

# READ ME

## 1 Image Analysis Tool

This application window is part of an **Image Processing Workflow** application. It allows users to configure various parameters related to image processing, particularly focusing on Gaussian filtering and image analysis. Below is a detailed breakdown of the components in the window:

### Input Fields

- **Gaussian Window:** A numeric input field where the user specifies the size of the Gaussian window for image filtering. For example, a default value of **121** can be used.
- **Gaussian Sigma:** A numeric input that defines the standard deviation for the Gaussian filter, affecting the smoothness of the filtering effect. The default value is **40**.
- **Dtime:** This input field represents a time-related parameter, such as the time-step used in processing or image acquisition, with a default value of **1E-5**.
- **Grid Size - Image Size:** A drop-down menu allows the user to select a grid size for the images, with options ranging from **8 to 64**, defining the resolution or sampling grid size for the image processing task.

### Checkbox Option

- **Enable Advanced Fitting:** A checkbox labeled **Enable Advanced Fitting (takes longer time)** allows the user to enable advanced image fitting techniques, which may provide more precise results but take longer to compute.

### File and Directory Selection

- **Select .tif Images:** A button labeled **Select .tif Images** allows users to select image files in the **.tif** format, commonly used for high-quality scientific images.
- **Select Output Directory:** This button allows the user to choose where the processed output files will be saved. It is currently marked as **No output directory selected**.

### Output Excel File Name

There is a text field where the user can specify the name of the Excel file in which the output data will be saved. The default value is **output\_data.xlsx**.

### Log/Status Area

A large text area is present below the file name input field. This space can be used for logging the progress of the image processing workflow, displaying statuses, or indicating errors.

## Run Workflow Button

At the bottom of the window, there is a button labeled **Run Workflow**, which initiates the image processing workflow using the selected input parameters and files.

## Summary

This application window is designed for scientific image processing tasks. It allows for custom Gaussian filtering, grid size selection, and advanced fitting techniques. Users can select input **.tif** image files and configure output settings, with results saved in an Excel file.

## 2 Data Analysis Tool

This application is a graphical user interface (GUI) designed for running data analysis based on two model options: a **Simple Model** and a **Model with Background (BG)**. Users can load data, set bounds, exclude data points, customize output preferences, and save their analysis results.

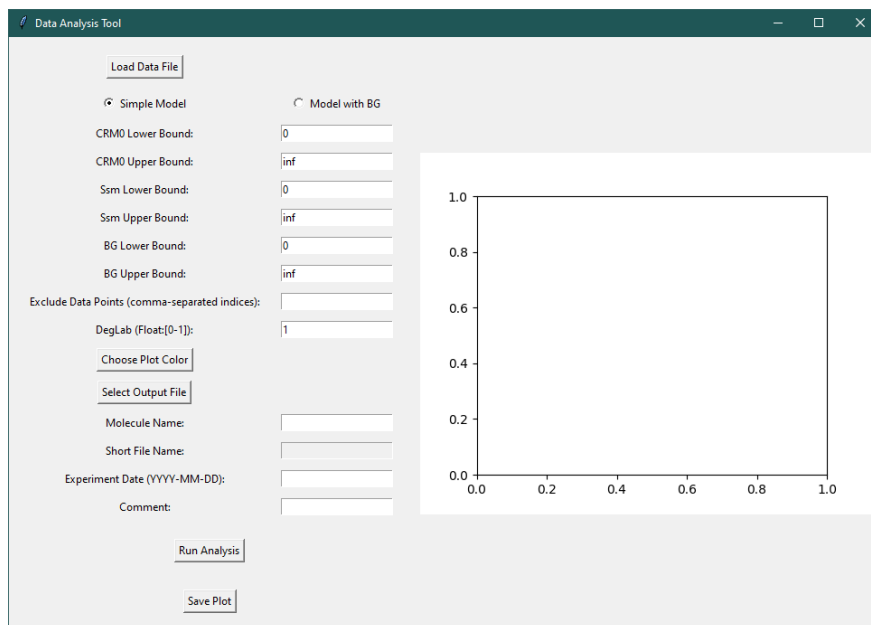


Figure 1: Start up screen of the application

## Features

- **Model Selection:**
  - **Simple Model**<sup>1</sup>: A basic analysis model without background contribution.
  - **Model with BG**: Includes an analysis with background contribution.
- **Bound Settings**: Set custom lower and upper bounds for CRMO, Ssm, and BG.
- **Exclude Data Points**: Manually exclude data points by specifying their indices.
- **DegLab Parameter**: Adjust the degree of labeling (Float[0-1]).

<sup>1</sup>Combining Fluorescence Fluctuations and Photobleaching to Quantify Surface Density: <https://pubs.acs.org/doi/10.1021/acs.analchem.1c05513>

- **Select Output file:** Choose an existing output Excel file or create a new one.
- **Metadata Input:** Add information such as Molecule Name, Short File Name (extracted from the Excel file), Experiment Date, and Comments.
- **Run and Save Analysis:** Execute the analysis and save the plot as an image file.

## How to Use

### 1. Load Data:

- Click the *Load Data File* button to select and upload your data file.
- **Input Data File:** The tool expects an Excel file to be loaded. If the Excel file doesn't contain 'CRMmoy', 'CRmoy' and 'CRMsem' as headers, it will raise an error.

### 2. Model Selection:

- Choose between the *Simple Model* or the *Model with BG* by selecting the corresponding radio button.

### 3. Set Bounds:

- Input values for the bounds (CRMO, Ssm, and BG), or leave the default values (0 for lower bound, inf for upper bound) for no restrictions.

### 4. Exclude Data Points:

- If you wish to exclude certain data points from the analysis, provide their indices in the input box, separated by commas (e.g., 1,3,5). The first row is equal to 0!

### 5. DegLab Parameter:

- Set the *DegLab* value, which should be a float between 0 and 1. Example: If you are using a mixture of 1:1 Molecule with no label and molecule with label, then the  $\text{DegLab} = 0.5$ .

### 6. Select Output File:

- Click the *Select Output File* button to choose a location and filename where the analysis results will be saved. This step needs to be performed only once every time the application is opened.

### 7. Enter Metadata:

- Input optional information such as Molecule Name, Short File Name, Experiment Date, and any additional comments.

### 8. Run Analysis:

- Click the *Run Analysis* button to execute the analysis and display the plot.

### 9. Save Plot:

- Click *Save Plot* to save the generated plot to the file specified in the *Select Output File* step.