

WimScratch

Analysis results in detail

Images uploaded to WimScratch are considered to be an individual experiment of a Time Series, as we will call it in this document. If you want to upload more than one of them, just put each Time Series (experiment images) inside a different folder and you will get individual results for each of them.

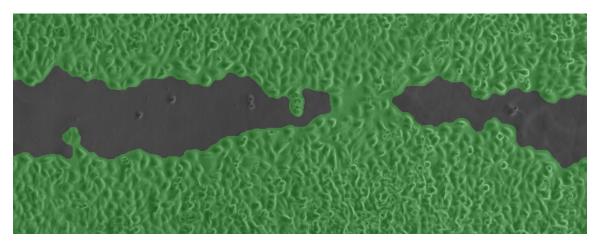
For every set of images that you upload to WimScratch, you will receive the following data files, bundled inside a zip file:

- A Results Control Image for each uploaded image, consisting of your original images with an overlay of the detected cells in it.
- A CSV format Results Data Sheet with the wound healing measured data for each uploaded image.
- A CSV format Time Series Data Summary of the individual sheets for each uploaded Time Series.
- A Time Series Chart representing the progress in time of the wound healing process for each uploaded Time Series.
- A CSV format Results Data Summary of the whole order.

Let's check them in detail.

1. Results Control Image

Every input image will produce a control image (saved in jpg format) to assess the accuracy of the analysis output. It consists of the input image with the detected migrating cells with a transparent green mask on them, as shown in the image below:





2. Results Data Sheet

Every input image will also generate an output CSV file (comma separated value format, similar to an Excel sheet) with the following measurements:

• Scratch Area [%]: the percentage of the image still covered by the scratch.

3. Time Series Data Summary

Every uploaded Time Series will also generate an output CSV summary file with some Standard Metrics of your wound healing process. You will get further Time Metrics if you include a timestamp in the filenames of your Time Series (as described in Step 2 of the WimScratch Application Note that you can download here).

Individual Standard Metrics:

- Image Nr / Time [Time units]: an ID to identify each image in the Time Series Chart (an artificial "timestamp"). If you include a timestamp in your filename, it will be the time inside the filename (in generic Time units that you can convert to the Time unit of your experiment, generally hours).
- Scratch Area [%]: the percentage of the image still covered by the scratch.
- Cell-covered Area [%]: the percentage of the image covered by cells.

Individual Time Metrics:

• **Speed [%/Time Units]:** the wound closing speed between this image and the next in the Time Series, measured as percentage of wound area covered divided by the time difference between the images.

It is calculated using the following formula (which means that there is no value for the last time-point as there is no next point to calculate it with):

Speed =
$$\frac{\text{CoveredArea (T2) - CoveredArea (T1)}}{\text{T2 - T1}}$$

• Acceleration [%/(Time Units)^2]: the wound closing acceleration between this image and the next in the Time Series, measured as difference of speed between this image and the next, divided by the time difference between them.

It is calculated using the following formula (which means that there is no value for the last two time-points as there is speed data to calculate it with):

Acceleration =
$$\frac{\text{Speed (T2) - Speed (T1)}}{\text{T2 - T1}}$$



General Time Metrics:

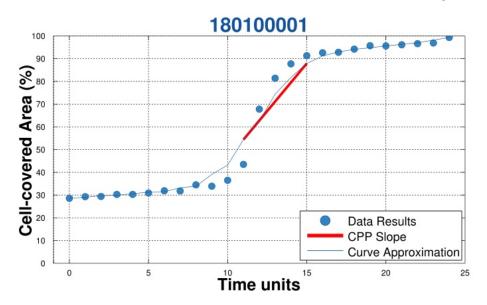
- Average Speed [%/Time Units]: the average wound closing speed.
- Start Point [Time units]: the time point of first image of the Time Series.
- End Point Cell-covered Area [%]: the percentage of image cell-covered area in the first image of the Time Series.
- End Point [Time units]: the time point of last image of the Time Series.
- End Point Cell-covered Area [%]: the percentage of image cell-covered area in the last image of the Time Series.

If your Time Series includes more than five images and the wound closing process is smooth enough in its transition from open to close, a Central Piece Proxi (CPP) can be calculated. The CPP presents the speed for the central piece of the experiment and indicates the initial delay in closure as well as the late stoppage in total coverage.

If a CPP can be calculated for your Time Series, the five parameters described above will also be calculated for it and included in your Summary.

4. Time Series Chart

In unison with the adage "A picture is worth a thousand words", the WimScratch solution provides a visualization of the complex data that conveys the experiment properties with just a single image, the Time Series Chart. The analysis data of each Time Series will be represented in a chart showing cell-covered area over time. In addition to the discrete data points, a curve approximation and the CPP slope is presented in a graphic file.



This chart represents linear cell migration of a 25 point analysis of a Scratch Assay.



5. Results Data Summary

Every uploaded order also produces a CSV file that summarizes the measurements contained in all the individual results data sheets of your order. This file will have the name of the order number followed by "_Summary". If you only upload one Time Series, it will be the same file as the Time Series CSV data summary.