
Alkylating agents
- Peptichemio
- Cisplatin
- Carboplatin
- Oxaliplatin

Nitrosoureas or related
- Mitomycin-C
- Cystemustin

Mitotic spindle poisons
- Docetaxel
- Vinorelbine

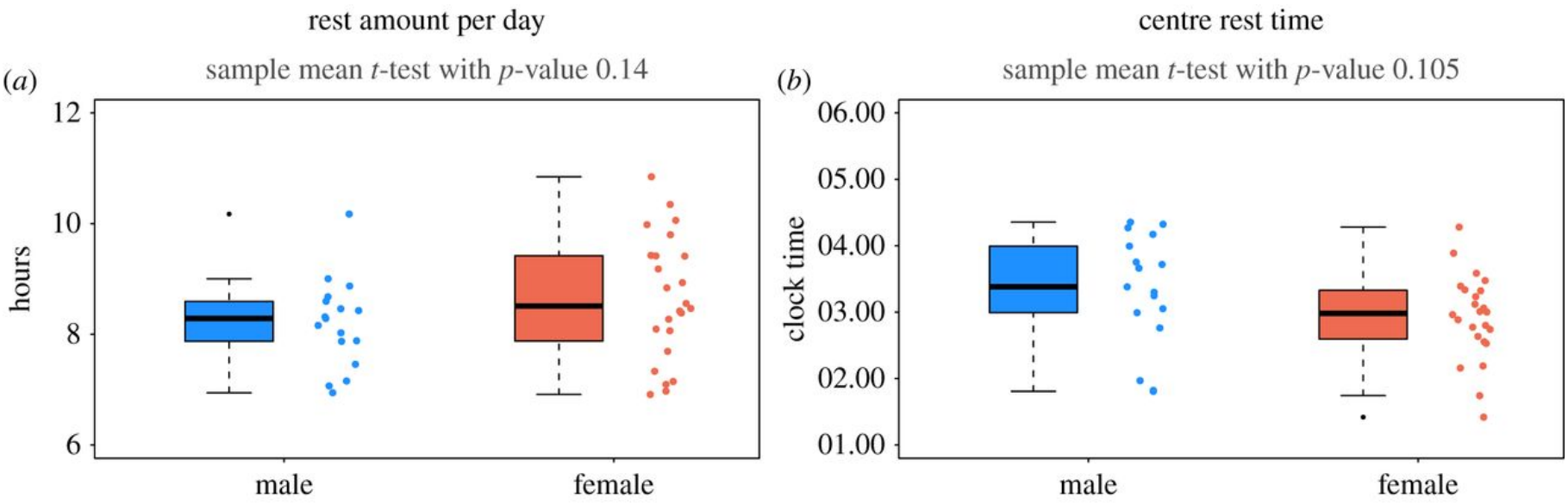
Cytokines
- Interleukin-2

Small kinase inhibitors
- Seliciclib



- **Chronotoxicity** demonstrated for 40 anticancer agents
- **Chronoefficacy** for 28 agents.

Large inter-patient variability in circadian rhythms



Analysis of rest-activity of 46 healthy individuals (19 male, 27 female; aged 21–75 years, median 35 years), wearing sensors for 4-7 days.