WORKSHOP ON PHARMACOKINETICS-PHARMACODYNAMICS OF ANTICANCER DRUGS: RESISTANCES AND SYNERGIES, *PARIS, DECEMBER 18-19, 2008*

Towards optimisation of cancer chronotherapeutics by taking into account patient-specific constraints: Mathematical models for individualised medicine

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http://www-c.inria.fr/bang/JC/Jean_Clairambault_en.html

Outline of the talk

1. Modelling (circadian) pharmacokinetics-pharmacodynamics (PK-PD)
…at the molecular level in blood and in tissues

2. Modelling cell proliferation and its control mechanisms
…at the level of cell populations in tissues

3. Modelling the circadian system and its disruptions
…at both the central and peripheral levels

4. Optimising chronotherapeutics: objectives and constraints

5. Individualising treatments: identifying patient-specific parameters

1. Action of classical cytotoxic drugs:

(*5FU, Oxaliplatin, Irinotecan*)

Pharmacokinetic-pharmacodynamic (PK-PD) modelling

Ordinary differential equations (ODEs)

Molecular PK-PD modelling in oncology

"Pharmacokinetics is what the organism does to the drug,
Pharmacodynamics is what the drug does to the organism"

- *Input*: an intravenous [multi-]drug infusion flow
- Drug concentrations in blood *and tissue* compartments (PK)
- Control of targets on the cell cycle *in tissues* (cell population PD)
- *Output*: a resulting growth rate in tumour and healthy tissues
- *Optimisation* = decreasing proliferation in tumour tissues while maintaining normal proliferation in healthy tissues

Example: 5FU (with drug resistance) + Leucovorin

P = Plasma [5FU]

F = Intracellular [FdUMP]

Q = Plasma [LV]

L = Intracellular [LV]

N = [nrf2] efflux Nuclear Factor

A = ABC Transporter activity

S = Free [TS] (not FdUMP-bound)

B = [FdUMP-TS] binary complex

T = [FdUMP-TS-LV] irreversible ternary complex

$$\frac{dP}{dt} = -k_0 P - \frac{aP}{b+P} - l_{DPD}\frac{P}{m_{DPD}+P} + \frac{i(t)}{V}$$

$$\frac{dF}{dt} = \frac{a}{\xi}\frac{P}{b+P} - \frac{AF}{c+F} - k_1 FS + k_{-1}B$$

$$\frac{dQ}{dt} = -k_2 Q + \frac{j(t)}{V}$$

$$\frac{dL}{dt} = \frac{k_2}{\xi}Q - k_3 L - k_4 BL$$

$$\frac{dN}{dt} = \frac{\kappa F^n}{\lambda^n + F^n} - \mu N$$

$$\frac{dA}{dt} = \mu N - \nu A$$

$$\frac{dS}{dt} = -k_1 FS + k_{-1}B + \theta_{TS}(S_0 - S)$$

$$\frac{dB}{dt} = k_1 FS - k_{-1}B - k_4 BL$$

$$\frac{dT}{dt} = k_4 BL - v_T T$$

Input = 5FU infusion flow

Output = blocked Thymidylate Synthase

where $l_{DPD} = l_{DPD\_BASE}\left\{1 + \varepsilon \cos\frac{2\pi(t-\varphi_{DPD})}{24}\right\}$ and $S_0 = S_{0\_BASE}\left\{1 + \delta \cos\frac{2\pi(t-\varphi_{TS})}{24}\right\}$

Simulation: 5 courses of 2 week-therapy courses

$i(t)=i_0[1+\sin\{2\pi(t-\varphi_{5FU}+9)/12\}]$ and $j(t)=j_0[1+\sin\{2\pi(t-\varphi_{LV}+9)/12\}$, then zero for 12 hours

4 days of 4FU+LV infusion,12 hours a day, every other week

P = Plasma [5FU]

F = Intracellular [FdUMP]

Q = Plasma [LV]

L = Intracellular [LV]

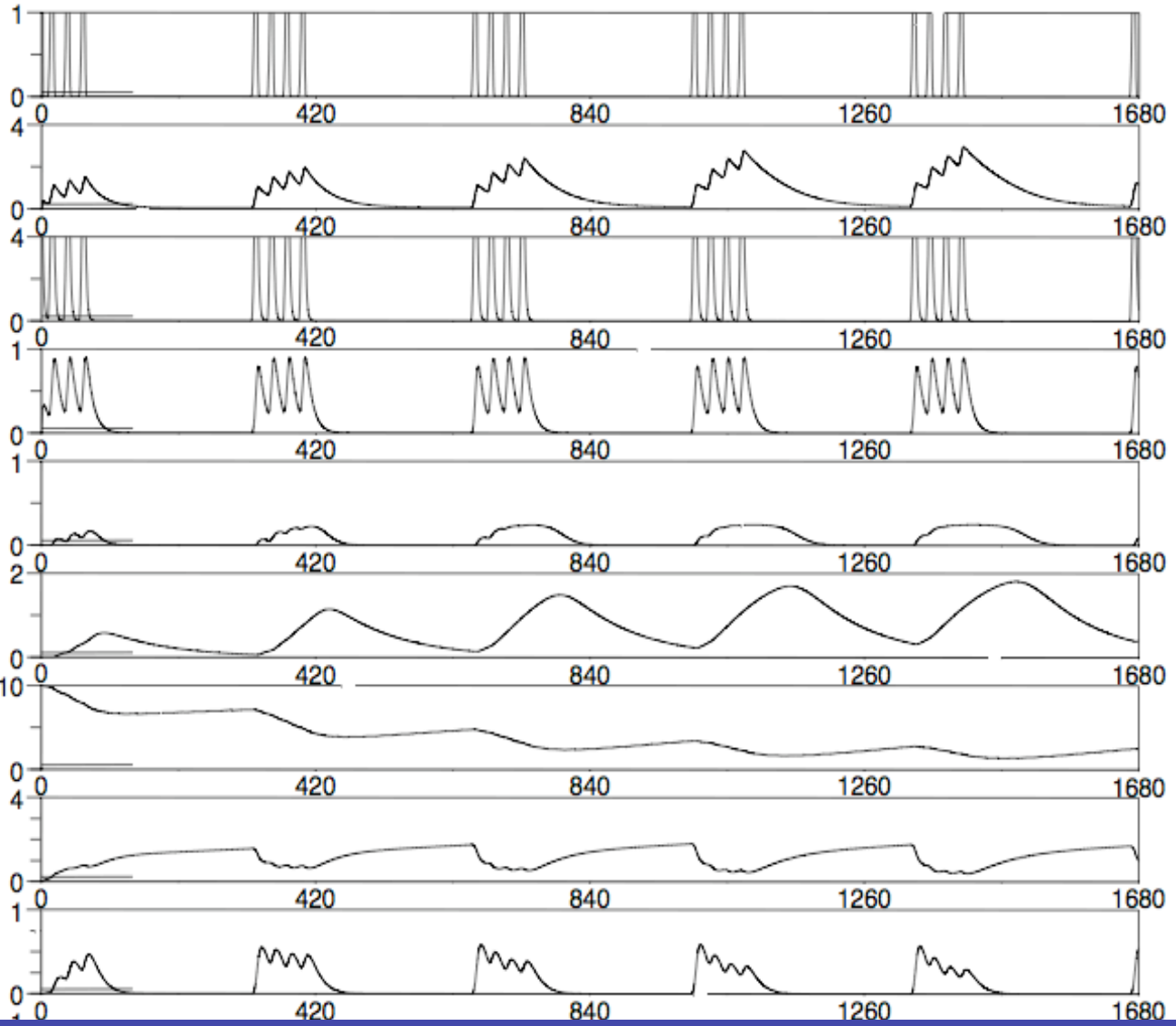
N = [nrf2] 5FU-triggered Nuclear Factor

A = ABC Transporter activity, nrf2-inducted

S = Free [TS] (not FdUMP-bound)

B = [FdUMP-TS] reversible binary complex

T = [FdUMP-TS-LV] stable ternary complex



Induction of ABC Transporter activity by FdUMP-triggered synthesis of nuclear factor *nrf2*

$$\frac{dF}{dt} = \frac{a}{\xi}\frac{P}{b+P} - \frac{AF}{c+F} - k_1 F S + k_{-1} B$$

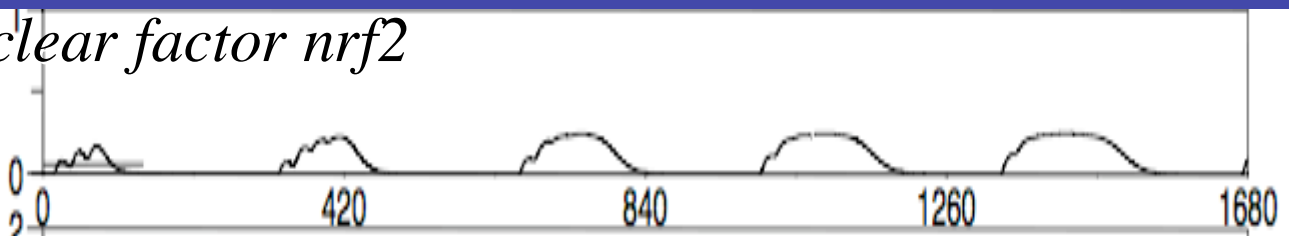
$$\frac{dN}{dt} = \frac{\kappa F^n}{\lambda^n + F^n} - \mu N$$

$$\frac{dA}{dt} = \mu N - \nu A$$

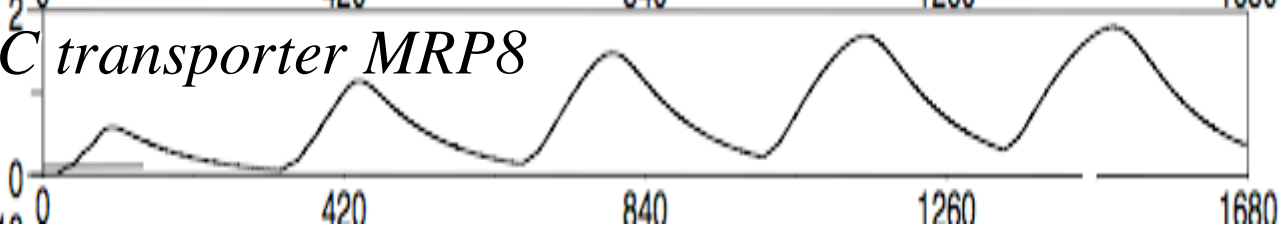
Nuclear factor *(nrf2)*

ABC Transporter (ABCC11=MRP8)

N=nuclear factor nrf2



A=ABC transporter MRP8

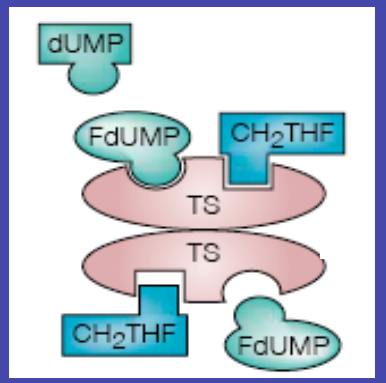


Targeting Thimidylate Synthase (*TS*) by FdUMP: Formation of binary and ternary *TS*-complexes

$$\frac{dS}{dt} = -k_1 FS + k_{-1} B + \theta_{TS}(S_0 - S)$$

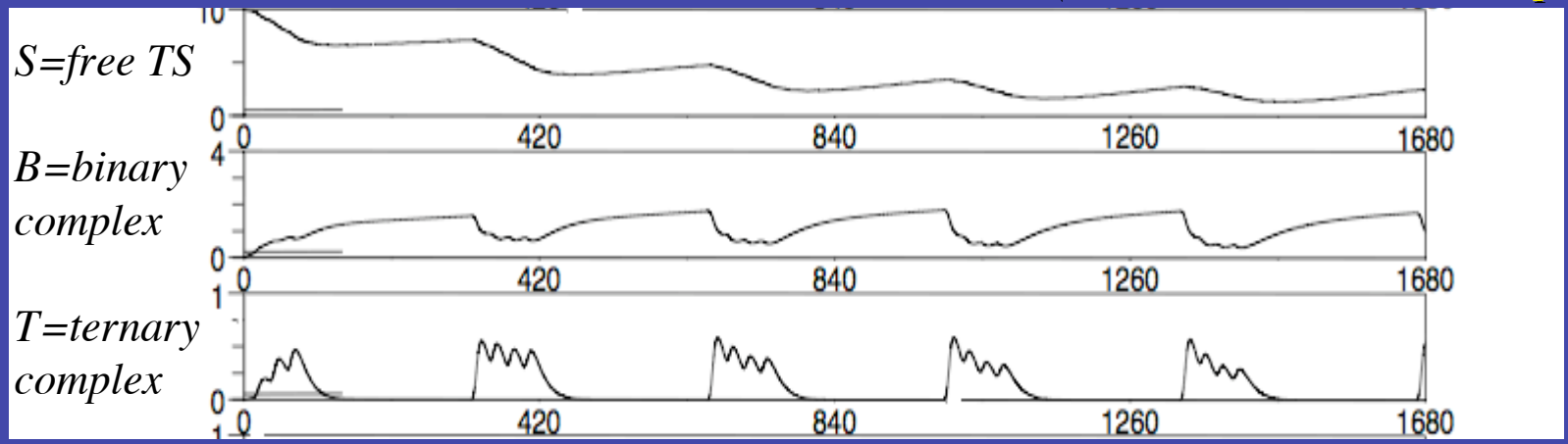
$$\frac{dB}{dt} = k_1 FS - k_{-1} B - k_4 BL$$

$$\frac{dT}{dt} = k_4 BL - v_T T$$



$F + S \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} F\text{-}S = B$ *(FdUMP-TS 2-complex)*

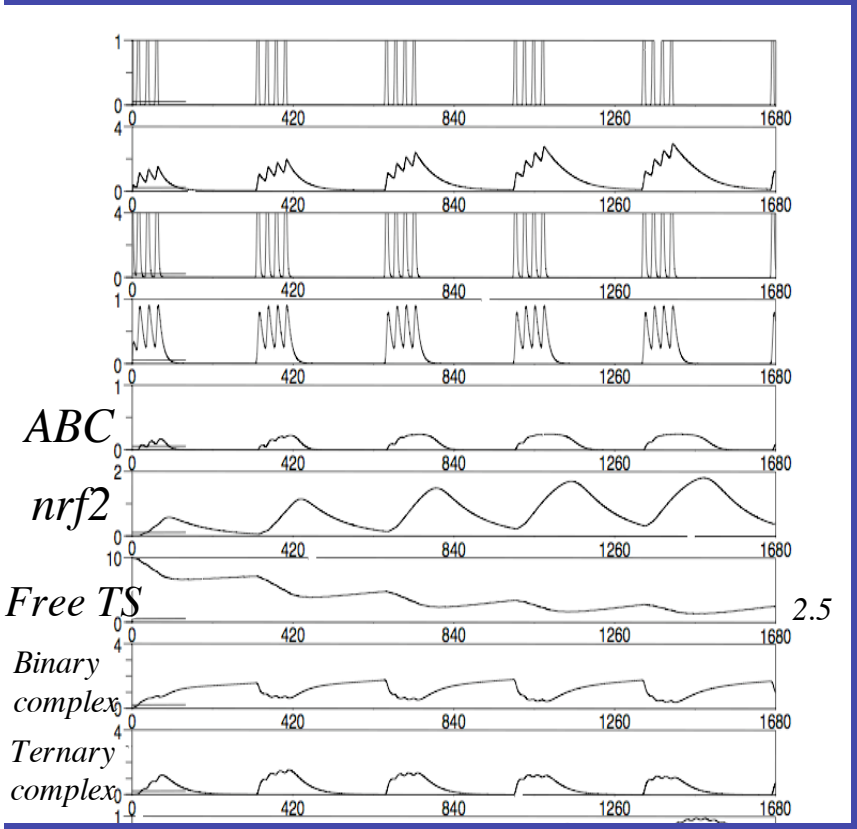
$B + L \xrightarrow{k_4} B\text{-}L = T$ *(FdUMP-TS-LV 3-complex)*



Examples of features of the model:

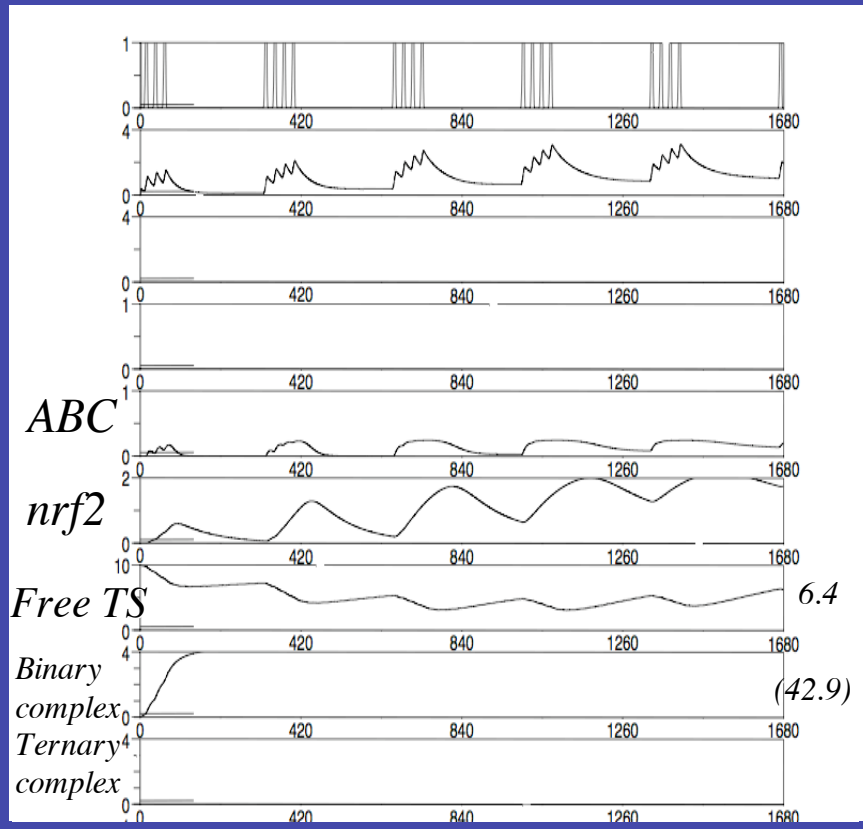
a) 5FU with/without LV in cancer cells (=ABC transporter MRP8+)

With Leucovorin added in treatment



Free TS decays to zero = cancer cells die

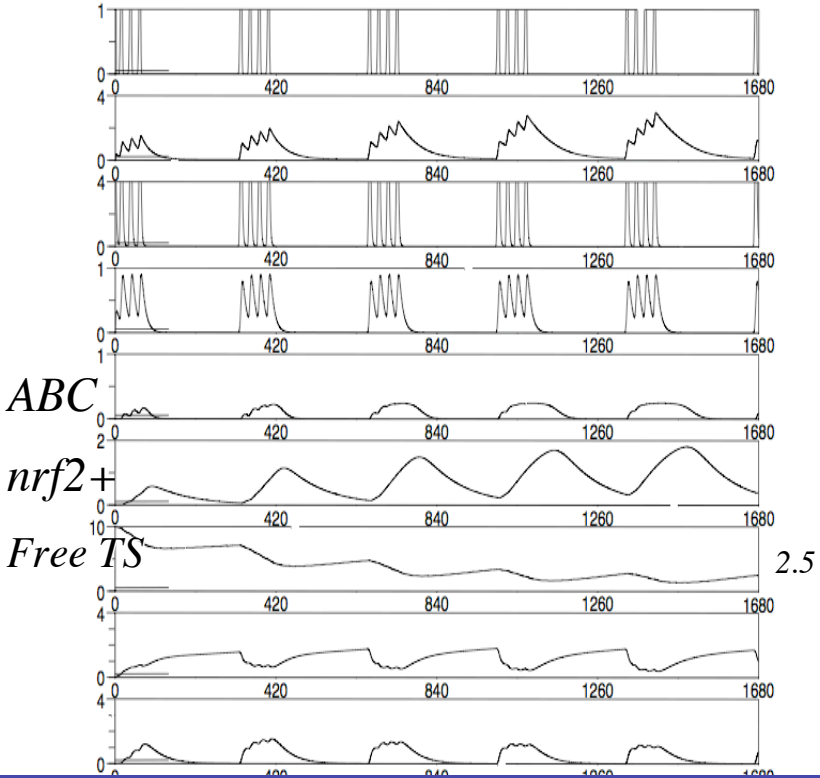
Without Leucovorin added



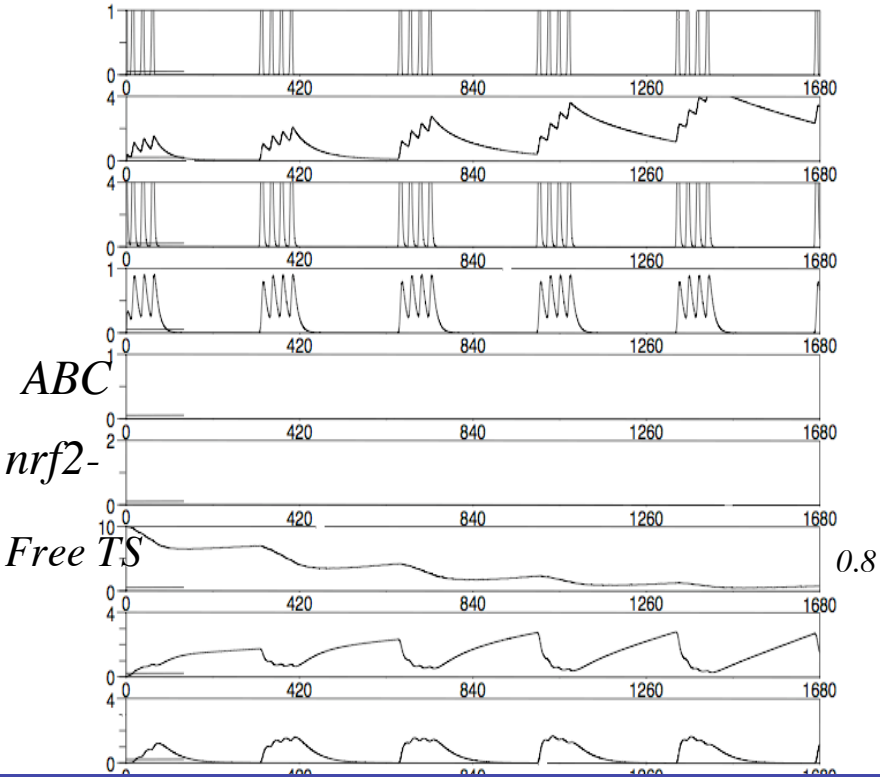
Free TS maintains its level = cancer cells survive

b) 5FU+LV with/without MRP8 (cancer vs. healthy cells)

Cancer cells (ABC+=MRP8+)



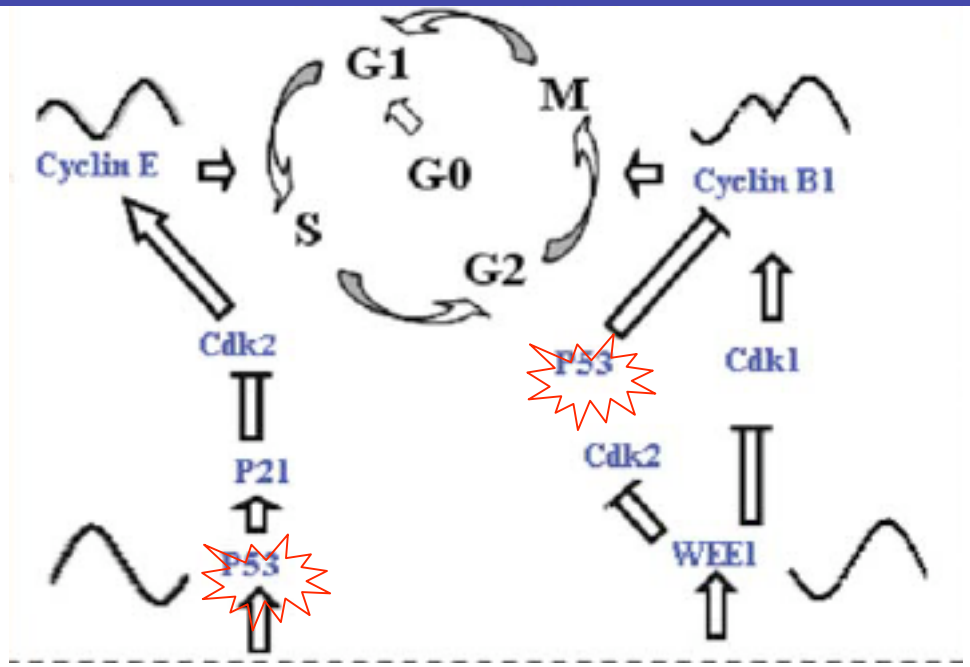
Healthy cells (ABC-=MRP8-)



Cancer cells resist more than healthy cells, due to lesser exposure to FdUMP

(actively effluxed from cells by ABC Transporter MRP8)

Yet to be accurately represented: p53 to connect DNA damage with cell cycle arrest and apoptosis



Needed: a p53-Mdm2 model (existing models by Ciliberto, Chickarmane,…) to connect DNA damage with cell cycle arrest at checkpoints and apoptosis

Future work (or work in progress) for PK-PD models

Such molecular (=physiological) modelling should be included in a multiscale whole body physiologically based ("WBPBPKPD") model:

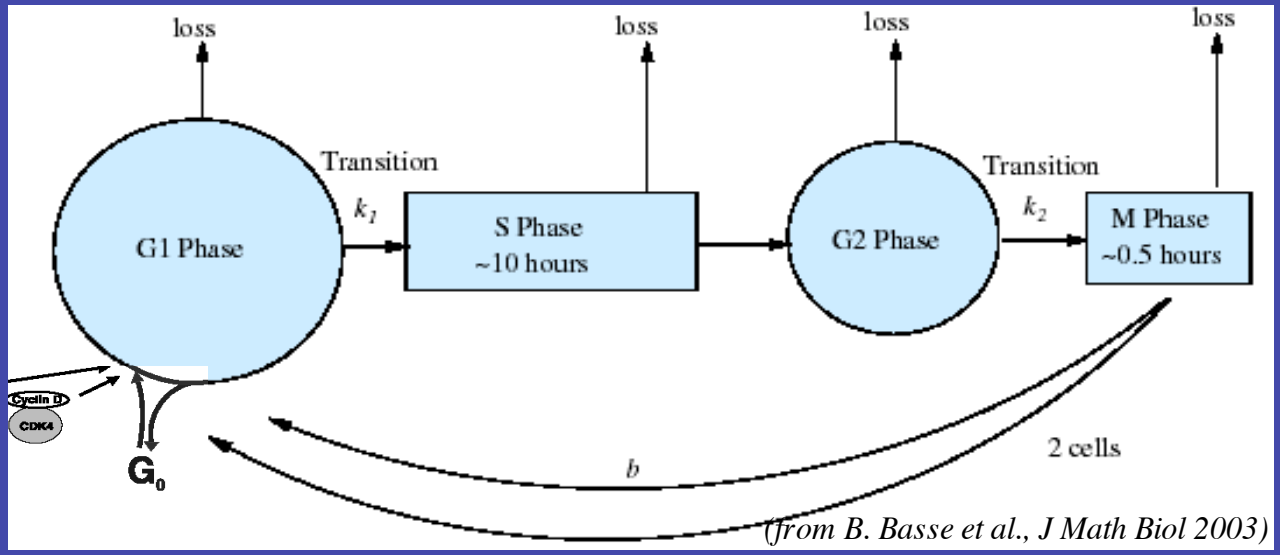
- From the cell to the tissue (cell environment, whole body regulations)
- From the tissue to the whole organism (compartmental modelling)
- From the individual patient to the population (populational PK-PD)

2. Modelling cell proliferation and its control mechanisms in cell populations

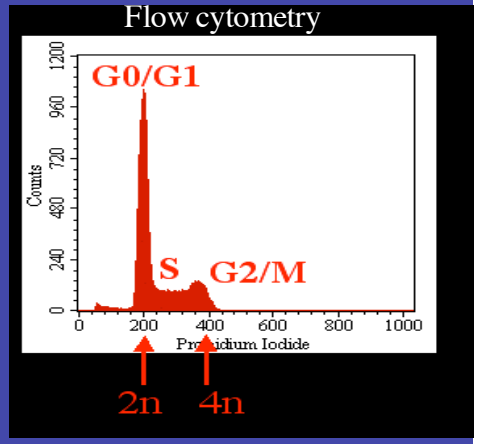
Age-structured partial differential equations (PDEs)

Modelling the cell division cycle in cell populations

Age-structured PDE models



(from B. Basse et al., J Math Biol 2003)



In each phase i, a Von Foerster-McKendrick-like equation:

$$\frac{\partial}{\partial t} n_i(t,a) + \frac{\partial}{\partial a}[v_i(a) n_i(t,a)] + d_i(t,a) n_i(t,a) + K_{i\to i+1}(t,a) n_i(t,a) = 0$$

$$v_i(0) n_i(t, a=0) = \int_{\alpha \geq 0} K_{i-1\to i}(t,\alpha)\, n_{i-1}(t,\alpha)\, d\alpha$$

$$K_{i\to i+1}(t,a) = \psi(t) \mathbf{1}_{a \geq a_i}(a)$$

n_i:=cell population density in phase i;
v_i:=progression speed;
d_i:=death rate;
$K_{i-1->i}$:=transition rate (with a factor 2for $i=1$)
d_i, $K_{i->i+1}$ constant or periodic w. r. to time t ($1 \leq i \leq I$, $I+1=1$)

Death rates d_i: ("loss"), "speeds" v_i and phase transitions $K_{i->i+1}$ are model targets for physiological (e.g. circadian) and therapeutic (drugs) control $\psi(t)$
[$\psi(t)$: e.g., clock-controlled CDK1 or intracellular output of drug infusion flow]

(Firstly presented in: JC, B. Laroche, S. Mischler, B. Perthame, RR INRIA #4892, 2003)

The simplest case: 1-phase model with division

$$\frac{\partial}{\partial t} n(t,a) + \frac{\partial}{\partial a}[n(t,a)] + [d(t) + K(t,a)]\, n(t,a) = 0$$

$$n(t, a=0) = 2 \int_{\alpha \geq 0} K(t,\alpha)\, n(t,\alpha)\, d\alpha$$

$$\text{where } K(t,a) = K_0 \psi(t) \mathbb{1}_{[a^*, +\infty[}(a)$$

$$\text{and } \psi(t) = \mathbb{1}_{[0,\tau[}(t), \text{1-periodic}$$

(Here, $v(a)=1$, a^* is the cell cycle duration, and $\tau < 1$ is the time during which the *periodic control* ψ is actually exerted on cell division)

Then it can be shown that the eigenvalue problem: $n(t,a) = e^{\lambda t} N(t,a)$

$$\frac{\partial}{\partial t} N(t,a) + \frac{\partial}{\partial a}[N(t,a)] + [\lambda + d(t) + K(t,a)]\, N(t,a) = 0$$

$$N(t, a=0) = 2 \int_{\alpha \geq 0} K(t,\alpha)\, N(t,\alpha)\, d\alpha$$

admits a unique *positive* 1-*periodic* eigenvector N, with a *positive* eigenvalue λ, a so-called Malthus exponent, or exponential growth rate for the cell population

General case (I phases): Existence of a nonnegative first eigenvalue λ and, if $\widetilde{N}_i(t,a) = e^{-\lambda t} n_i(t,a)$, of eigenvectors N_i, bounded solutions to the problem :

$$\begin{cases} \frac{\partial}{\partial t} N_i(t,a) + \frac{\partial}{\partial a} N_i(t,a) + [d_i(t,a) + \lambda + K_{i\to i+1}(t,a)] N_i(t,a) = 0, \\ N_i(t,a=0) = \int_{\alpha \geq 0} K_{i-1\to i}(t,\alpha)\, N_{i-1}(t,\alpha)\, d\alpha, \quad 2 \leq i \leq I \\ N_1(t,a=0) = 2 \int_{\alpha \geq 0} K_{I\to 1}(t,\alpha)\, N_I(t,\alpha)\, d\alpha, \quad \text{with} \sum_{i=1}^{I} \int_{a\geq 0} N_i(t,a) da = 1 \end{cases}$$

with a function $\rho\,(a)$ such that the asymptotics of $\widetilde{N}_i(t,a) = e^{-\lambda t} n_i(t,a)$ follow:

$$\int_{\alpha \geq 0} \left| \widetilde{N}_i(t,\alpha) - \rho\,(\alpha)\, N_i(t,\alpha) \right| \varphi_i(t,\alpha) d\alpha \to 0 \qquad \text{as } t \to \infty$$

the φ_i being solutions to the dual problem; this can be proved by using an entropy principle (GRE). Moreover, if the control (d_i or $K_{i->i+1}$) is constant, or if it is periodic, so are the N_i, with the same period in the periodic case

[Term $d_i + \lambda + K_{i->i+1}$ for the same N_i to be solutions: the higher the d_i, the lower the λ]α

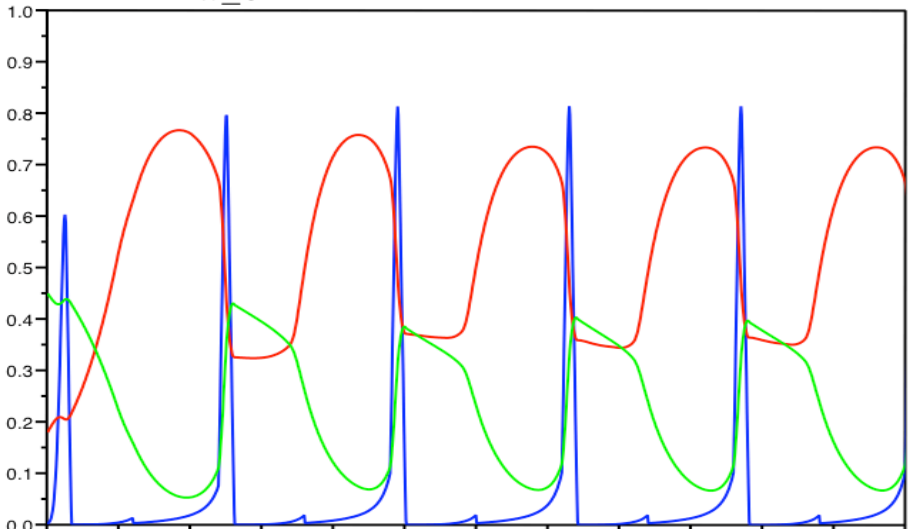
Michel, Mischler, Perthame, C. R. Acad. Sci. Paris Ser. I (Math.) 2004; J Math Pures Appl 2005
JC, Michel, Perthame, C. R. Acad. Sci. Paris Ser. I (Math.) 2006; Proc. ECMTB Dresden 2005, Birkhäuser 2007
JC, Gaubert, Perthame, C. R. Acad. Sci. Paris Ser. I (Math.) 2007; JC, Gaubert, Lepoutre, Submitted

To sum up: a growth exponent for the cell population

Proof of the existence of a unique growth exponent λ, the same for all phases *i*, such that the $\widetilde{N}_i(t,a) = e^{-\lambda t} n_i(t,a)$ are asymptotically (i.e., for large times) bounded, and asymptotically periodic if the control is periodic

Surfing on the exponential growth curve, example (periodic control case): 2 phases, control on G_2/M transition by 24-h-periodic CDK1-Cyclin B (A. Goldbeter's model)

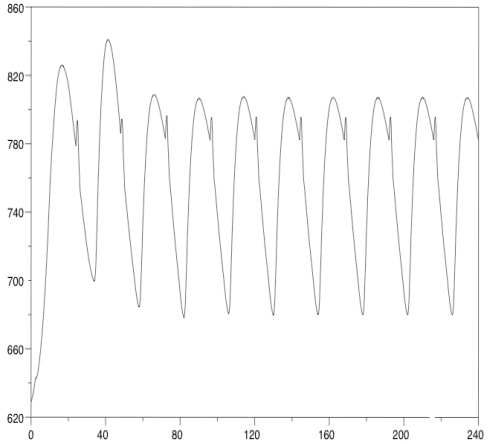
$$\mathcal{N}_i^{tot}(t) = e^{-\lambda t} \int_{\alpha \geq 0} n_i(t,\alpha) d\alpha, \quad i = 1,2$$



ψ=CDK1 **All cells in G1-S-G2 (phase *i*=1)** **All cells in M (phase *i*=2)**

Entrainment of the cell division cycle by CDK1 at the circadian period

All cells, surfing on the exponential growth curve

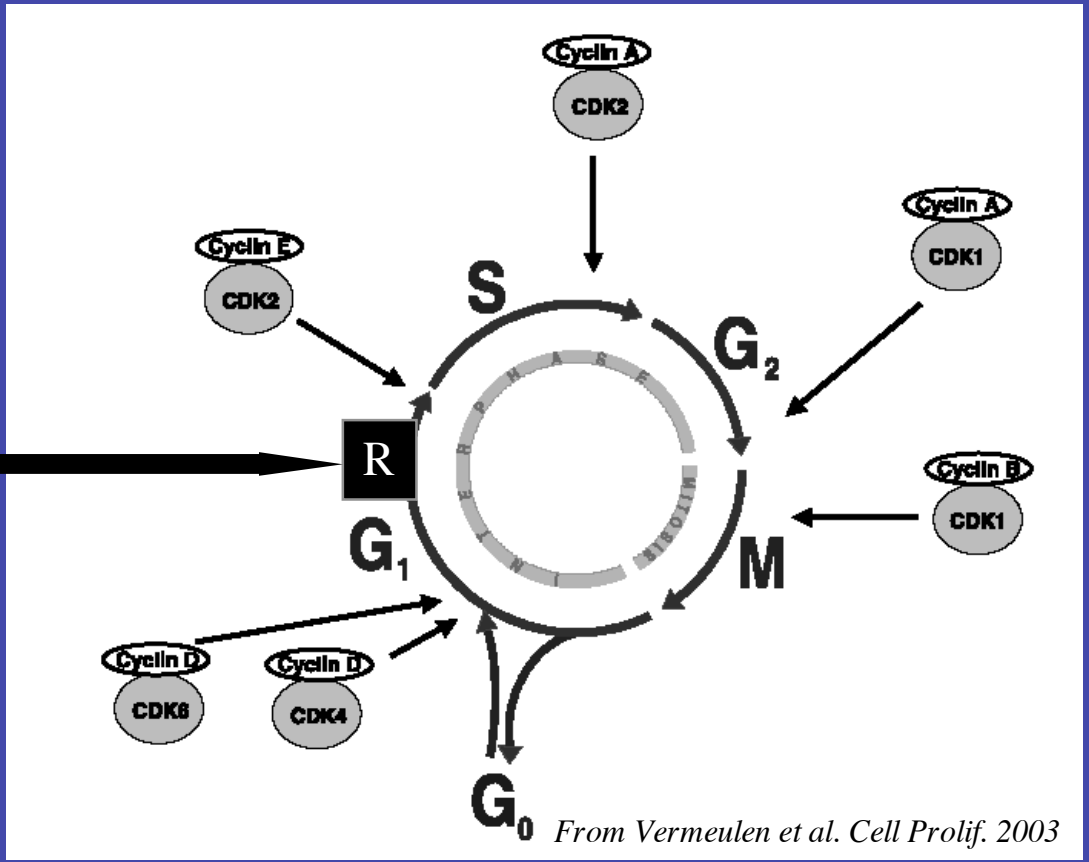


Complementary modelling: exchanges between G_1 and G_0 phases (proliferating and quiescent cell populations)

after R:
mitogen-independent
progression through G1 to S
(no way back to G_0)

Restriction point
(late G_1 phase)

before R:
mitogen-dependent
progression through G1
(possible regression to G_0)



From Vermeulen et al. Cell Prolif. 2003

Most cells do not proliferate physiologically, even in fast renewing tissues (e.g. gut)

Exchanges between proliferative (G_1SG_2M) and quiescent (G_0) cell compartments are controlled by mitogens and antimitogenic factors in G_1 phase

Exchanges between proliferative (p) and quiescent (q) phases: healthy and tumour tissue cases: G_0 to G_1 recruitment differs

$$\begin{cases} \dfrac{\partial}{\partial t}p(t,a,x) + \dfrac{\partial}{\partial a}\left(\Gamma_0 p(t,a,x)\right) + \dfrac{\partial}{\partial x}\left(\Gamma_1(a,x)\,p(t,a,x)\right) = \\ -\left(L(a,x) + F(a,x) + d_1\right)p(t,a,x) + G(N(t))\,q(t,a,x), \\ \dfrac{\partial}{\partial t}q(t,a,x) = L(a,x)\,p(t,a,x) - \left(G(N(t)) + d_2\right)q(t,a,x). \end{cases}$$

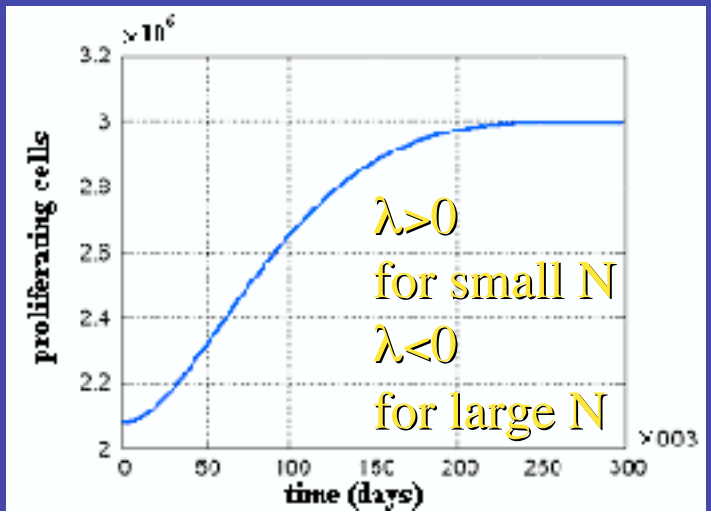
$N=\Sigma p+q$ (total number of cells at time t)

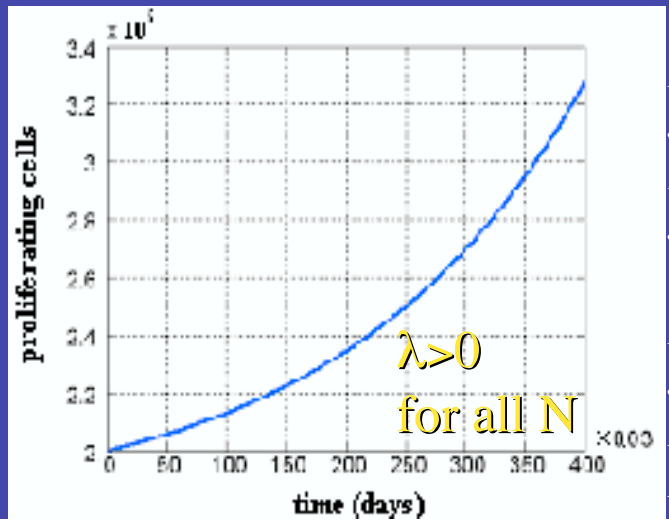
$$G(N) = \frac{\alpha_1\theta^n}{\theta^n + N^n}$$

Healthy cells: tissue homeostasis

$$G(N) = \frac{\alpha_1\theta^n + \alpha_2 N^n}{\theta^n + N^n}$$

Tumour cells: exponential growth





Bekkal Brikci, JC, Ribba, Perthame J Math Biol 2008

Bekkal Brikci, JC, Perthame Math Computer Modelling 2008

Work in progress and future work on cell populations

1) Merging the linear (Von Foerster-McKendrick) and non linear (Gyllenberg-Webb-like) models, together with cell population synchronisation control on transitions $K_{i->i+1}$ [and on velocities $v_i(a)$]

2) Representing the action of different drugs (cytotoxics, EGFR antagonists,…) on molecular-based targets in *various tissue environments*: with mitogens and antimitogens, genomic [in]stability, cell population [de]synchronisation (by *adaptive dynamics equations*?)

3. The circadian system and its disruptions

('Circa diem'=approximately one day)

Representing physiological and disrupted control functions

ψ_i on cell cycle phase transitions (G_1/S, G_2/M)

The circadian system

(SCN = Suprachiasmatic Nuclei, located in the hypothalamus)

Central coordination

CNS, hormones, peptides, mediators

Pineal

NPY **NPV**

Melatonin

Entrainment by light

RHT

Glutamate

SCN

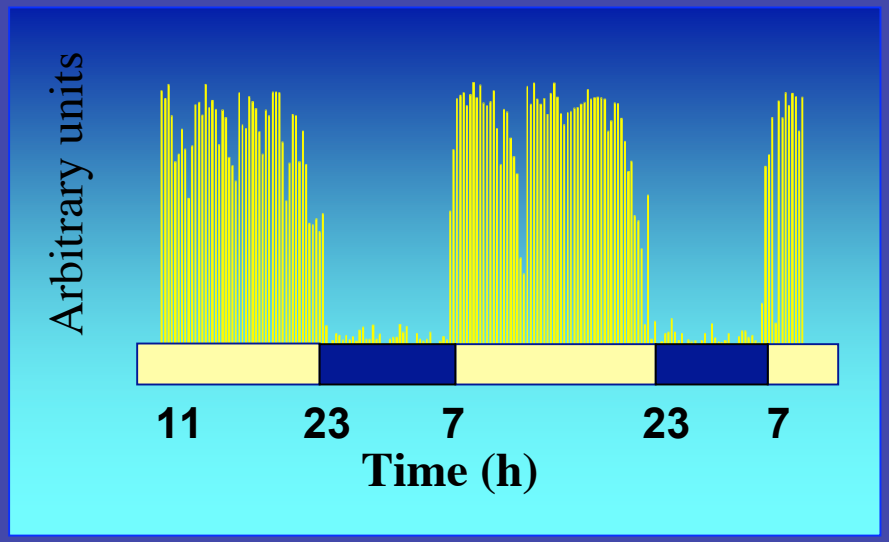
TGF α, EGF

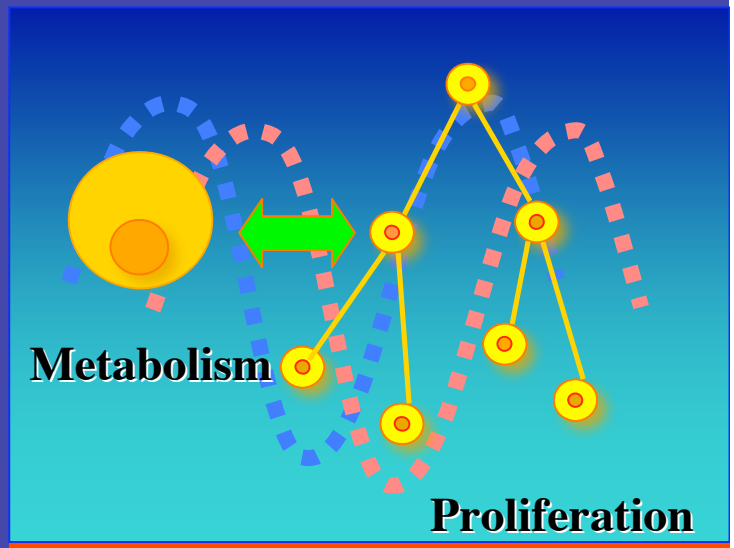
Prokineticin

Glucocorticoids

Food intake rhythm

Autonomic nervous system





Rest-activity cycle: open window on SCN central clock

Peripheral oscillators

Lévi & Schibler, Ann Rev Pharmacol Toxicol 2007

(from Francis Lévi, INSERM U 776 Rythmes Biologiques et Cancers)

The SCN pacemaker as a network of coupled oscillators:

Leloup-Goldbeter simplest circadian clock with diffusive coupling between neurons

$$\frac{dmRNA(i)}{dt} = V_s \frac{K^n}{K^n + Z(i)^n} - V_m(i) \frac{mRNA(i)}{K_m + mRNA(i)}$$

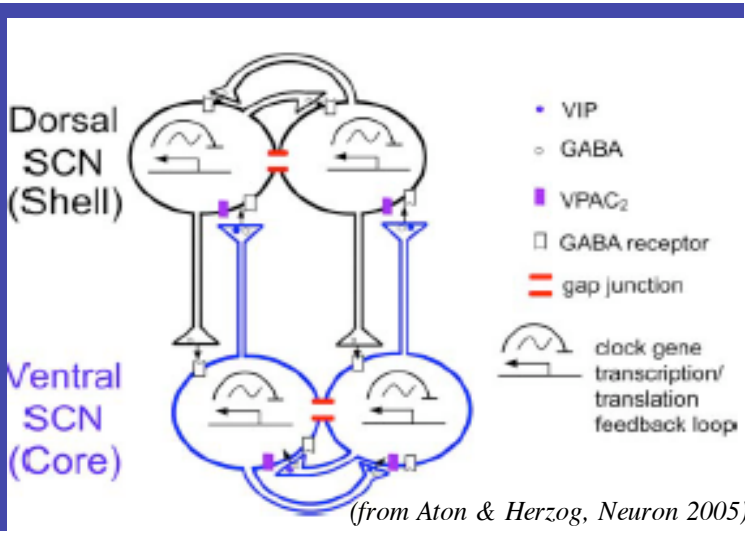
$$\frac{dPER(i)}{dt} = k_s mRNA(i) - V_d \frac{PER(i)}{K_d + PER(i)} - k_1 PER(i) + k_2 Z(i) + K_e \sum_{j \neq i} [PER(j) - PER(i)]$$

$$\frac{dZ(i)}{dt} = k_1 PER(i) - k_2 Z(i)$$

(after Leloup, Gonze, Goldbeter, J Biol Rhythms 1999)

V_s : $V_s = 1.6\,(1 + L\cos(2\pi t/24))$ target of entrainment by light L; K: target of transcriptional inhibition (e.g. by cytokines); $V_m(i)$: the carrier of variabilility of the oscillatory period in this model

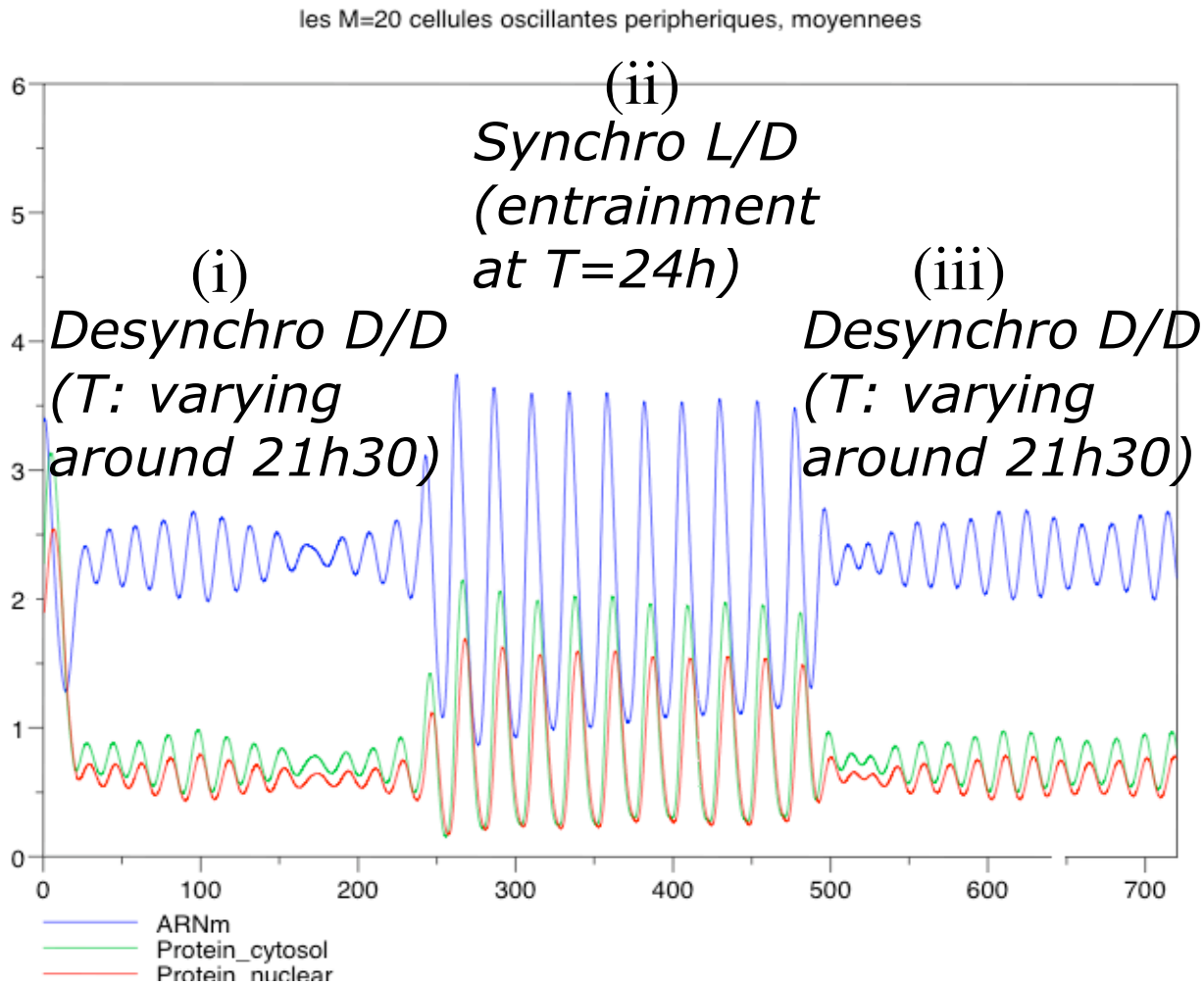
3 variables for the i^{th} neuron that communicates with all other ($j \neq i$) neurons of the SCN through cytosolic PER protein, with coupling constant K_e: electric? gap junctions? VIP / $VPAC_2$ signalling?



(from Aton & Herzog, Neuron 2005)

Result: example of disrupted clock: averaged *peripheral* oscillator

(i) without *central pacemaker* entrainment by light; (ii) with it; (iii) without it again



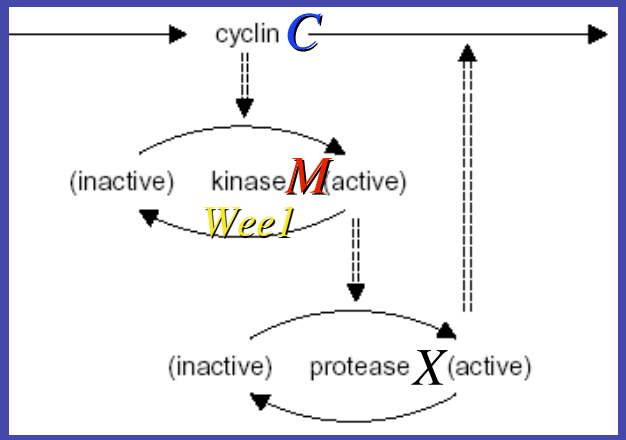
Resulting Per to control Wee1, that inhibits CDK1= ψ, in proliferating cells

JC, Proc. IEEE-EMBC 2006, IEEE-EMB Mag 2008

Relating circadian clocks to the cell division cycle

ODEs to describe progression in the cell cycle at the single-cell level

A. Golbeter's minimal model for the G_2/M transition (the « mitotic oscillator »)



Wee1

$$\frac{dC}{dt} = v_i - k_d C - v_d X \frac{C}{K_d + C}$$

$$\frac{dM}{dt} = v_1 \frac{C}{K_c + C} \frac{(1 - M)}{K_1 + (1 - M)} - V_2 \frac{M}{K_2 + M},$$

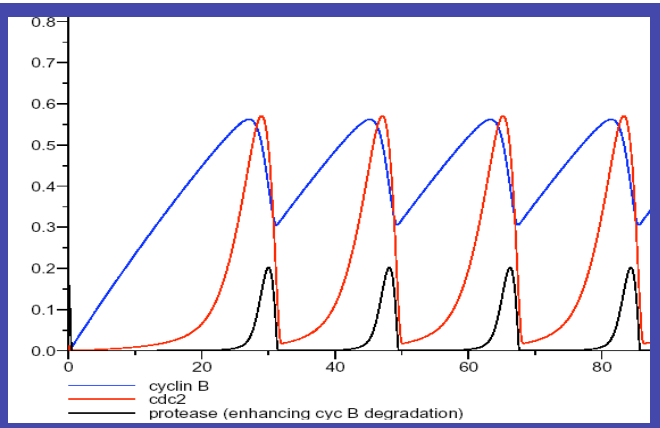
$$\frac{dX}{dt} = v_3 M \frac{(1 - X)}{K_3 + (1 - X)} - V_4 \frac{X}{K_4 + X}.$$

C = cyclin B, M = cyclin dependent kinase cdk1, X = degrading protease

Input: *Per=Wee1*; output: M=Cdk1=ψ

Switch-like dynamics of dimer Cyclin B-cdk1

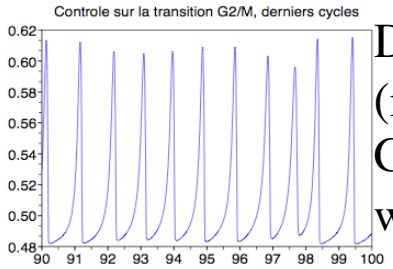
Adapted to describe G_2/M phase transition



(A. Goldebeter Biochemical oscillations and cellular rhythms, CUP 1996)

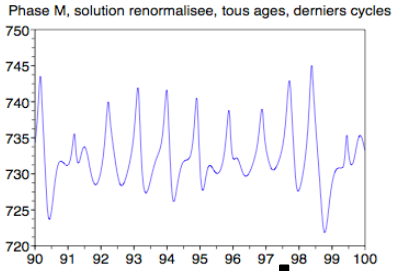
Clock perturbations and cell population proliferation

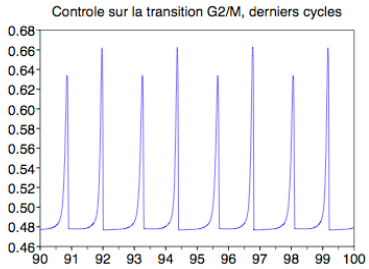
(Wee1 here identified as averaged Per in the circadian clock model)



Desynchronised Wee1 (no entrainment by light): Control cdk1= ψ with perturbed clock

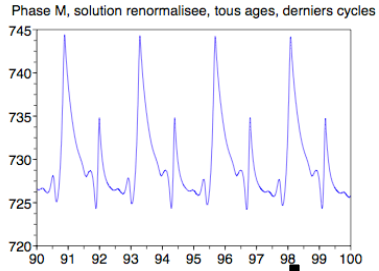
Resulting irregular cell population dynamics in M phase





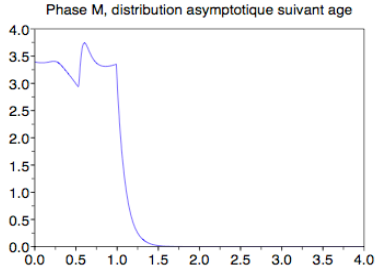
Synchronised Wee1 (entrainment by light): Control cdk1= ψ with unperturbed clock

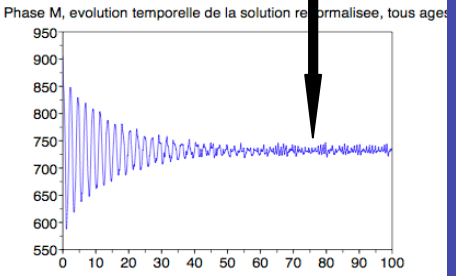
Resulting regular cell population dynamics in M phase



Wee1=Per is desynchronised at the central (NSC) level

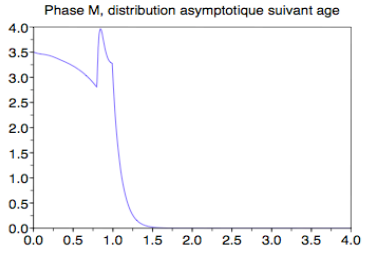
Resulting λ=0.0466

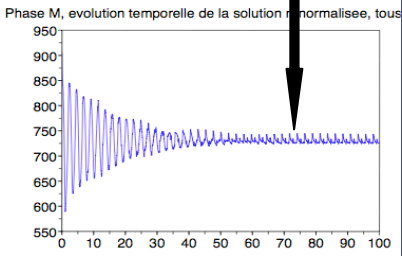




Wee1=Per is synchronised at the central (NSC) level

Resulting λ=0.0452





4. Optimisation of anticancer pharmacotherapy

1) *Objective function* to be minimised: cell population growth rate or cell population density in tumour tissues

2) *Control function*: instantaneous [dynamic] intravenous infusion = [multi-]drug delivery flow via external programmable pumps

3) *Constraints* to be satisfied:
   - maintaining healthy cell population over a tolerability threshold
   - taking into account circadian phases of drug processing systems (model prerequisite)
   - *maintaining normal tissue synchronisation control by circadian clocks*
   - limiting resistances in tumour cells (*e.g. controlling induction of nrf2)*
   - others: maximal daily dose, maximal delivery flow,…

4) *With adaptation* of drug delivery flow to *patient-specific parameters* (clock phases, enzyme genetic polymorphism, target protein levels,…)

Drug resistance: a constraint in optimising strategies? Different mechanisms of resistance in tumour cells

1. Efflux pumps (ABC transporters): Innate? Acquired: how should they be induced by drugs? (*activation of dormant transporters? Proteic synthesis?*)

2. Mutations of the target (*e.g. of BCR-Abl protein for Imatinib resistance*): representation within the frame of PDEs structured according to a genetic trait?

3. Overexpression of drug processing enzymes or other detoxicating molecules

4. Overexpression of DNA mismatch repair enzymes (*resistance to radiotherapy*)

5. Environmental factors (*micro-, e.g. hospicells; or macro-, e.g., insensitivity to the immune system*)

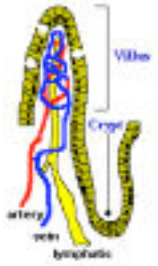
Example of chronotherapy optimisation with respect to treatment tolerability in a single-drug case

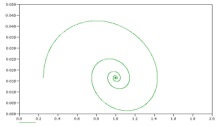
Oxaliplatin to treat Glasgow osteosarcoma in mice, with a jejunal toxicity limit

PK-PD simplified model for cancer chronotherapy

Healthy cells (jejunal mucosa)

$$\begin{aligned}
\frac{dP}{dt} &= -\lambda P + \frac{i(t)}{V}\Phi(t) \\
\frac{dC}{dt} &= -\mu C + P \\
\frac{dZ}{dt} &= -\{\alpha + f(C,t)\}Z - \beta A + \gamma \\
\frac{dA}{dt} &= Z - Z_{eq}
\end{aligned}$$

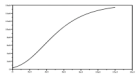




(PK)

Tumour cells

$$\begin{aligned}
\frac{dP}{dt} &= -\lambda P + \frac{i(t)}{V}\Phi(t) \\
\frac{dD}{dt} &= -\nu D + \xi_D P \\
\frac{dB}{dt} &= \left[a \ln \frac{B_{max}}{B} - g(D,t)\right] B
\end{aligned}$$



(homeostasis=damped harmonic oscillator)

(tumour growth=Gompertz model)

(« chrono-PD »)

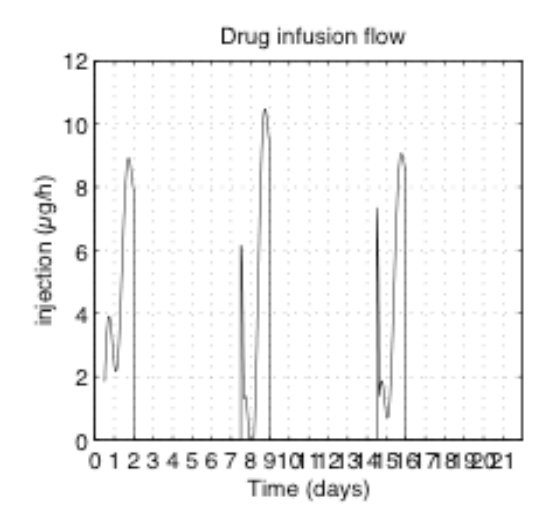
$f(C,t)=F.C^{\gamma}/(C_{50}{}^{\gamma}+C^{\gamma}).\{1+\cos 2\pi(t-\varphi_S)/T\}$

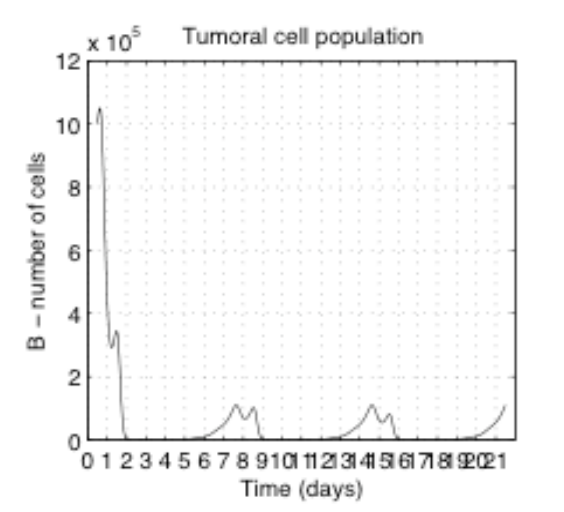
$g(D,t)=H.D^{\gamma}/(D_{50}{}^{\gamma}+D^{\gamma}).\{1+\cos 2\pi(t-\varphi_T)/T\}$

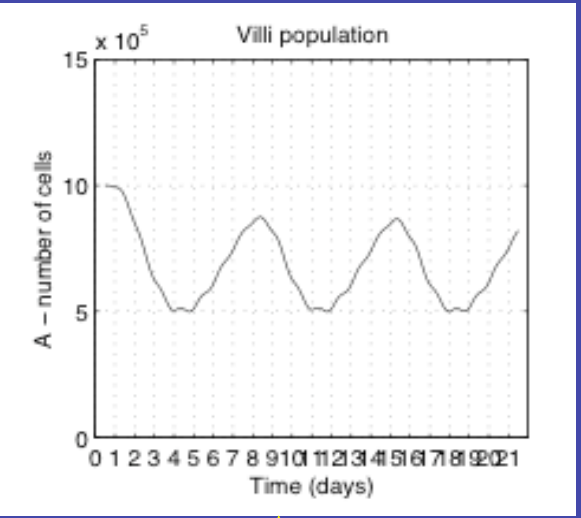
Aim: balancing IV delivered drug anti-tumour efficacy by healthy tissue toxicity

JC Adv Drug Deliv Rev 2007

Optimal control: results of a tumour stabilisation strategy using this simple PK-PD model







Objective: *minimising the maximum of the tumour cell population*

Constraint : *preserving the jejunal mucosa according to the patient's state of health*

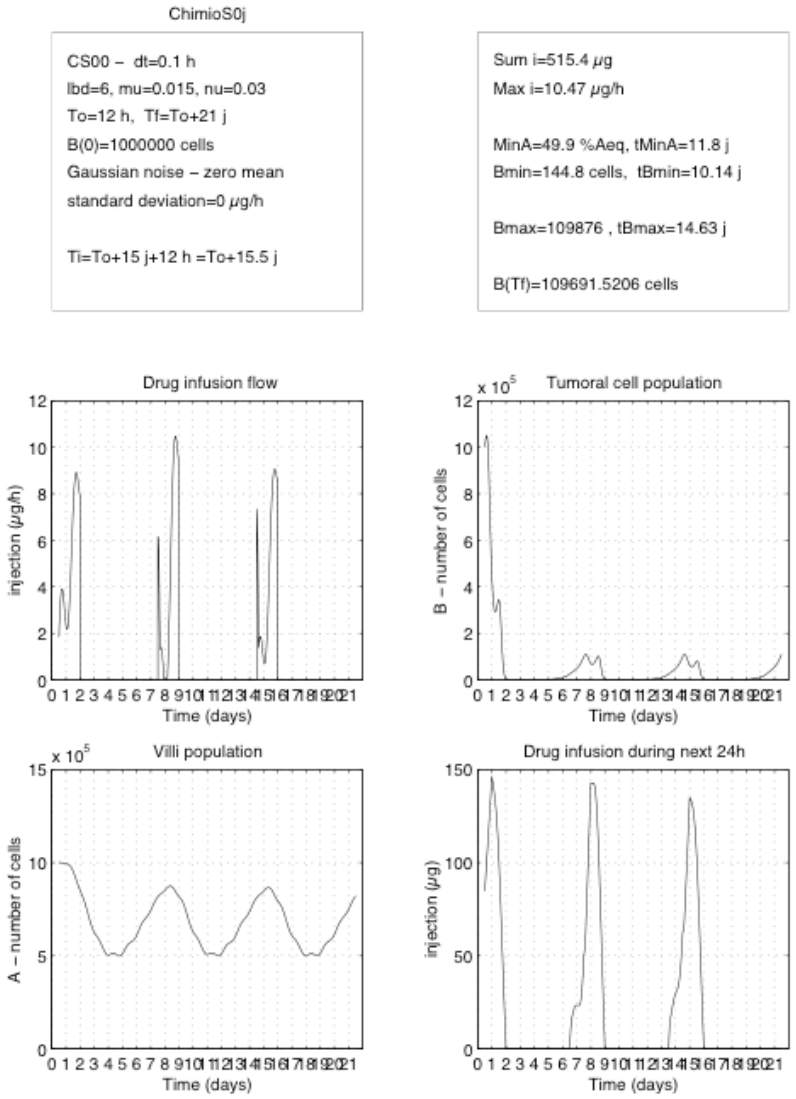
Result : optimal infusion flow adaptable to the patient's state of health

(according to a parameter τ_A: here preserving at least τ_A=50% of enterocytes*)*

(Basdevant, JC, Lévi, M2AN 2005)

Example of treatment adaptation to the patient

For τ_A = 50 %:



Varying τ_A:

Numerical results for 1.5 days of infusion + 5.5 days of recovery:

τ_A	max B(t)	min B(t)
40 %	28 000	6
50 %	102 000	147
60 %	305 000	2700

τ_A= 1- tolerability
(the more fragile
the patient, the higher is τ_A)

5. Individualising treatments: (circadian profiling, genotyping)

Identifying patient-specific parameters

a) Circadian clock timing: phases of rhythmic cell cycle determinants (*cyclins, p53*) and drug metabolism enzymes (*e.g., DPD*) and molecules (*e.g. red. Glutathione*)

b) Expression and activity levels of drug processing enzymes (uptake, degradation, efflux, e.g. *DPD, UGT1A1, P-gp*) and cellular targets (*e.g. TS, Topoisomerase I*)

c) DNA mismatch repair enzyme gene expression (*e.g. ERCC1, ERCC2*)

Aim: patient genotyping to individualise optimised drug delivery schedules

… But accurate biomarkers for treatment adaptation are still required!

Conclusion: ongoing modelling work in 3 directions

- *ODEs* for (as much as possible WBPB) PK-PD modelling
  to represent the dynamics of drug concentrations in cells and tissues

- Physiologically structured (possibly including space) *PDEs* with prescribed targets to represent cell and tissue proliferation dynamics

- *Optimisation methods* to propose optimal control of [multi-] drug infusion flows, considering possible drug synergies and [innate or acquired] drug resistances

- *…Account being taken in the models of whole body (e.g., circadian) known physiological controls on PK and PD determinants, and individual (patient-specific) parameters to be identified (what biomarkers?) for personalised medicine*