IL-13 induced JAK2/STAT5 signaling model in MedB-1 cells

Reference: Raia et al. Dynamic Mathematical Modeling of IL13-induced Signaling in Hodgkin and Primary Mediastinal B-cell Lymphoma Allows Prediction of Therapeutic Targets. Cancer Research 71, pp. 693-704, 2011.

Folder: `/Examples/Raia_CancerResearch2011`.

Facts: The model contains 205 data points, 39 free parameters and 4 experimental conditions.

Summary

This page provides a guided tour through the Data2Dynamics software using the dynamical model of JAK2/STAT5 signal transduction published in Raia et al., Cancer Research 2011.

Like most projects in Data2Dynamics this example is based on

- a **model definition** file specifying inputs, reactions, observations, and error model
- **data definition** files depicting experimental setups different from those defined previously
- **data** files containing the measured data
- a **setup** file to initialize the software, load model as well as data and create the associated C files

These definition, data and setup files are located in `/Examples/Raia_CancerResearch2011`, and their respective subfolders.

The Data2Dynamics software works hierarchical in the sense that specifications in the model definition are considered as defaults but may be overwritten for particular data sets by specifications in the respective data definition file. If on the other hand a data definition file is empty except the mandatory keywords in capitals, the defaults from the model definition are used. The latter case is the situation in this example.

The following tasks are routinely performed after model and data have been loaded:

- **Plotting** to visualize the data and/or the dynamics generated from the current set of parameters
- **Fitting** to adjust the parameters to describe the data as good as possible
- **Uncertainty analysis** to assess parameter identifiability and calculate confidence intervals

Model definition

The file `il13_jak2_stat5.def` in the `/models` subfolder specifies the ODE model, i.e. names and units of the dynamic variables and their dynamic interactions by providing reaction schemes. A model definition file is divided in several sections that are described in detail at theSetting up models page. Most prominently the differential equation system is defined in the

REACTIONS section as conversion scheme.

```
...
REACTIONS
Rec             ->  IL13_Rec        CUSTOM "il13_level * Kon_IL13Rec * 2.265 * Rec"
Rec             ->  Rec_i           CUSTOM "Rec_intern * Rec"
Rec_i           ->  Rec             CUSTOM "Rec_recycle * Rec_i"
...
```

Each line represents a biochemical reaction. Reactants and products are specified on the left and right of the reaction arrow `->` and the reaction rate is specified next. Here, the membrane associated receptor `Rec` either forms a complex `IL13_Rec` with the ligand `IL13` (first line) or is internalized (second line). The internalized receptor `Rec_i` is again recruited to the membrane (third line). Each reaction rate is added to the right-hand-side of the ODEs for the products and subtracted for the reactants. The Data2Dynamics software parses the specified reaction rates and considers each term that is neither a dynamic variable nor an input as parameter, which can be estimated later from the data. As `il13_level` is set to the input dose for each experiment the introduced parameters from the reactions above are `Kon_IL13Rec`, `Rec_intern` and `Rec_recycle`.

Data definition

Later in the same file `il13_jak2_stat5.def` the observation functions are specified to make the default connection between dynamic variables and experimental data. In particular the observation functions are defined in the OBSERVABLES section and the associated error model after the keyword ERRORS.

```
...
OBSERVABLES
RecSurf_obs         C   au  conc.   0   0   "Rec + IL13_Rec + p_IL13_Rec"
IL13_cell_obs       C   au  conc.   0   0   "scale_IL13_cell_obs * IL13_cell"
...
ERRORS
RecSurf_obs         "RecSurf_obs1 + RecSurf_obs2*RecSurf_obs"
IL13_cell_obs       "IL13_cell_obs1 + IL13_cell_obs2*IL13_cell_obs"
...
```

In these exemplary lines the total amount of receptor on the surface is defined as the observable `RecSurf_obs`. As the IL13 concentration can only be observed on a relative scale, the observational parameter `scale_IL13_cell_obs` is introduced. For each observable it is required to specify an error model, in this case a relative error `RecSurf_obs2*RecSurf_obs` with an offset `RecSurf_obs1` is implemented.

Data file

The file `MedB1_real_data.def` in the `/data` subfolder is empty except for keywords hence the observables defined in the model definition above are not altered.

The associated file `MedB1_real_data.xls` contains the experimental data.

```
time    il13_level  pIL4Ra_obs  pJAK2_obs   IL13_cell_obs   RecSurf_obs     ...
0.0     0           NaN         NaN         0.616           1.289752779
5.0     0           NaN         NaN         NaN             NaN
10.0    0           NaN         NaN         NaN             1.57854438
...
```

Note that the headers of this file must match either the independent variable (here: `time`), or a model parameter (e.g. `il13_level`), or the name of an observable (e.g. `RecSurf_obs`). Columns with unrecognized headers are ignored.

Setup

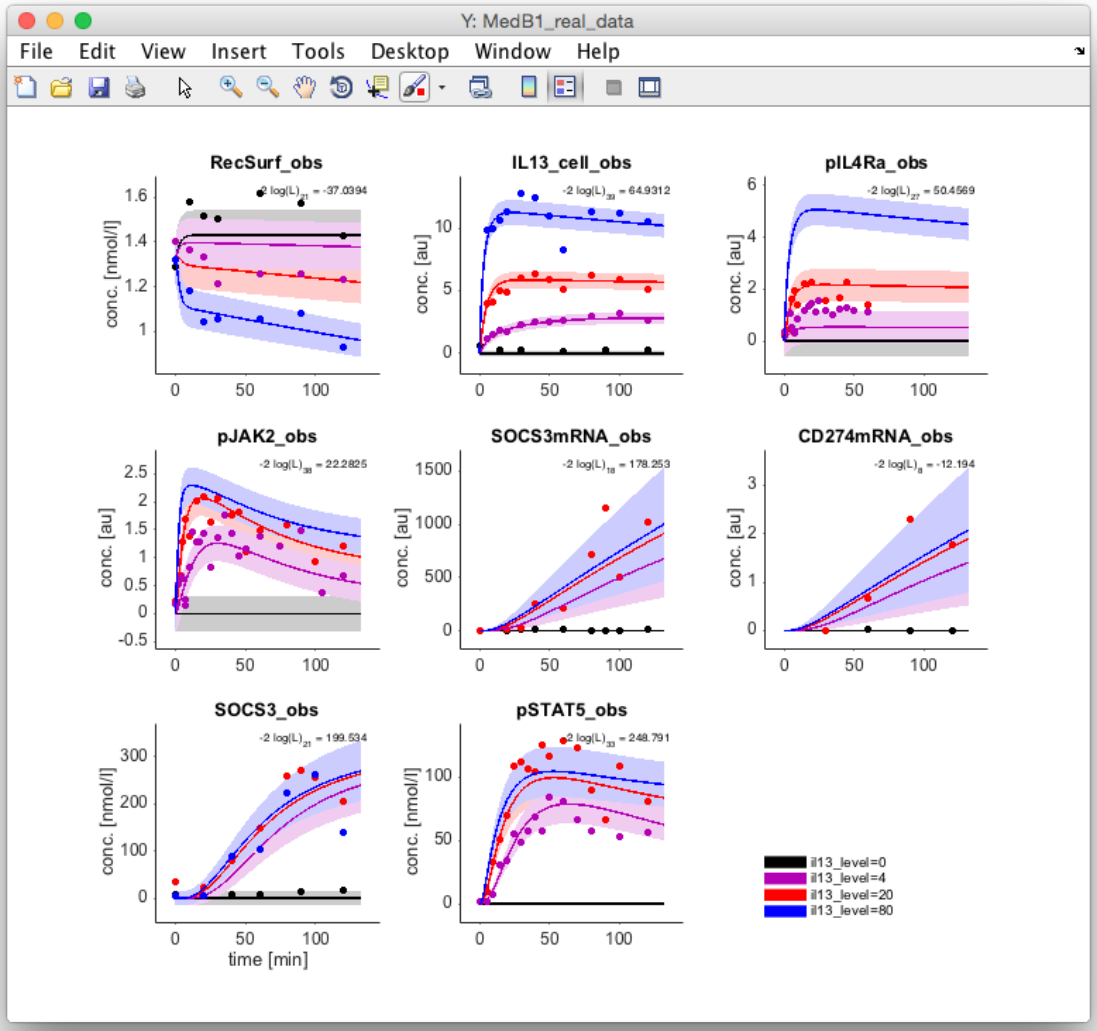
Executing the script `Setup.m` loads model and data in MATLAB and automatically generates the C-files. After the setup script is finished once, the full functionality of the Data2Dynamics framework can be applied for the analyses. All information about model and data are now available in the MATLAB environment as a global variable `ar`. Below some basic functions are explained briefly, for further information see Extended Functionalities and Function Reference.

Calling `arPrint` shows the list of all model parameters in the MATLAB command window. The output contains: the parameter index (#), a classifier (D=parameter involved in dynamics, E=error model parameter), the parameter name, its current values (values), upper bound (ub) and lower bound (lb), if the values are given on a log10-scale and the respective non-log10 parameter value (10^value), if the parameter is free for fitting (fitted, 0=fix, 1=fitted, 2=constant), and the prior distribution for this parameter (prior).

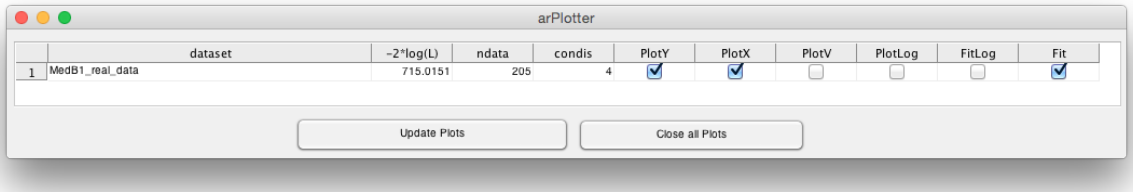
```
Parameters: # = free, C = constant, D = dynamic, I = initial value, E = error model

            name                        lb       value       ub          10^value
#   1|D   | CD274mRNA_production     |       -5       -1.7         +3 | 1     +0.021 |
#   2|D   | DecoyR_binding           |       -5       -2.4         +3 | 1    +0.0039 |
#   3|D   | JAK2_p_inhibition        |       -5       -1.1         +3 | 1     +0.078 |
...
```

Plotting

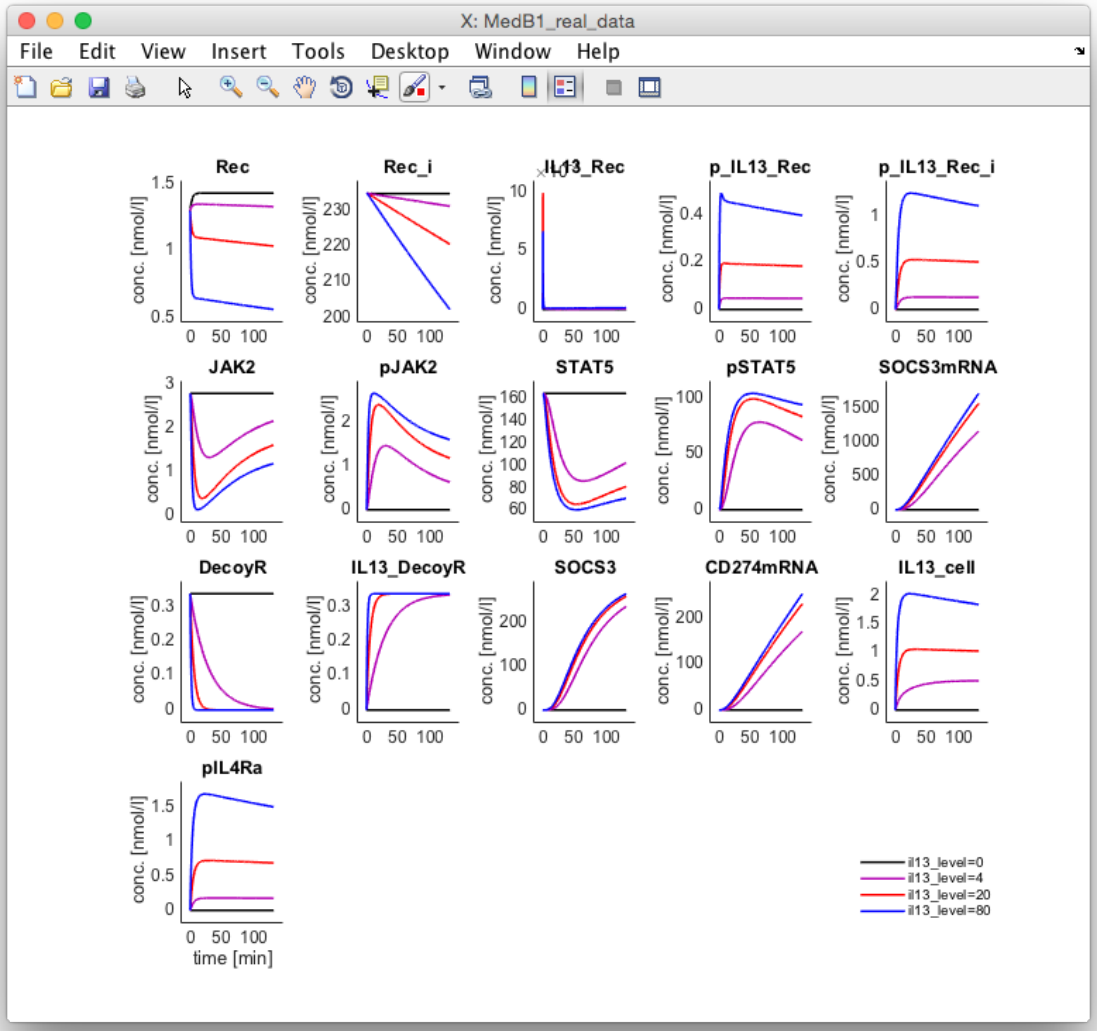
The user has several possibilities to specify which kind of plots should be shown by the default plotting command `arPlot`. After the setup, one figure for each data set is shown (one in total for this example).



One way to also visualize the underlying ODE solutions is to invoke `arPlotter`, which opens a small graphical user interface that allows selecting data sets and available quantities to display by the `arPlot` command.



The list contains the data set name (dataset), its current log-likelihood value (-2*log(L)), the number of data points in this data set, the number of experimental conditions, if the data plot is shown (PlotY), if the internal model dynamics are shown (PlotX), if the model reaction fluxes are shown (PlotV), if the data set if plotted on a log10 y-axis (PlotLog), if it is fitted on a log10 y-scale (FitLog) and if it is fitted at all (Fit). After activating the checkbox `PlotX` and hitting the button `Update Plots` a new figure is opened showing the dynamics of the internal variables.



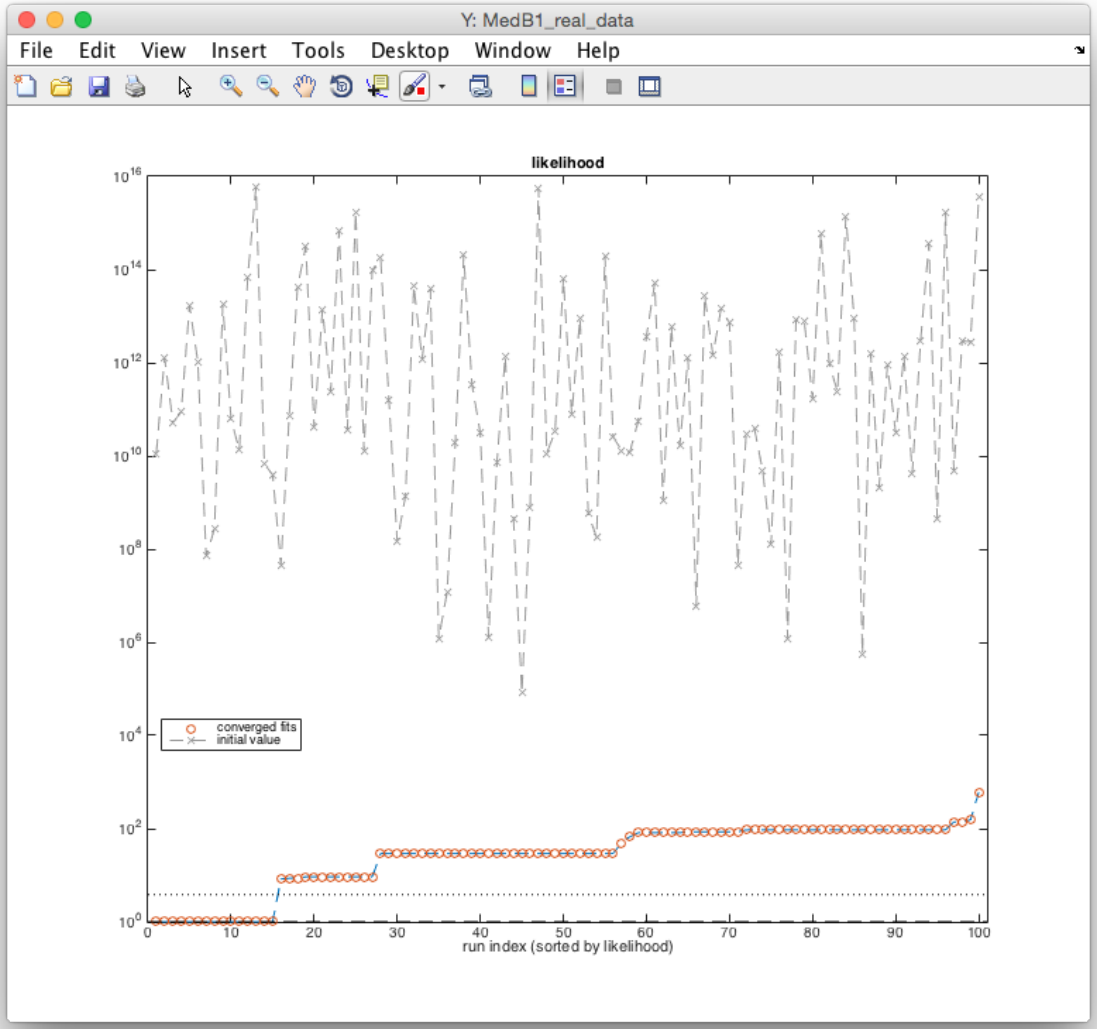
All commands executed in the Data2Dynamics framework via a graphical user interface can of course also be performed at the MATLAB command line, which enables the creation of analysis workflows.

Fitting

`arFit` starts the selected numerical optimization algorithm to calibrate the model to the experimental data. Several optimizers `ar.config.optimizers` can be selected specified by adjusting the index `ar.config.optimizer`. If the option flag in the global variable `ar.config.showFitting = true` model calibration is shown for each iteration by internally calling `arPlot`. In this example, optimal parameters values are already loaded from a workspace in the setup file, see line `arLoadPars`. If this line is commented the parameter values are set to default values of 0.1 as can be verified by the `arPrint` command. `arFit` then nicely demonstrates the efficiency of the implemented optimization technique by showing the iterative improvements.

To check whether the current local optimum is also globally optimal a sequence of `n` fits for random initial guesses is executed using the command `arFitLHS(n)`. For deterministic

optimization the initial parameter guesses are generated by random sampling. `arFitLHS` replace previous values if a better fit is found. The resulting likelihoods of the `n` fits are displayed in a sorted manner by `arPlotFits`.

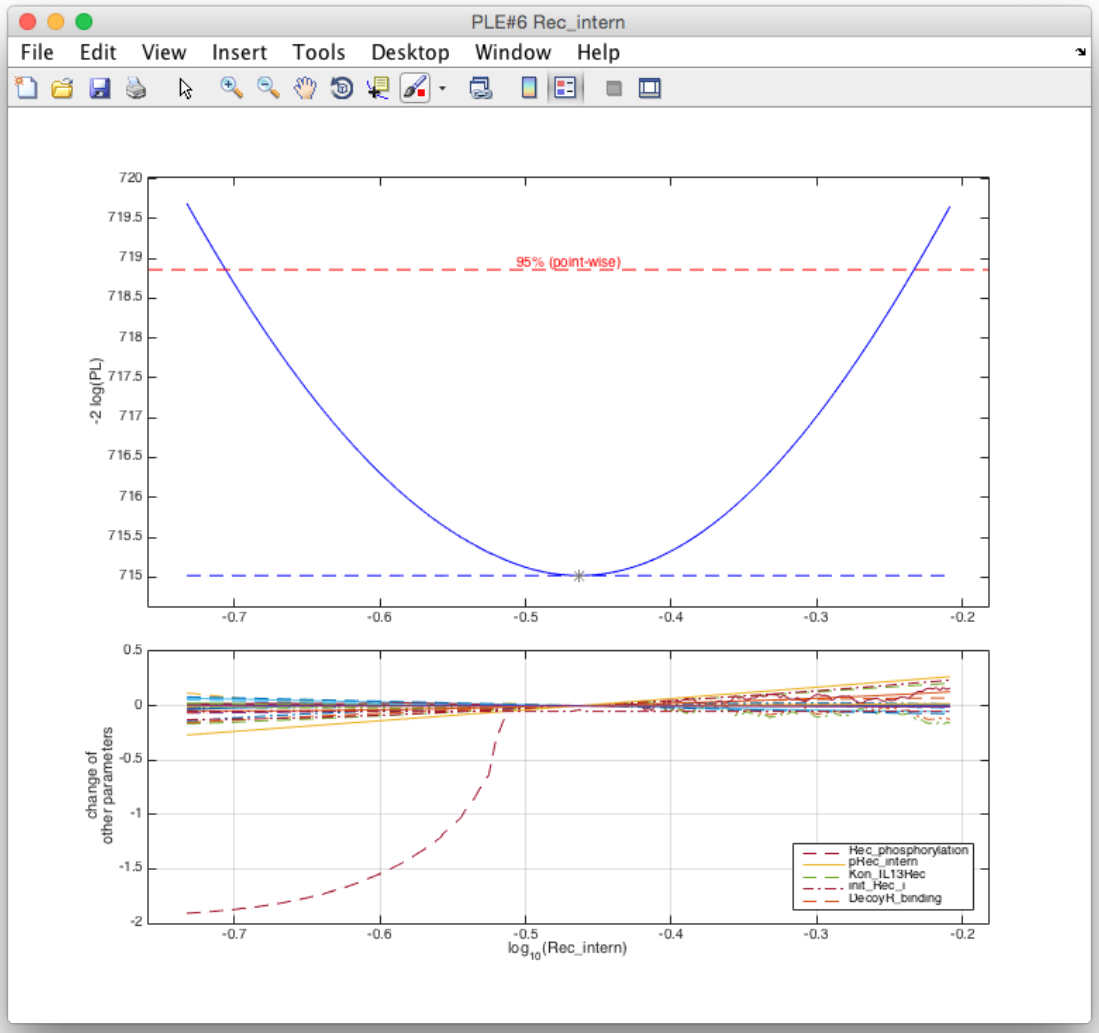


This figure shows the result for `n=100` independent fits from random initial starting points and indicates 1) the improvement of the initial likelihood value by several orders of magnitude (gray crosses vs. red circles), and 2) the plateaus after optimization demonstrating that local minima are reliably detected. The more often the same local minima are found, the better the chance that all existing local minima including the global one were found. See Raue et al., PLOS ONE 2013 for further explanation.

Profile Likelihood

A convenient way to assess whether a parameter of a non-linear model has a finite confidence region (i.e. is identifiable, see Raue et al., Bioinformatics 2009 for further explanation) is by calculating the profile likelihood. This concept generalizes the concept of standard errors and Fisher-Information for the non-linear setting. After initializing the profile likelihood calculation by `arPLEInit`, the profiles for all parameters are calculated

automatically by `ple`. The calculation for a specific parameter, e.g. for the internalization rate `Rec_intern` might be performed by `ple('Rec_intern', m)`. The second argument `m` can be used to specify the maximum number of steps (default: 50) along the profile. For this example, 200 points in lower an upper direction were calculated via `ple('Rec_intern', 200)`.

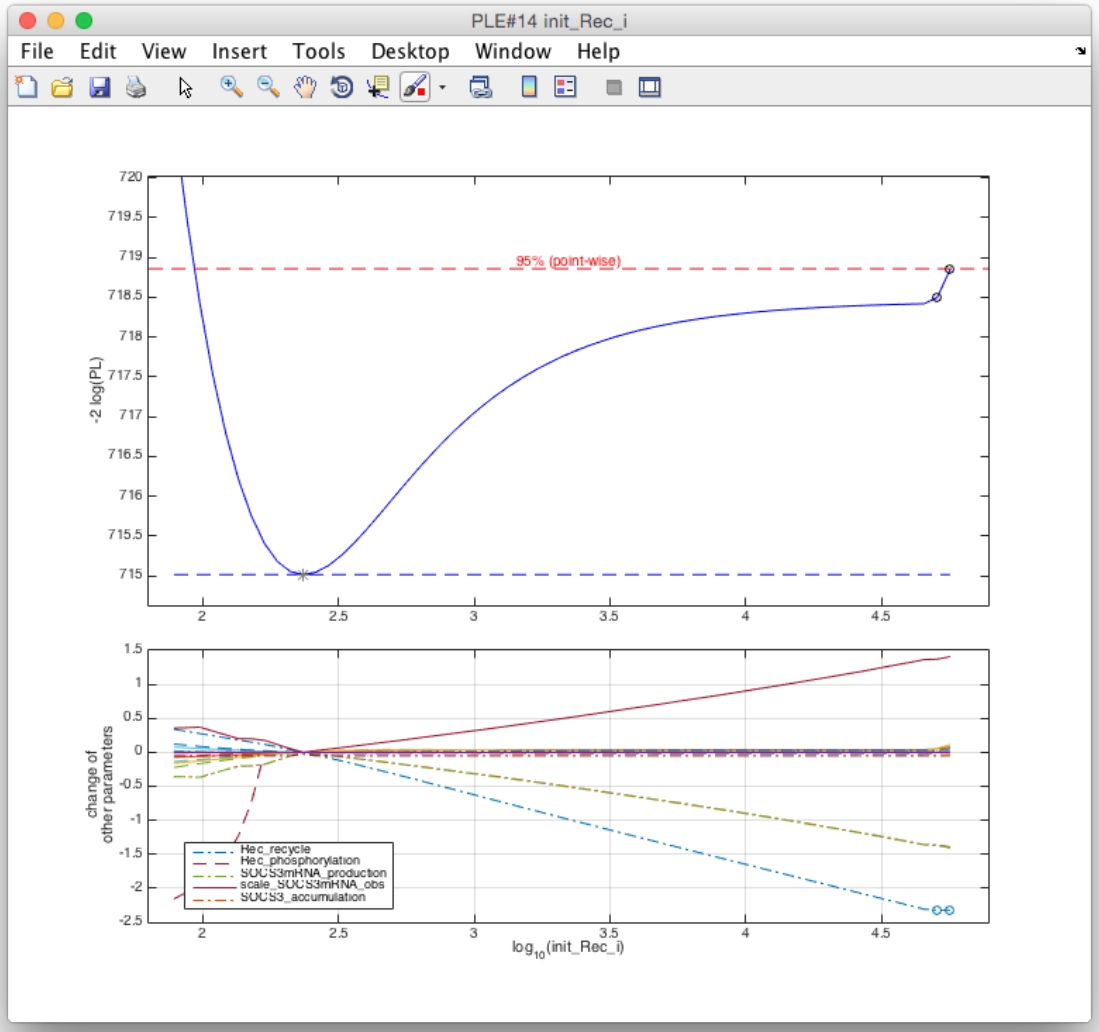


In the upper panel the profile likelihood is plotted. The parameter is identifiable because the profile (blue line) crosses the statistical threshold (red) at both sides of the optimum. The lower panel shows along the profile, i.e. for specific values of `Rec_intern`, how all other parameters have to be adjusted to describe the data. These dependencies are of particular interest for non-identifiabilities as demonstrated in the following exemplary profile.

The second parameter examined for identifiability is the initial concentration of the IL13 receptor `init_Rec_i`. Here, the default settings only calculate a part of the profile of interest. The following commands quickly provide the whole profile likelihood.

```
k = arPrint('init_Rec_i');          % Find index of parameter init_Rec_i
global ar; global pleGlobals;       % Get access to global variables
ar.ub(k) = 5;                       % Set upper boundary of init_Rec_i to 10^5 for MLE
pleGlobals.ub(k) = 5;               % Set upper boundary of init_Rec_i to 10^5 for PLE
ple('init_Rec_i',[],[],.1,.1)       % Calculate profile with fixed stepsize of 0.1
```

The resulting figure shows the profile likelihood of `init_Rec_i`.



The following conclusions can be drawn from this plot:

- The parameter is significantly larger than 2 (on the log10-scale)
- The profile becomes flat for values > 4
- The increase at around 4.7 induced is due to another parameter (`Rec_recycle`) hitting its lower boundary as indicated by the circles in the lower panel. To check this hypothesis, the profile is recalculated with a less stringent lower boundary for the receptor recycling parameter.

```
j = arPrint('Rec_recycle');      % Find index of Rec_recycle
ar.lb(j) = -8;                   % Set lower boundary of Rec_recycle to 10^-8 for MLE
pleGlobals.lb(j) = -8;           % Set lower boundary of Rec_recycle to 10^-8 for PLE
ple(k,[],[],.1,.1)               % Re-calculate profile of init_Rec_i
```

The resulting profile likelihood is flat in upper direction, which demonstrates that the receptor concentration `init_Rec_i` can not be significantly restricted by the data towards large numbers. Hence `init_Rec_i` is practically non-identifiable.