Supplementary material: Mathematical modeling predicts response to chemotherapy and drug combinations in ovarian cancer

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Text S1 Mathematical model

Model structure

In this section, details of the mathematical model are presented. To model tumor growth and the dynamics of drug resistance to platinum in patients with high-grade serous ovarian cancer (HGSOC), we developed a stochastic continuous time multi–type branching process model. In this model, during each time step, a cell can i) divide to produce two daughter cells that are identical to the mother cell, ii) divide to produce one daughter cell which is identical to the mother cell and one cell with one additional resistance mechanism, iii) die to produce no cell. Schematic of the model is shown in Figure 1.

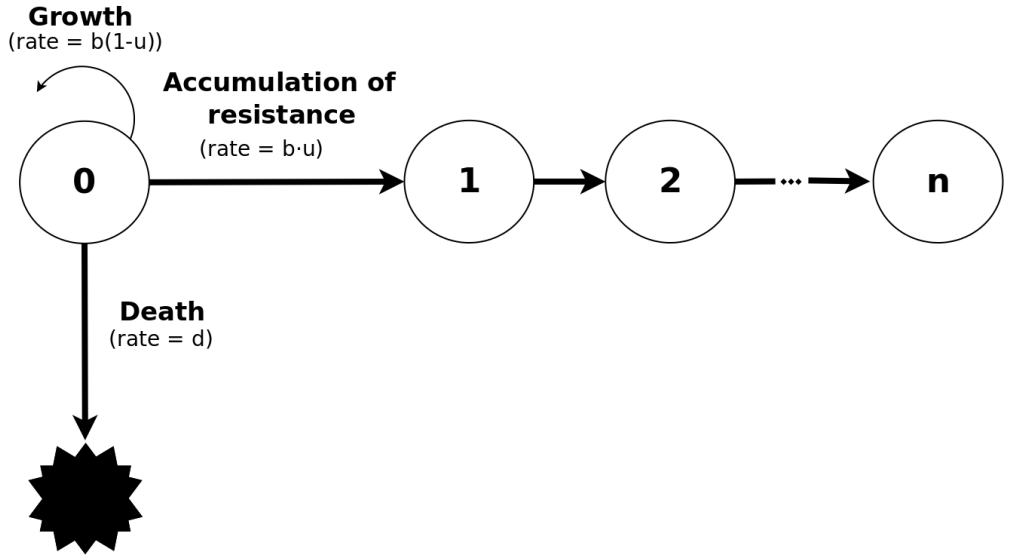

Figure 1: Schematics of the model.

First, we describe the tumor growth without treatment intervention. In this case, all type of cells grow with a positive net growth rate $\lambda = b - d > 0$. We assume that cells with mutations divide with the same rate as wild-type (sensitive) cells. In other words, resistance is neutral in absence of treatment. Sensitive cells grow exponentially with birth rate b and die with death rate d. In addition, wild-type cells have probability u per cell division to acquire one resistance mechanism leading to new subclone. Earlier studies defined parameter u as mutation rate, where 'mutation' is used collectively to include all genetic aberrations that can occur during cell division. While mutations play a major role in targeted drug resistance, in platinum resistance other aberrations, such as genomic instability and copy number alterations, are important. Thus, in our approach u is referred to as transition rate to indicate the fact that resistance is not solely determined by mutations. Finally, resistance accumulates sequentially. That is, each cell division event leads to the accumulation of one new resistance mechanism at a constant rate.

A tumor is diagnosed when the colony reaches M cells, after which the primary treatment starts. Two types of intervention are included in the model: surgery and chemotherapy. Surgery is modeled as removal of a fraction of β cancer cells. All cell types can be eliminated by surgery with equal probability. Chemotherapy is modeled by increasing death rate of sensitive cells proportionally to division rate. That is, during treatment sensitive cells die at rate $d' = d + b \cdot d_{chemotherapy}$, where $d_{chemotherapy}$ is the chemotherapy effect.

Chemoresistance is not a binary but a quantitative factor, and we have taken this into account by allow-

ing the model to contain cells with different (accumulative) resistance mechanisms. Thus, chemotherapy effect ($d_{chemotherapy}$) for a cell with n resistance mechanisms accumulated is: $d_{chemotherapy} = d_{sensitive} \cdot (0.01)^n$, where $d_{sensitive}$ is the value of chemotherapy effect for sensitive cells. Thus, chemotherapy effect decreases exponentially as a function of the resistance mechanisms.

Computer simulation

We performed Monte Carlo computer simulation using an approximation of exact stochastic simulation algorithm developed by Gillespie [3]. We used an approximation algorithm due to its computational efficiency.

First, slow events (unfaithful division) are separated from fast events (faithful division, cell death). The propensities of slow reactions are: $a_i(x)$, for $i = 1, 2 \ldots n$, where n is the number of slow events. The propensities of fast reactions are: $b_i(x)$ for $j = 1, 2 \ldots m$, where m is the number of fast events. Next, time of the next slow reaction is calculated:

$$\tau = \frac{1}{\sum_{i=1}^{n} a_i(x)} ln(\frac{1}{R_1}), \tag{S1}$$

where R_1 is a random number from a unit-interval uniform distribution. Next, the index of next slow event is given as the smallest k satisfying:

$$\sum_{k=1}^{n} a_k(x) > R_2 \cdot \sum_{i=1}^{n} a_i(x), \tag{S2}$$

where R_2 is a random number from a unit-interval uniform distribution. The system is then updated and fast events that occur until time of the first slow event τ. Fast events are updated using tau-leaping, *i.e.,* for every fast event the number of times each event occurs during the time interval $[t, t + \tau)$ is:

$$k_j = Poisson(a_j \cdot \tau) \quad , for\ j = 1, 2, \ldots, m \tag{S3}$$

and the system is updated:

$$x(t + \tau) = x(t) + \sum_{j} k_j \cdot v_j, \tag{S4}$$

where v_j is the state change vector.

Key assumptions of the model

1. Cells are growing exponentially with net growth rate λ.
2. Cancer is detected when the colony reaches a certain size (M).
3. The mutated cells grow at the same rate as wild–type cells, *i.e.*, they are neutral in absence of treatment.
4. The resistance mechanisms accumulate sequentially and with equal contribution to resistance.
5. Rate of accumulation of resistance in absence of treatment is the same as during the treatment.
6. The effect of chemotherapy is modeled by the drug–induced death rate, $d_{chemotherapy}$, which leads to negative net growth rate: $\lambda = b - d < 0$.
7. Surgery is modeled by removal of a fraction of cancer cells. All cell types can be removed with equal probability.

Text S2 Standard-of-care simulations

To investigate chemotherapy resistance, we performed computer simulations of the model. The simulation consists of three phases: 1) pre–treatment phase in which the tumor grows until diagnosis, 2) treatment phase that includes platinum-based chemotherapy and surgery, and 3) post–treatment phase where the tumor grows until relapse.

The first phase starts with a single sensitive cell and proceeds with exponential tumor growth in the absence of treatment interventions. The phase ends when the total number of tumor cells reaches M cells. The tumor is then diagnosed and the treatment phase starts. Treatment includes two types of interventions: Chemotherapy and surgery. Chemotherapy is modeled by reducing the growth rate of sensitive and partially–resistant cells. Surgery is modeled by removing a fraction of tumor cells at one time point. The treatment phase is simulated in a framework that follows the first-line standard-of-care therapy guidelines for HGSOC. That is, in our simulations, the HGSOC patient is treated with three cycles of neoadjuvant chemotherapy (NACT). Next, a debulking surgery is performed aiming for optimal cytoreduction, followed by another three cycles of adjuvant chemotherapy. If tumor reduction after NACT is below 40%, we assume that the patient does not respond to chemotherapy and platinum–free interval is zero. The last phase of simulation starts after the treatment and ends at the time of the first recurrence. In this phase of simulation, the tumor grows without perturbation until the total number of cells reaches $M_{relapse}$ cells.

Text S3 In-silico analysis of patient response to a therapy

We performed *in silico* treatment response analysis using the simulation approach described in Supplementary Text 2. Each virtual HGSOC patient is described with two parameters: tumor burden at diagnosis M and chemotherapy effect $d_{chemotherapy}$.

For each virtual HGSOC patient, we sampled tumor burden at diagnosis (M)from a log–normal distribution. Also chemotherapy effect ($d_{chemotherapy}$) was sampled from a log–normal distribution. The log–normal distribution was selected because it describes the best distributions for M and $d_{chemotherapy}$ among six tested statistical distributions (details in Supplementary Text S4).

We simulated each virtual HGSOC patient by calculating platinum–free interval (PFI) estimates. With the PFI estimates in a cohort of 1,000 virtual patients, we performed Kaplan–Meier analysis to obtain platinum–free interval plots. Kaplan–Meier analysis was carried out in MATLAB using ecdf function with no censoring.

Text S4 Parameter calibration

Tumor growth

The average cell cycle duration in an ovarian cancer cell is 36 hours [4] and it is defined as the time between cell divisions in the absence of cell death. This implies that the division rate is: $b = \frac{1}{36}[\frac{1}{hour}] = 0.667[\frac{1}{day}]$. Late–stage HGSOC tumors double in volume approximately every 2.5 months [1], leading to net growth rate $\lambda = \frac{log(2)}{DT} = 0.0058[\frac{1}{day}]$, where DT is the doubling time. To achieve the observed λ, we set the cell death rate to $d = \lambda - b$. In our simulations, the parameters b and d are constant for all cell types.

Size of tumors at clinical diagnosis

We modeled the probability of diagnosis as a function of tumor size as follows. First, we estimated tumor burden using metabolic tumor volume (MTV) values extracted from ^{18}F-FDG-PET/CT images. Next, the MTV values were converted to a number of cells assuming 10^9 cells in $1cm^3$ tumor bulk according to [2]. The number of cells was fitted to normal, exponential, Weibull, log–normal, logistic, and log–logistic probability distributions. The Bayesian Information Criterion (BIC) was calculated to measure the goodness of fit. The best agreement with the data was obtained for log–normal probability distribution with parameters $\mu = 26.59$ and $\sigma = 0.47$. Converting μ of the log–normal distribution to μ of the normal distribution, the total number of cancer cell at diagnosis equals $M = 3.9548 \cdot 10^{11}$ cells.

Chemotherapy effect

Patients in the calibration cohort received a median of three cycles of NACT where each cycle is 21 days. This means $\tau_{NACT} = 63$ [days] of neoadjuvant treatment. Without loss of generality, we can assume that τ_{NACT} is also the time interval between acquisition of PET/CT images before and after NACT. Parameter $d_{chemotherapy}$ was estimated for each patient separately as follows. We set all parameters except $d_{chemotherapy}$ and M to default values listed in Table 2. Next, for each patient we extracted a number of cells before (M) and after (M_{NACT}) chemotherapy. In addition, we know that time between two PET/CT scans is $\tau_{NACT} = 9$ [weeks]. Thus, we estimated $d_{chemotherapy}$ numerically with bisection method to find minimum of the following objective function:

$$J = \hat{M_{NACT}} - M_{NACT} = \sum_{i=1}^{n} x_i(t_{NACT}) - M_{NACT}.$$

Debulking surgery effect

The optimal debulking surgery is defined as a residual disease of 10 [mm] or less [5]. Tumor with 10 mm diameter corresponds to tumor volume of 10 [cm^3]. Ovarian cancer tumor is diagnosed when tumor burden is of order 10^{11} cells. Thus, optimal surgery removes at the minimum fraction of $\beta\frac{10^9}{10^{11}}$ and corresponds to two cell–log kill fraction of cells removed by surgery.

Rate of resistance accumulation

We utilized clinical data from our cohort to estimate the value of u. From the calibration cohort we excluded 12 patients who did not proceed to IDS after NACT and 17 patients who did not progress during follow-up time, leaving 33 patients. Next, we performed Kaplan–Meier analysis. By iterative

model calibration, we chose the values of u using the smallest deviation between patient data and the model prediction.

Parameter u was estimated by fitting to survival plot in the calibration cohort. Each patient was simulated as explained in Supplementary Text 2 and 3. Next, a cohort of 1000 "virtual patients" was created, and based on the created cohort, we performed Kaplan–Meier analysis. The obtained platinum–free interval plot was compared with the plot from the calibration cohort. Mean squared error (MSE) was calculated to measure deviation between the two survival estimates. The simulation was performed for a wide range of transition rate $10^{-8} - 10^{-4}$. The computation was repeated 100 times and the mean value of MSE was calculated. The smallest MSE was obtained for parameters listed in Table 2 in the manuscript.

Text S5 Sensitivity analysis method

Local sensitivity analysis

Parameter sensitivity analysis is a valuable tool to explore the impact of the parameters to outcome. We applied local sensitivity analysis to find the key parameters that affect platinum-free interval values.

The relative sensitivity coefficient of a response Y at time t with respect to a parameter p_i is [6]

$$S_i(t) = \frac{\partial Y}{\partial p_i} : \frac{Y}{p_i} \approx \frac{\Delta Y_i}{Y_i} : \frac{\Delta p_i}{p_i}, \quad for\ \Delta p_i \to 0 \tag{S5}$$

where ΔYi represents the change in Y with perturbation Δp_i

Then, the sensitivity coefficient of the survival percentage with respect to parameter p_i was calculated according to formula:

$$S_i = \frac{\int_0^T (Y(t,p_i) - Y(t,\tilde{p_i}))dt}{\int_0^T (Y(t,p_i)dt} : \frac{\Delta p_i}{p_i} = \int_0^T S_i(t)dt \tag{S6}$$

where $Y(t,p_i)$ and $Y(t,\tilde{p_i})$ represents value of response Y with parameter p_i and varied parameter $\tilde{p_i}$ at time t.

We performed a local sensitivity analysis to single-parameter perturbation. Each parameter was increased by 0.1% from its estimated value (listed in Table 2). The parameter perturbation was chosen to be small enough to reduce error in numerical approximation of sensitivity coefficient. Then, we calculated relative change in area under platinum-free interval survival plot curve by solving numerically equation (S6). The calculations were repeated 50 times and the average sensitivity coefficient was calculated.

References

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Supplementary figure 1

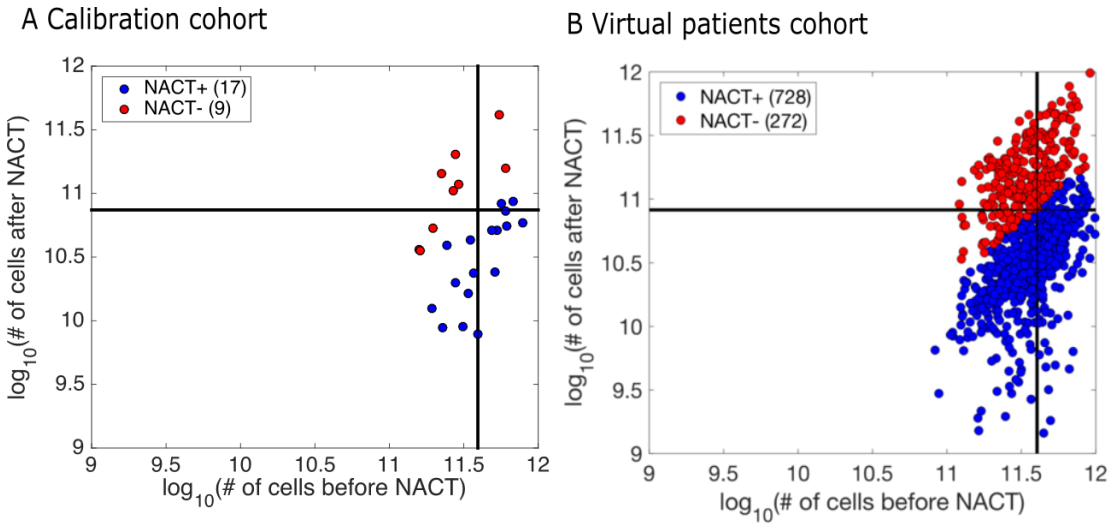

Figure S1: Initial response to platinum chemotherapy. The plots show tumor burden before and after NACT for two cohorts: calibration cohort of 26 patients and 1,000 virtual patients. Black line represents average number of tumor cells before and after NACT. Patients were stratified into two groups: $NACT^{+}$ (tumor reduction above average) and $NACT^{-}$ (tumor reduction below average). Virtual patient cohort faithfully reproduces the patient responses to platinum-based chemotherapy observed in our calibration cohort.