CAR**T***math*

Overview

CAR**T***math* is a user-friendly interface using R (R CORE TEAM, 2020) package Shiny (CHANG et al., 2020) to run the three population mathematical model developed in Rodrigues (2019). This model describes the tumor response to CAR-T cell immunotherapy in immunodeficient mouse models. It encompasses interactions between tumor cells, effector and long-term memory CAR-T cells such as tumor-induced immunosuppression, memory pool formation, conversion of memory T cells into effector T cells in the presence of tumor cells, and individual specificity considered as uncertainties in the model parameter values (RODRIGUES et al., 2020). Other simulation results are already published in Barros et al. (2020).

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Basic usage

Running CART*math*: users can choose a pre-set dataset or customize every option. Datasets generate the same figures from Rodrigues et al. (2020). On "Mathematical Model" one can view the model to be simulated. To generate the graph result, click the "Run Simulation" button on "Simulation". Wait for a few seconds and the corresponding simulation result will be displayed.

Running CART*math* Customized mode: to customize your run, choose one of the "Customized" options on Dataset box, and make any changes on parameters as tumor or CAR-T cell numbers.

Modifying previous simulations: after choosing a pre-set dataset, users can modify any basic or advanced parameters. To generate a new graph, click the "Run Simulation" button on "Simulation". Wait for a few seconds and the new graph will be displayed.

Graphs download directly from SETUP page: pass the mouse over the graph and click on the "camera" symbol. Download the graph on png format.

Graphs customizations: at the "Graphs" page, users can customize graph "y" and "x" axis labels, scales, and formats for each population in an independent plot. Line width, color, and grid can also be changed. The graph with the three populations will acquire all changes made by each population graph independently and it is possible to choose by a single or double y-axis.

Table download: at the "Table" page, users can visualize and download the table with the simulation values for each cell population. For simplification, one value for each day is displayed for each population. The ordering of the lines in increasing or decreasing values can be changed by clicking the arrows next to each population title. The simulation results are available for download in the form of tables in csv format containing all the simulation steps ("Download full table" button) or in a reduced version ("Download summarized table" button). In the full table, there is a printing frequency to avoid the generation of very large files, but without compromising accuracy.

Report: a report is generated for each simulation containing the values of all basic and advanced model settings and a graph with the three cell population dynamics. Users must press the "Generate report" button to generate the report before downloading it, by clicking on the "Download report" button.

CAUTION: in the Customized mode, the advanced parameters assigned *a priori* must be carefully evaluated. Different types of tumor and CAR-T cells may require significant variations in their values. A model calibration widget will be available in the next version of CAR**T***math*.

Available Datasets

I. CAR-T Immunotherapy in HDLM-2 tumor (RODRIGUES et al., 2020 – Figure 3a)

Hodgkin's Lymphoma tumor cell lineage HDLM-2 in NSG mice and CAR-T 123 immunotherapy (against CD123 antigen). Experimental data from Ruella et al. (2017) - Figure 4.

Initial conditions:
Tumor cell number (T): 2×10^6 cells.
CAR-T cell number (C_T): 1.5×10^6 cells.
CAR-T cell injection on day 42.

II. CAR-T Immunotherapy with Challenge in HDLM-2 tumor (RODRIGUES et al., 2020 – Figure 2a)

Hodgkin's Lymphoma tumor cell lineage HDLM-2 in NSG mice and CAR-T 123 immunotherapy. Challenge on day 250. Experimental data from Ruella et al. (2017) - Figure 5.

Initial conditions:
Tumor cell number (T): 2×10^6 cells.
CAR-T cell number (C_T): 2×10^6 cells.
CAR-T cell injection on day 42.
Challenge with 1×10^6 tumor cells on day 250.

III. CAR-T Immunotherapy with Fractionated Doses in HDLM-2 tumor (RODRIGUES et al., 2020 – Figure 3d)

Hodgkin's Lymphoma tumor cell lineage HDLM-2 in NSG mice and CAR-T 123 immunotherapy applied according to a hypothetical scenario of dose fractionation.

Initial conditions:
Tumor cells (T): 2×10^6 cells.
CAR-T cell (C_T): 5×10^5 cells/dose.
CAR-T cell injection on days 42, 49, 56, 63.

IV. CAR-T Immunotherapy in RAJI tumor (RODRIGUES et al., 2020 - Figure 2b)

RAJI tumor cell in SCID/Beige mice. CAR-T 19 immunotherapy (against CD19 antigen). Experimental data from Ninomiya et al. (2015) - Figure 2.

Initial conditions:
Tumor cells (T): 3×10^6 cells.
CAR-T cell (C_T): 1×10^7 cells.
CAR-T cell injection on day 7.

V. CAR-T Immunotherapy in RAJI-IDO$^+$ tumor (RODRIGUES et al., 2020 - Figure 5a)

RAJI-IDO$^+$ tumor cell in SCID/Beige mice. CAR-T 19 immunotherapy. Experimental data from Ninomiya et al. (2015) - Figure 3.

Initial conditions:
Tumor cells (T): 3×10^6 cells.
CAR-T cell (C_T): 1×10^7 cells.
CAR-T cell injection on day 7.

VI. **CAR-T Immunotherapy with IDO inhibitor (1-MT) in RAJI-IDO$^+$ tumor (RODRIGUES et al., 2020 - Figure 5b)**

RAJI-IDO$^+$ tumor cell in SCID/Beige mice. CAR-T 19 immunotherapy with IDO inhibitor (1-MT). Experimental data from Ninomiya et al. (2015) - Figure 3.

Initial conditions:
Tumor cells (T): 3×10^6 cells.
CAR-T cell (C_T): 1×10^7 cells.
CAR-T cell injection on day 7.

VII. **"Custom slow growth (HDLM-2)" or "Custom rapid growth (RAJI)"**

By choosing one of these options, the user can modify any basic or advanced settings for the slow tumor growth (refered to the Hodgkin's Lymphoma tumor cell lineage HDLM-2) or rapid tumor growth (refered to the RAJI tumor cell lineage).

Basic Parameters

Dataset: choose a pre-set dataset or a full customized option.

Maximum simulation time (days): set the maximum simulation time wanted to evaluate CAR-T cell immunotherapy results.

Tumor cell number (x10^6): set the initial tumor cell dose injected into the mice.

Dose type: set if it will be administrated a single dose of CAR-T cell or several doses (Fractionated). If Fractionated dose is selected, set the number of doses to be performed.

CAR-T cell number (x10^6): set the CAR-T cell dose injected into the mice. If Fractionated dose is selected, set each corresponding dose.

Day of CAR-T cell injection: set the day after tumor injection when CAR-T cell are inoculated. If Fractionated dose is selected, set the injection day of each dose.

Challenge: assign challenge if a second tumor cell injection will be performed.

Challenge day: set how many days after the first tumor injection a second injection will be performed.

Number of tumor cell in challenge (x10^6): set how many tumor cells will be injected at the challenge.

Advanced Parameters

Advanced parameters estimation was performed with *in vitro* and *in vivo* experimental results from Ruella et al. (2017) to the Hodgkin's Lymphoma tumor cell lineage HDLM-2 and Ninomiya et al. (2015) to the RAJI tumor.

- **Tumor proliferation rate (r)**: $ln(2)/td$, in which "td" is the time (in days) it takes for the number of tumor cells to double. Unit: 1/day.
- **Tumor death by CAR-T (γ)**: probability of tumor cell death per day due to contact with effector CAR-T cells. Unit: 1/(cell.day).
- **Inverse of tumoral support capacity (b)**: the inverse of the maximum number of tumor cells that the environment can sustain. Unit: 1/cell.
- **CAR-T proliferation rate (ϕ)**: $ln(2)/tc$, in which "tc" is the time (in days) it takes for the number of CAR-T cells to double. Unit: 1/day.

- **CAR-T cell reduction rate (ρ)**: differentiation rate of effector CAR-T cells into memory CAR-T cells. Unit: 1/day.
- **CAR-T inhibition (α)**: probability of effector CAR-T cell inhibition per day caused by the tumor. For example, the action of PD-L1/PD-1 signaling. Unit: 1/(cell.day).
- **Memory conversion rate (ϵ)**: proportion of differentiated effector CAR-T cells which become memory CAR-T cells. Unit: 1/day.
- **Memory death rate (μ)**: inverse of the half-life time of memory CAR-T cells. Unit: 1/day.
- **Conversion coefficient (θ)**: proportion of memory CAR-T cells which are converted into effector CAR-T cells per day driven by antigen contact on tumor cells. Unit: 1/(cell.day).
- **Numerical parameter, Δt value**: Simulation time step size. Typical values range from 10^{-2} up to 10^{-6} (day). In general, smaller time steps produce more accurate results but require more computational processing. A $\Delta t = 10^{-2}$ yields convergent solutions for Ruella et al (2017) experiments.

The "Reset advanced parameters" button enable the user to reset the original values of the advanced parameters.

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Development team

Emanuelle A. Paixão, Gustavo T. Naozuka, Andrea M. P. Valli,
Luciana R. C. Barros, and Regina C. Almeida.