MATHEMATICAL MODELS OF TUMOR GROWTH INHIBITION IN XENOGRAFT MICE AFTER ADMINISTRATION OF ANTICANCER AGENTS GIVEN IN COMBINATION

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Accelera
Experience results.

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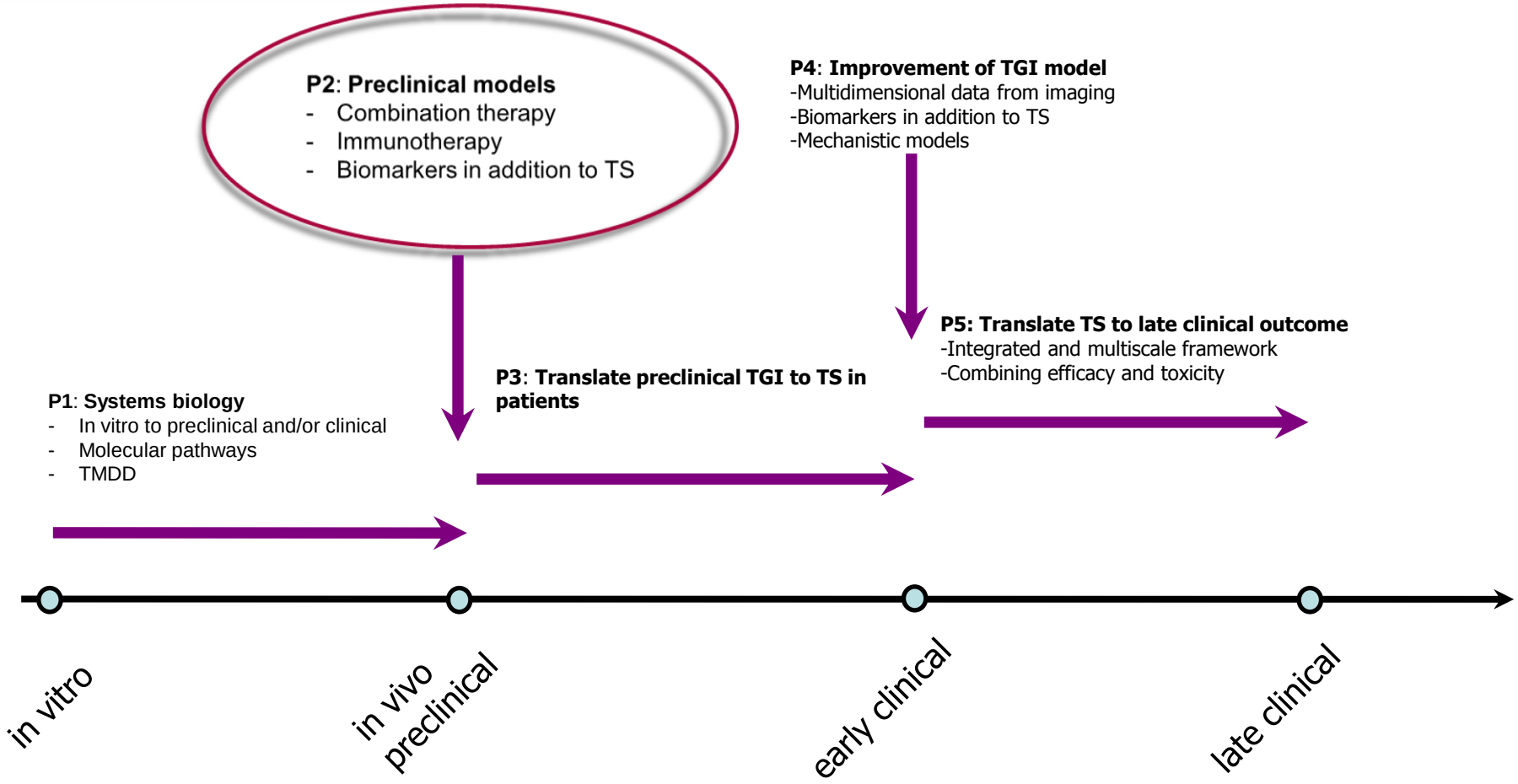
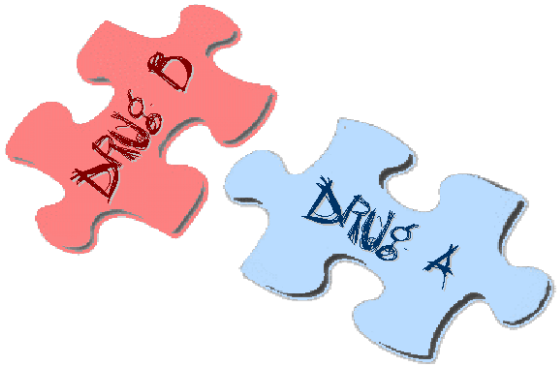
New model development: course of action in the oncology area



Combination therapies

- The pharmacological therapy in oncology patients is always polychemiotherapy.
- The evaluation of the most promising combinations is a fundamental step in early drug development.
  - ad-hoc *in vitro* and *in vivo* experiments routinely performed;
  - identification of drug combinations that yield an enhanced pharmacological effect;
  - prioritization of combinations taking into account the interaction intensity.

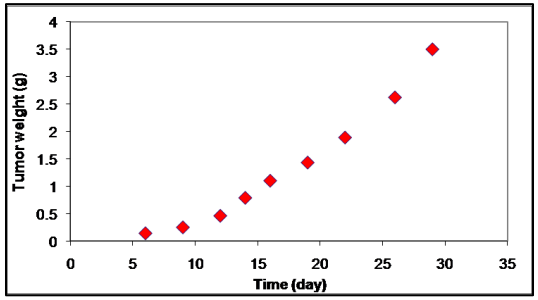


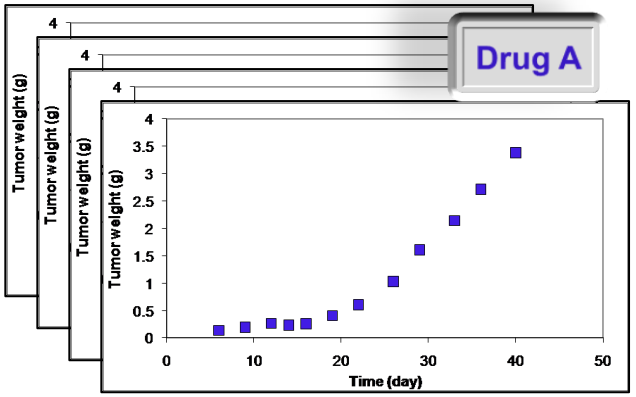
Typical experiments in xenograft mice

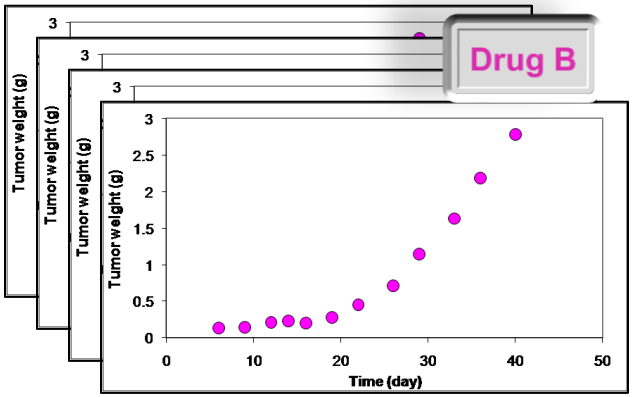
- **xenograft models:** after the inoculation of human tumor fragments, mice bearing a palpable tumor are randomized into control groups and groups treated with an anticancer drug given as a single agent or in combination with another drug. At different time points the tumor weight of each animal is recorded.

SINGLE AGENT ARMS

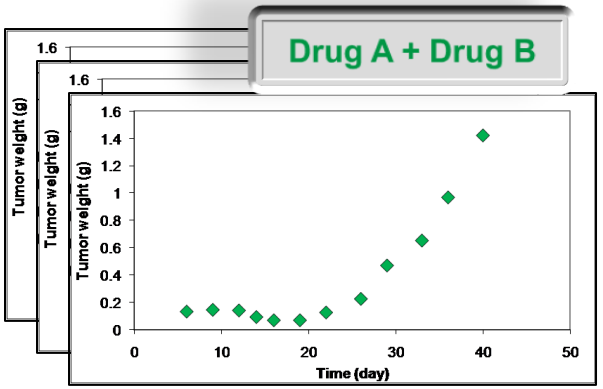
CONTROL ARM





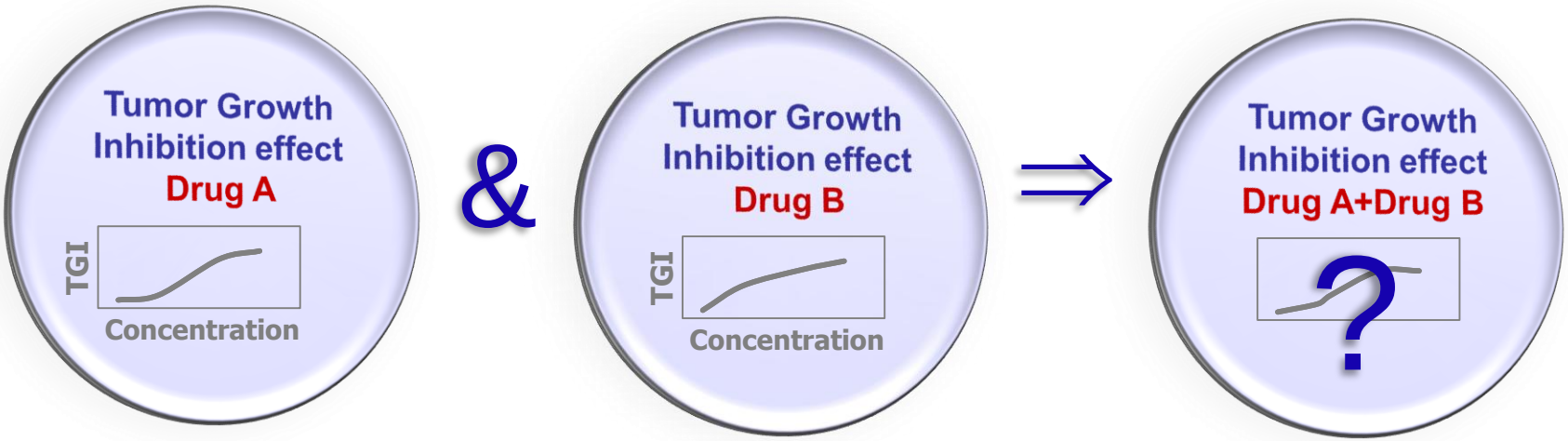


COMBINATION ARMS



Assessment of combination therapies in xenograft mice

- The assessment of the combination of two or more compounds with respect to single compound regimens is an open question, regarding the quantitative interpretation of interaction in terms of:
  - nature (additive, synergistic or antagonist effects);
  - intensity.



Minimal model of TGI under the hypothesis of no interaction

TGI model to assess the drug effect interaction

Definition of no interaction between drugs

Theoretical definition of the concepts of interaction/no interaction by formulating a mathematical model in a probabilistic framework.

- Starting from cellular level assumptions, a *minimal model* able to define and simulate the **no interacting (or zero-interacting or "additive") behavior of an arbitrary number of antitumor drugs** used in combination regimens has been obtained.
  - Stochastic model based on a minimal set of probabilistic assumptions on tumor cells (single cell model, cell population model, Poisson events).

Minimal model of TGI under the hypothesis of no interaction

- Drugs damage cells that after an irreversible process die (e.g. cytotoxic or target oriented agents).
- Tumor cells are divided in two groups: proliferating and non-proliferating.
- The process is governed by a minimal set of probabilistic assumptions at cellular level:
  - the probability that a proliferating cell generates a new cell is a decreasing function of the tumor weight (**birth function**);
  - the probability that a proliferating cell becomes non-proliferating is an increasing function of the plasma i-th drug concentration (**damage function**);
  - the **time-to-death** of a non-proliferating cell is a random variable whose distribution reflects the nondeterministic delay between the i-th drug action and the cell death.
- We assume that:
  - a cell already damaged by one or more drugs can be damaged again by other drugs;
  - two or more drugs can hit the same cell and the associated damage processes evolve independently (only possible way to fulfill the no interaction hypothesis!).

Minimal model

$$\frac{dx_p(t)}{dt} = [\lambda_B(w(t)) - \sum_{i=1}^{K} \lambda_{D_i}(c_i(t))]x_p(t)$$

$$\frac{\partial y_{d_i}(t,s_i)}{\partial t} = \frac{\partial y_{d_i}(t,s_i)}{\partial s_i} + f_{s_i}(s_i)\lambda_{D_i}(c_i(t))x_p(t) - \sum_{j=1,j\neq i}^{K} \lambda_{D_j}(c_j(t))y_{d_i}(t,s_i), \quad i = 1,\ldots,K$$

$$\frac{\partial y_{d_{ij}}(t,s_i,s_j)}{\partial t} = \frac{\partial y_{d_{ij}}(t,s_i,s_j)}{\partial s_i} + \frac{\partial y_{d_{ij}}(t,s_i,s_j)}{\partial s_j} + f_{s_j}(s_j)\lambda_{D_j}(c_j(t))y_{d_i}(t,s_i) + f_{s_i}(s_i)\lambda_{D_i}(c_i(t))y_{d_j}(t,s_j) +$$
$$- \sum_{h=1,h\neq i,j}^{K} f_{s_h}(s_h)\lambda_{D_h}(c_h(t))y_{d_{ij}}(t,s_i,s_j), \quad i,j = 1,\ldots,K; i \neq j$$

$$\frac{\partial y_{d_{ijh}}(t,s_i,s_j,s_h)}{\partial t} = \frac{\partial y_{d_{ijh}}(t,s_i,s_j,s_h)}{\partial s_i} + \frac{\partial y_{d_{ijh}}(t,s_i,s_j,s_h)}{\partial s_j} + \frac{\partial y_{d_{ijh}}(t,s_i,s_j,s_h)}{\partial s_h} +$$
$$+ f_{s_j}(s_j)\lambda_{D_j}(c_j(t))y_{d_{ih}}(t,s_i,s_h) + f_{s_h}(s_h)\lambda_{D_h}(c_h(t))y_{d_{ij}}(t,s_i,s_j) +$$
$$+ f_{s_i}(s_i)\lambda_{D_i}(c_i(t))y_{d_{jh}}(t,s_j,s_h) - \sum_{l=1,l\neq i,j,h}^{K} f_{s_l}(s_l)\lambda_{D_l}(c_l(t))y_{d_{ijh}}(t,s_i,s_j,s_h),$$
$$i,j,h = 1,\ldots,K; i \neq j,h; j \neq h$$

$$\vdots$$

$$\frac{\partial y_{d_{1\ldots K}}(t,s_1,\ldots,s_K)}{\partial t} = \sum_{i=1}^{K} \frac{\partial y_{d_{1\ldots K}}(t,s_1,\ldots,s_K)}{\partial s_i} + \sum_{i=1}^{K} f_{s_i}(s_i)\lambda_{D_i}(c_i(t))y_{d_{\ldots(i-1)(i+1)\ldots}}(t,\ldots,s_{(i-1)},s_{(i-1)}\ldots)$$

$$w(t) = x_p(t) + \sum_{i=1}^{K}\int_0^\infty y_{d_i}(t,s)ds + \sum_{i,j=1,i\neq j}^{K}\int_0^\infty\int_0^\infty y_{d_{ij}}(t,s_1,s_2)ds_1ds_2 +$$
$$+ \sum_{i,j,h=1,i\neq j,h,j\neq h}^{K}\int_0^\infty\int_0^\infty\int_0^\infty y_{d_{ijh}}(t,s_1,s_2,s_3)ds_1ds_2ds_3 + \ldots$$

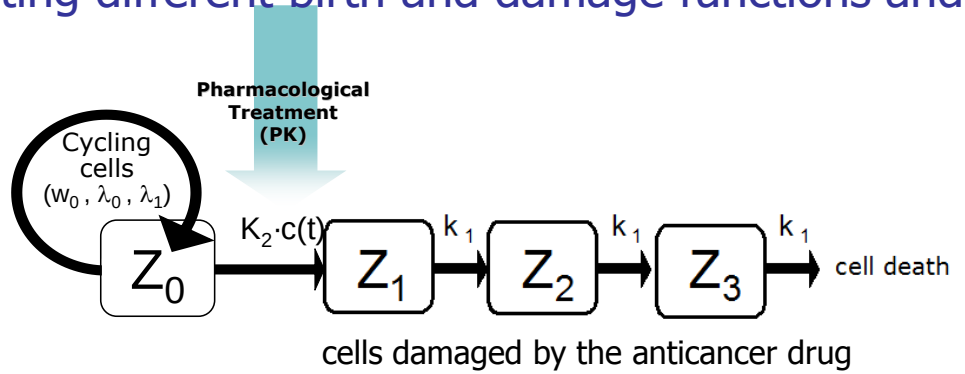
IEEE Trans. on Biomed. Eng. 59, 2161-2170, (2012)

A special case: the TGI_{add} model

- The minimal model defines a class of models, that can be specialized in several different models by properly selecting different birth and damage functions and time-to-death distributions.

The Simeoni TGI model

Cancer Research 64, 1094-1101, 2004



w_0: tumor weight at the inoculation time
λ_0 : exponential growth rate
λ_1 : linear growth rate
k_1: rate constant of transition
k_2: drug potency index

A special case: the TGI_{add} model

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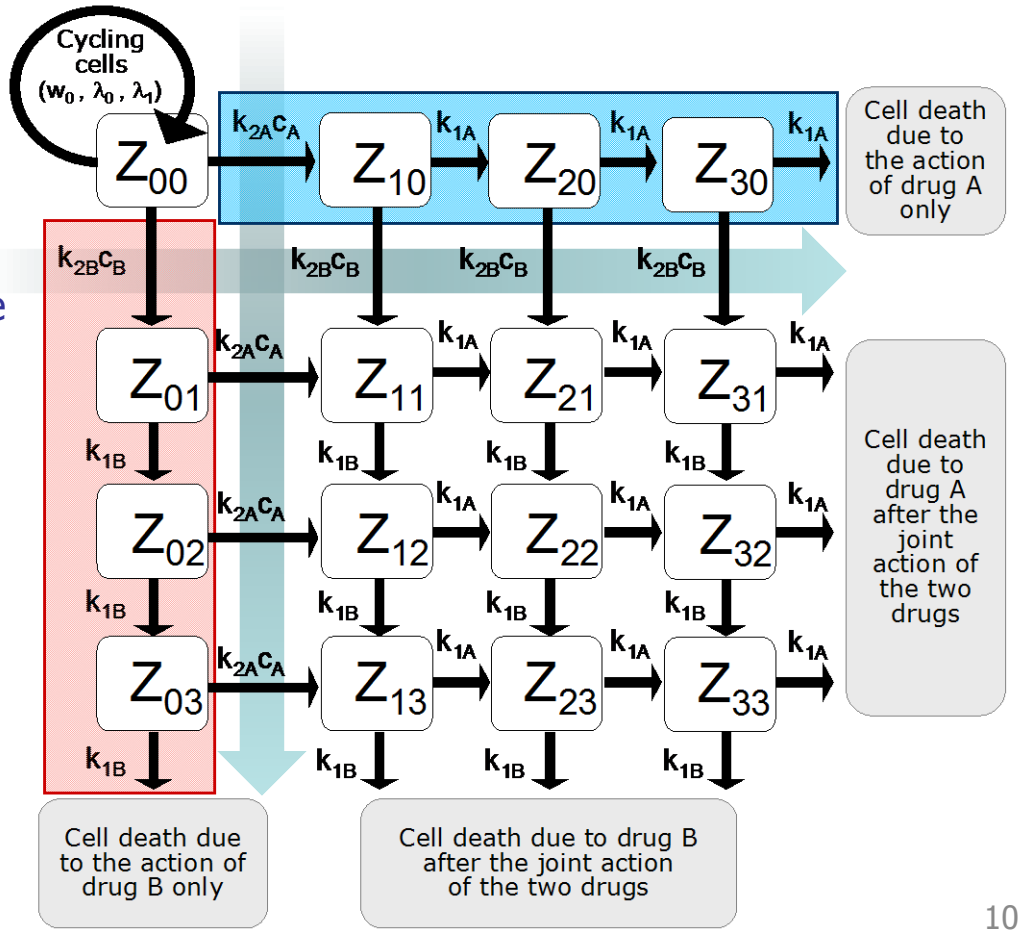
Considering the Simeoni TGI model and only two co-administered drugs

The TGI_{add} model

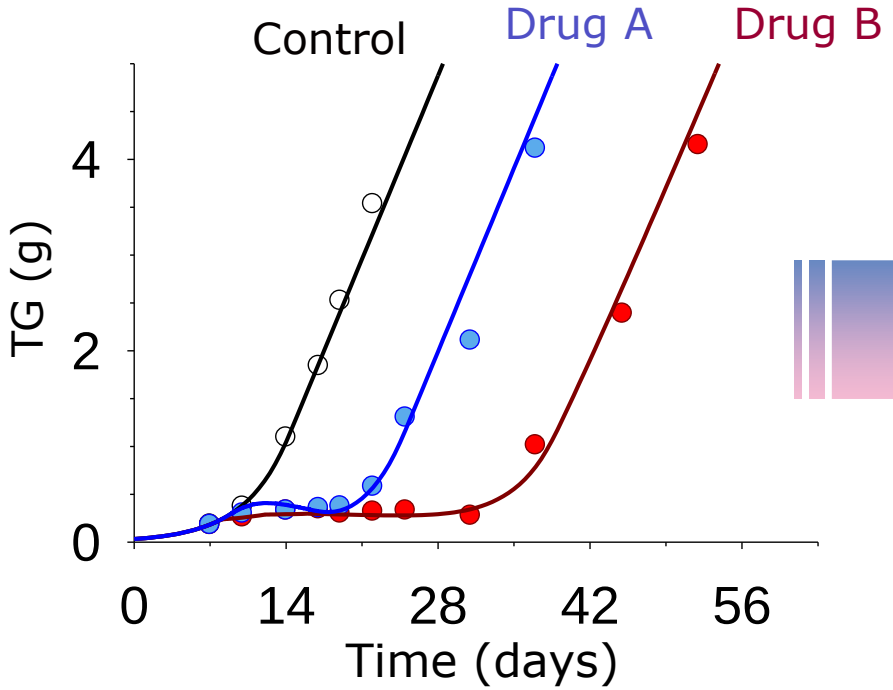
w_0: tumor weight at the inoculation time
λ_0 : exponential growth rate
λ_1: : linear growth rate
k_{1A}: rate constant of transition - Drug A
k_{2A}: potency index of Drug A
k_{1B}: rate constant of transition - Drug B
k_{2B}: potency index of Drug B

- Seven combination regimens involving five compounds have been tested.

Eur J of Cancer 45, 3336-3346, (2009)



Testing additivity (I)

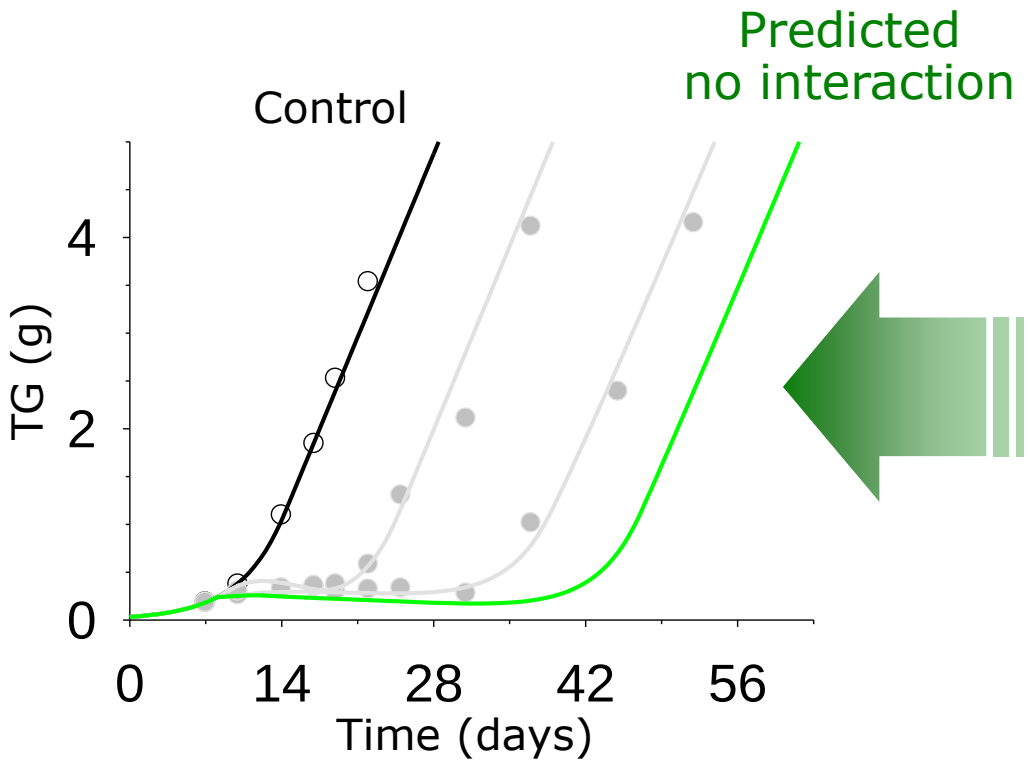


Single agent arms fitting (k_{1A}, k_{1B}, k_{2A}, k_{2B})

Parameter	Estimate	CV (%)
K_1 (day^{-1})	0.0951	38.97
K_1 (day^{-1})	0.879	24.37
K_2 ($\mu M^{-1} ml\ day^{-1}$)	1.83	9.51
K_2 ($\mu M^{-1} ml\ day^{-1}$)	0.306	11.17
λ_0 (day^{-1})	0.249	7.75
λ_1 (day^{-1})	0.274	8.68
W_0 (g)	0.0322	18.43

Testing additivity (II)

- Tumor growth curve generated by the model under the assumption of no interaction between drugs.



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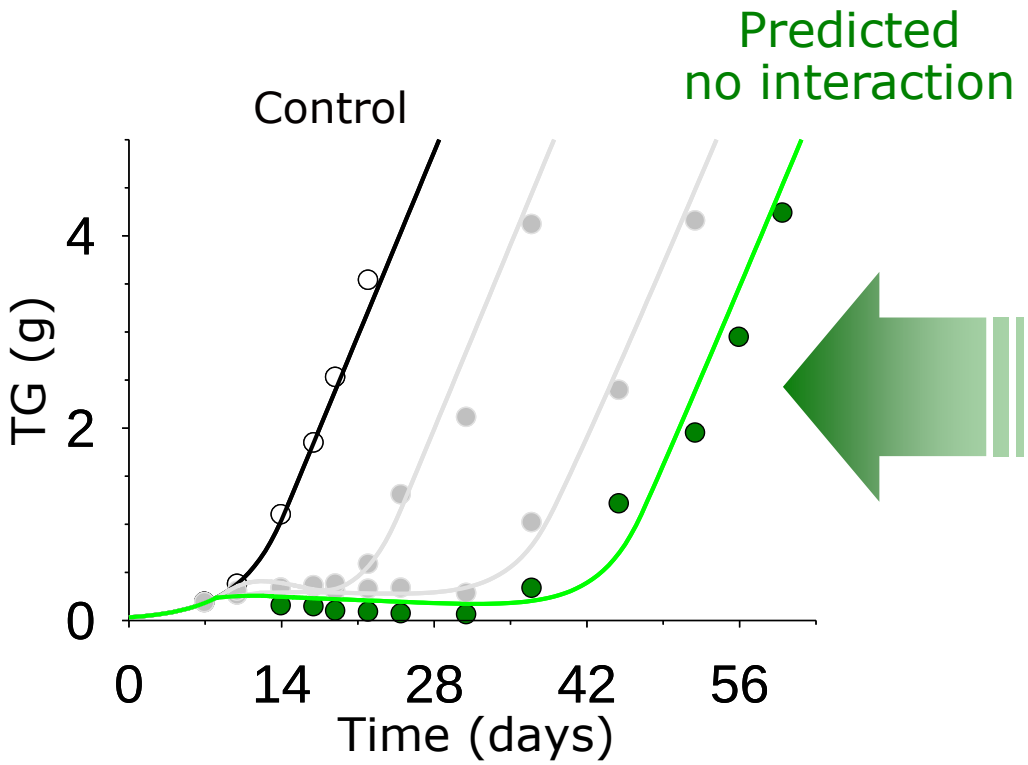

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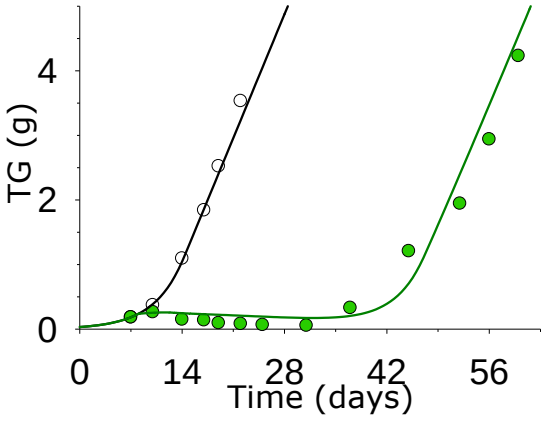
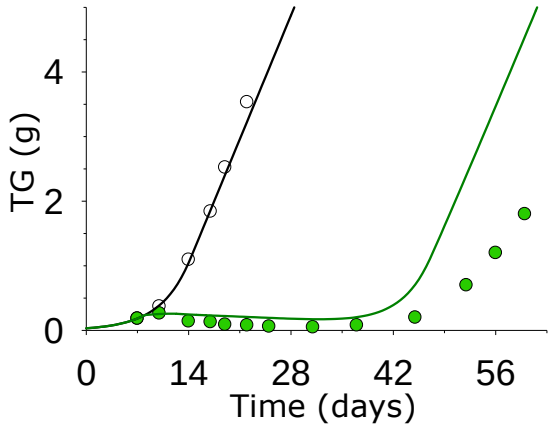
Testing additivity (III)

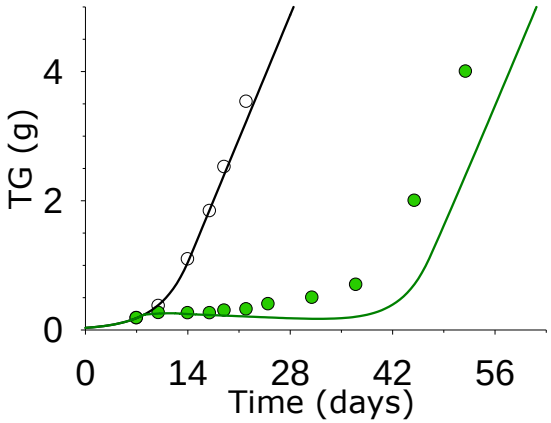
- By comparing the predicted curve with actual tumor weight data, possible departures from additivity can be ascertained:
  - tumor weights lying below or above the predicted additivity indicate synergism or antagonism



ADDITIVE EFFECT



SYNERGISTIC EFFECT



ANTAGONISTIC EFFECT

- Deviations from this behavior are evaluated comparing the predicted curves with experimental data through a statistical test (*χ2 statistics).*

Characteristics of the no interaction models

Cons

- No assessment of the strength of the interaction is given by the model.

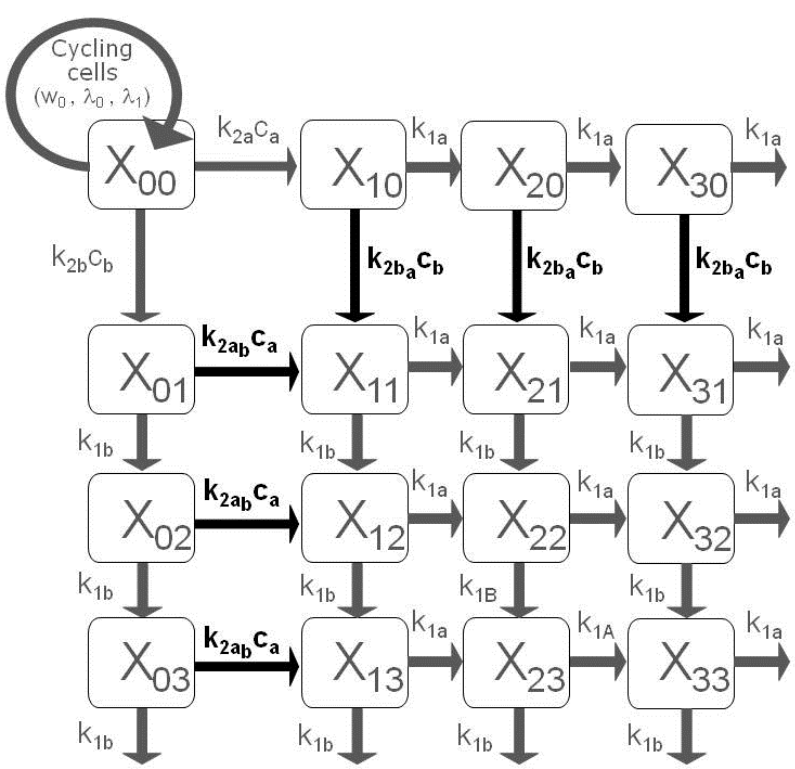
- Predictions of new administration schedules are not possible.

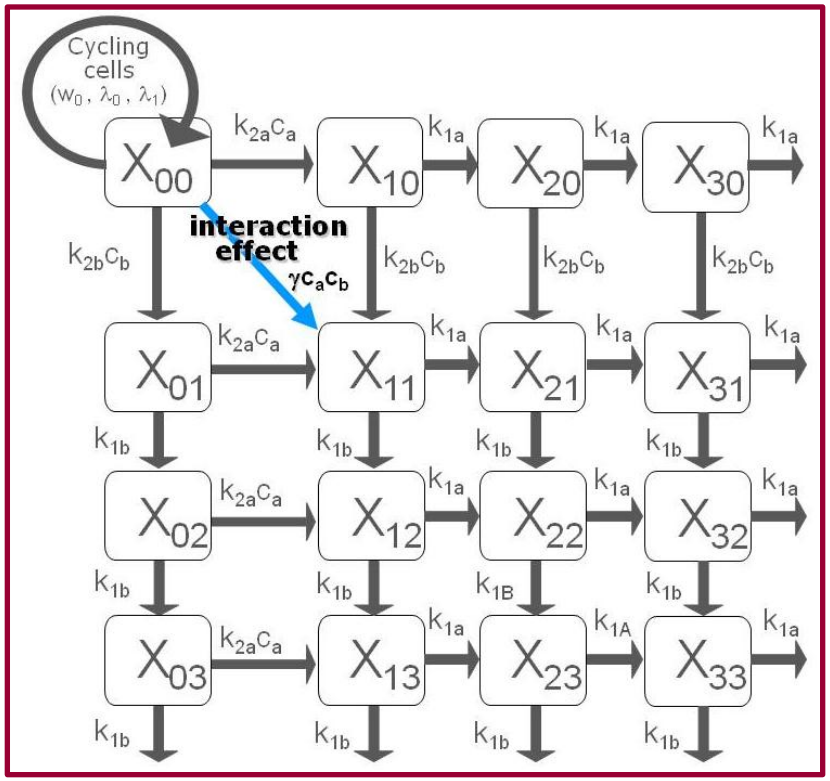
Pros

- The simulation of the no interaction behavior paves the way to the objective assessment of "additivity" and, then, to the characterization of the nature of a possible interaction (i.e., synergistic or antagonist).

- The architecture of no interaction model (e.g. the structure of mortality chains) plays a key role also in the architecture of the new developed combination model.

The TGI combination model



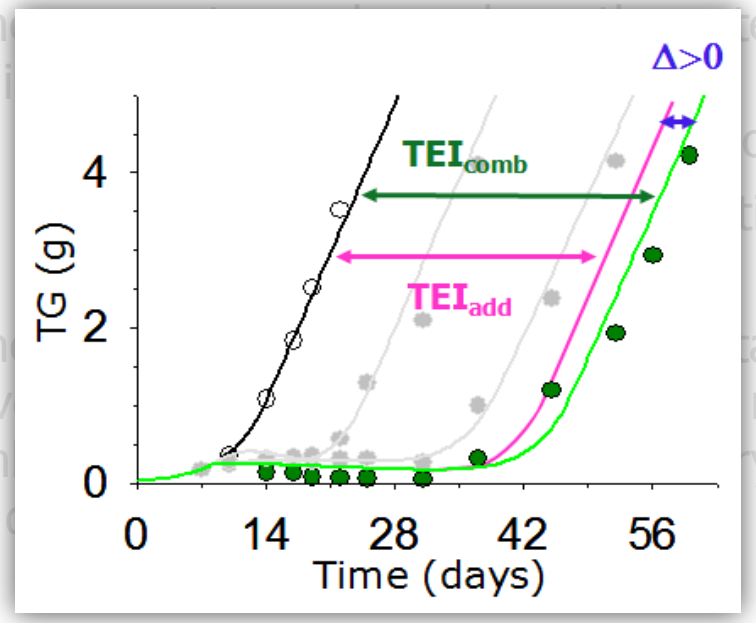


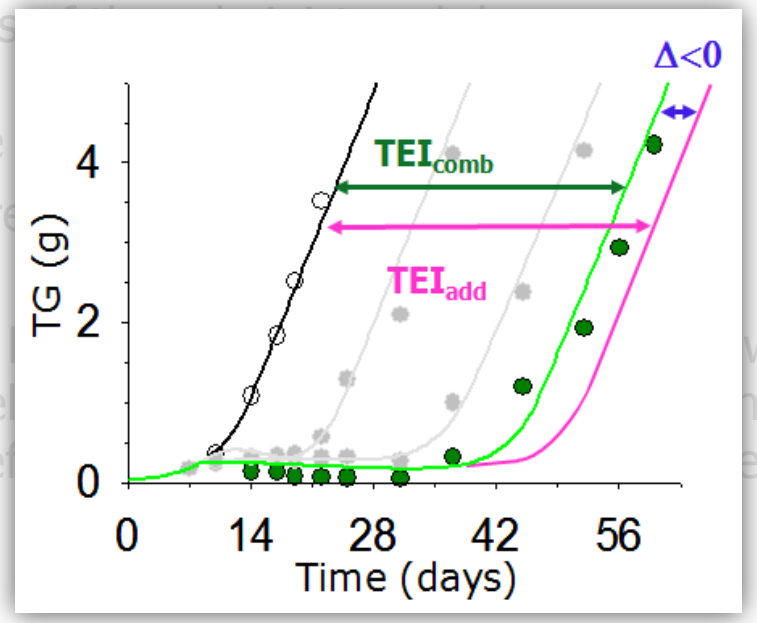
- If the value of γ is higher than, lower than or close to zero, the drug effect interaction has a synergistic, antagonistic or additive nature, respectively.
- Only the interaction effect on the proliferating cells can be significantly appreciated on the tumor mass dynamics.

Measurement of the interaction intensity

- The parameter γ depends on the potencies of the administered drugs.
- It is not possible to use it:
  - directly as an absolute measure of the interaction intensity;
  - for comparing different combination treatments and to rank them in accordance to it.
- The evaluation of the horizontal distance between the predictive tumor growth curve defined by the zero-interaction model and the curve obtained by the new combination TGI model provides a very useful index to quantify the contribution of the drug effect interaction.

Measurement of the interaction intensity





$$TEI_{comb} \simeq \frac{k_{2a}AUC_{c_a} + k_{2b}AUC_{c_b} + \gamma AUC_{c_a c_b}}{\lambda_0}$$

Portion due to the interaction term defined with Δ

Synergistic combination index (for Δ>0)

$$SC \equiv 100 \cdot \frac{\Delta}{TEI_{comb}}$$

Antagonistic combination index (for Δ<0)

$$AC \equiv 100 \cdot \frac{-\Delta}{TEI_{add}}$$

Model identification

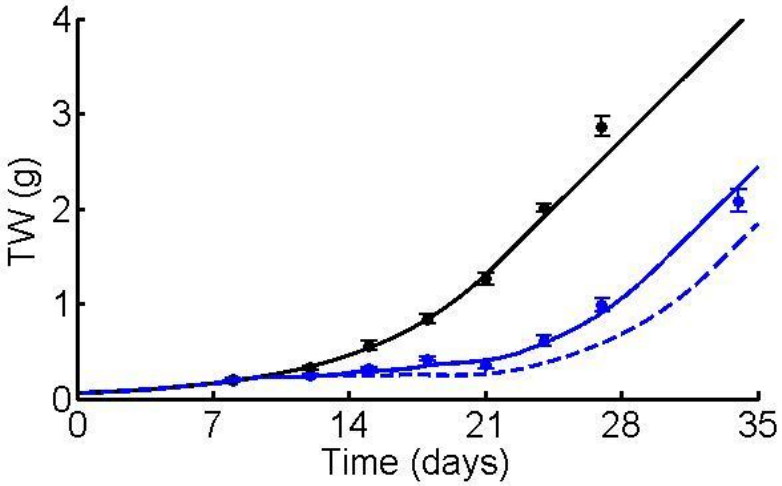
- The relevance and applicability of the proposed PK-PD model was demonstrated analyzing 11 studies involving three tumor cell lines, four new compounds as well as four drugs already on the market.

Drug A	Drug B	Cell line	Combination arms
Drug C1	Gemcytabine	BxPC3	One combination arm
	Cisplatin	A2780	Two combo – different schedule
Drug C2	Irinotecan (CPT-11)	HT29	Two combo – different dose
	5-fluoracil (5-FU)	HT29	Two combo – different dose
Drug C4	Gemcytabine	BxPC3	Two combo – different dose
Drug C5	Irinotecan (CPT-11)	HT29	Two combo – different dose

- Similar model identification strategy as for the TGI_{add} model.

Drug C1 given in two combination experiments

Drug C1 + Gemcytabine



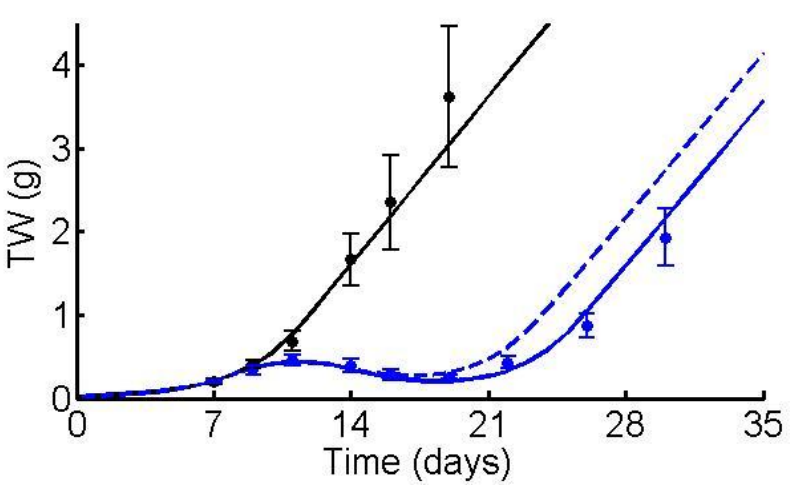
$\gamma = -0.88 \mu M^{-2} day^{-2} (CV = 10\%)$

$RMSE = 0.19 g$

$AC = 24\%$

Significant role of negative interaction

Drug C1 + Cisplatin



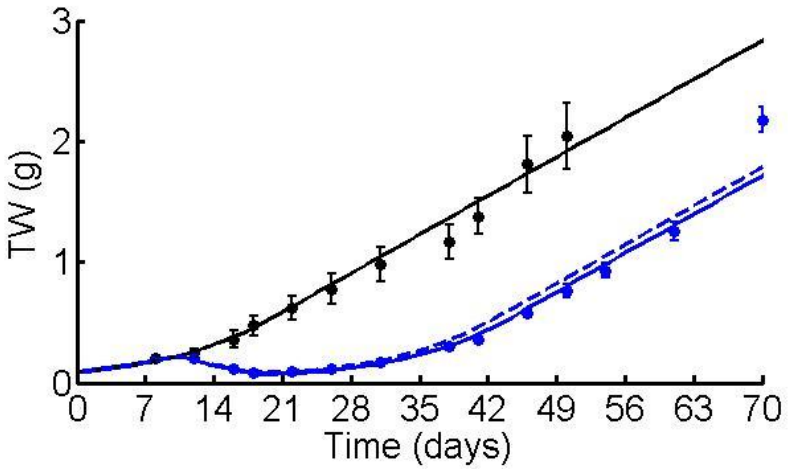
$\gamma = 8 \mu M^{-2} day^{-2} (CV = 15\%)$

$RMSE = 0.17 g$

$SC = 14.7\%$

Moderate synergistic effect

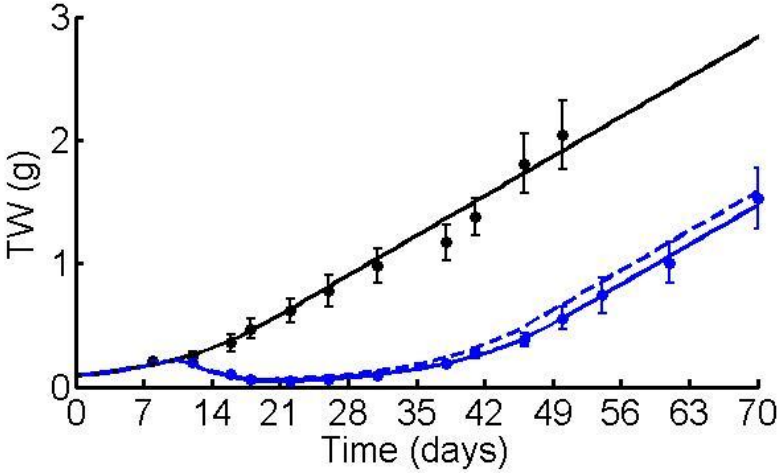
Drug C2 and 5-FU given in two combination regimens



$\gamma = 0.131 \mu M^{-2} day^{-2} (CV = 42\%)$

$RMSE = 0.11g$

$SC = 6\%$



$\gamma = 0.157 \mu M^{-2} day^{-2} (CV = 36\%)$

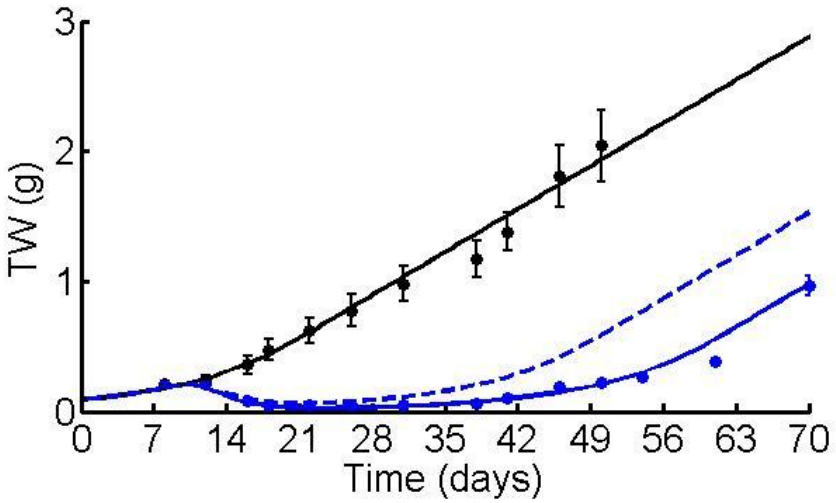
$RMSE = 0.06g$

$SC = 8\%$

Very limited synergistic interaction effect

A changing of about 33% in the dose level of Drug C2 does not affect significantly the interaction intensity!

Drug C2 given in combination with CPT-11



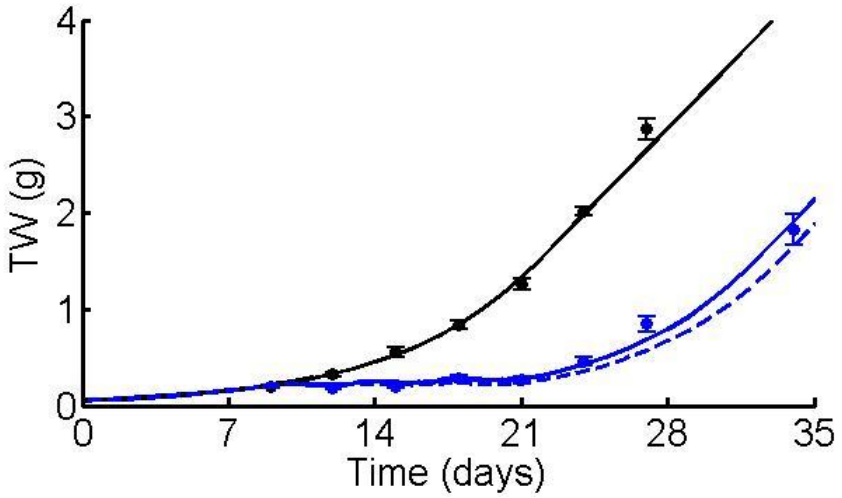
$\gamma = 3.45 \mu M^{-2} day^{-2} (CV = 11\%)$ $RMSE = 0.07 g$

$SC = 29\%$

Significant synergistic interaction effect

Almost one-third of the total inhibition is due to the interaction effect.

Drug C4 given in combination with Gemcytabine

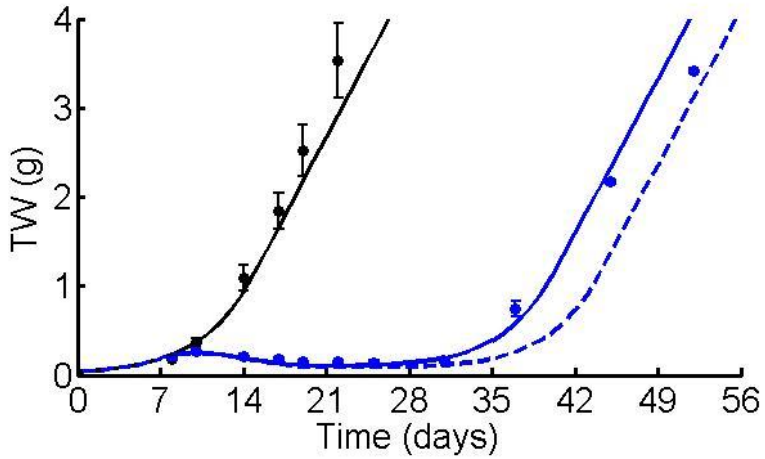


$\gamma = 0.045 \mu M^{-2} day^{-2} (CV = 28\%)$ $RMSE = 0.09g$

$AC = 10\%$

Slight antagonistic interaction effect

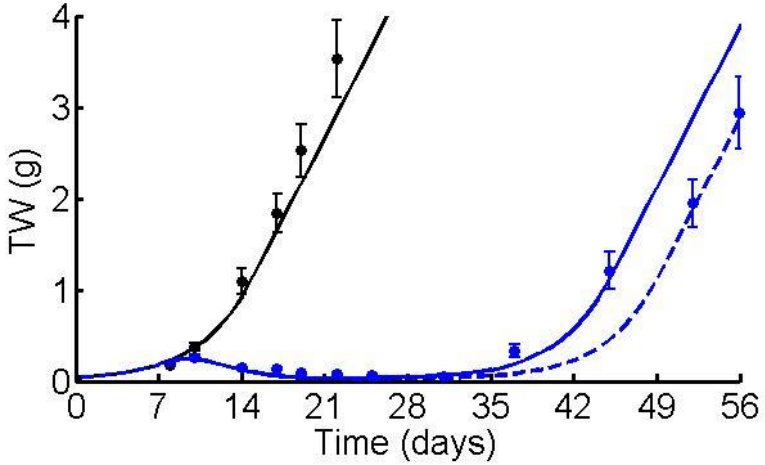
Drug C5 and CPT-11 given in two combination regimens



$\gamma = -2.04 \mu M^{-2} day^{-2} (CV = 18\%)$

$RMSE = 0.25g$

$AC = 14\%$



$\gamma = -1.05 \mu M^{-2} day^{-2} (CV = 30\%)$

$RMSE = 0.36g$

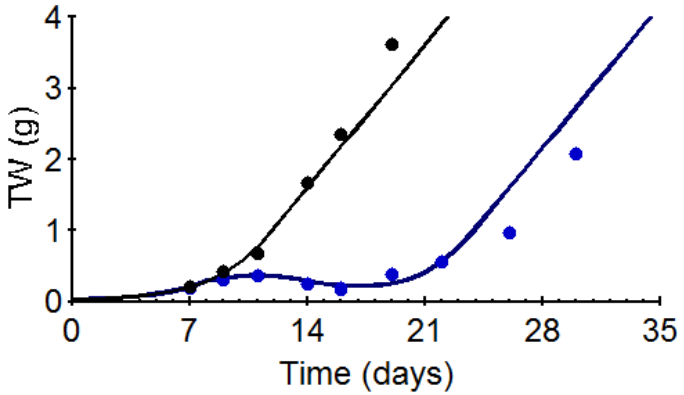
$AC = 12\%$

Possible antagonistic interaction effect

Although the two interaction parameters γ are two fold in the two conditions the combination indexes are very similar.

TGI predictive power

Drug C1 + Cisplatin

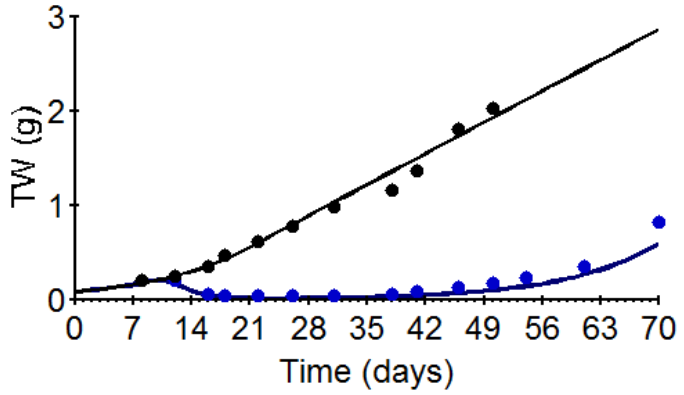


$SC_{predicted} \approx 0\%$

$SC_{estimated} = 8\%$

Different schedules

Drug C2 + CPT-11

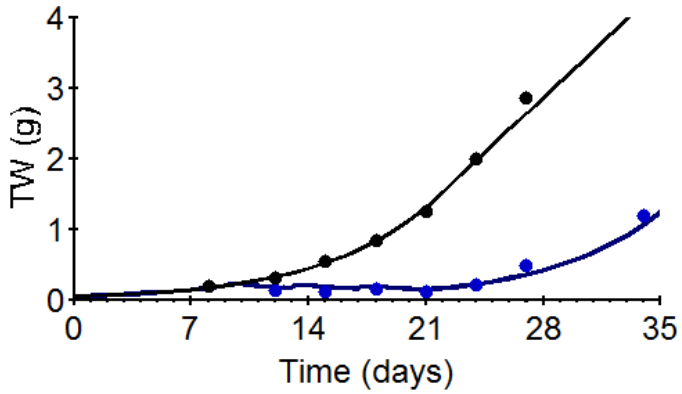


Different doses
(30% dose increasing)

$SC_{predicted} = 32\%$

$SC_{estimated} = 21\%$

Drug C4 + Gemcytabine



Different doses
(100% dose increasing)

$AC_{predicted} = 14\%$

$AC_{estimated} = 9\%$

Achievements

- The minimal model represents a general class of models, that provide the reference growth curve under the hypothesis of no interaction.

- The proposed PK-PD model is able to characterize the drug potency and the drug effect interaction independently from dose levels and schedules.
  - Model prediction capabilities demonstrated analyzing a large number of studies.
  - Useful tool to facilitate the optimization strategies in combination therapies, thus reducing time and costs.

- This approach is of practical use as it can be applied to assess combination therapy in standard xenograft experiments and it enables to identify synergistic drug combinations.

- It is used in Nerviano Medical Science Labs to analyze routinely performed experiments.

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ORIGINAL ARTICLE

A predictive pharmacokinetic–pharmacodynamic model of tumor growth kinetics in xenograft mice after administration of anticancer agents given in combination

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