Guideline for FLIm Data Visualization Tool

1. Introduction

The purpose of this guideline is to facilitate users with generating the FLIm map using the FLIm data visualization tool. In this guideline, we introduce the procedure for producing the FLIm map. We also exhibit the images resulted from different combinations of parameters in order for users to generate the desired results by properly selecting the parameters.

The tool is a graphical user interface (GUI) which is built using ***GUIDE*** in ***MATLAB*** of version ***R2019a*** under *macOS*. While the majority of the script is written in *MATLAB* under *macOS*, the figures and resultant images in this guideline were produced in *MATLAB* under *Windows 10*.

2. Installation

To run the GUI, you will need to install ***MATLAB*** and also to install ***Image Processing Toolbox*** in order to successfully execute the plotting action.

Following setting the environment, all of the scripts below are necessary to be included in exactly the same path/folder when running this tool:

(a) FLIm_Data_Visualization.fig

(b) FLIm_Data_Visualization.m

(c) replotImage.m

(d) processImgInterpGUI.m

(e) IDW.m

(f) NaturalNeighbor.m

(g) OriginalCircle.m

(h) SNRmap.m

3. GUI

To run the GUI, simply open **FLIm_Data_Visualization.m** in *MATLAB* and click the **Run** button in the toolbar at the top of the window as Fig. 1 shows. Following clicking the button, the GUI will be displayed like Fig. 2. Note that the appearance of the GUI might vary due to the different resolution of the screen you use; however, it will not have any effect on the operation of the functions of the tool.

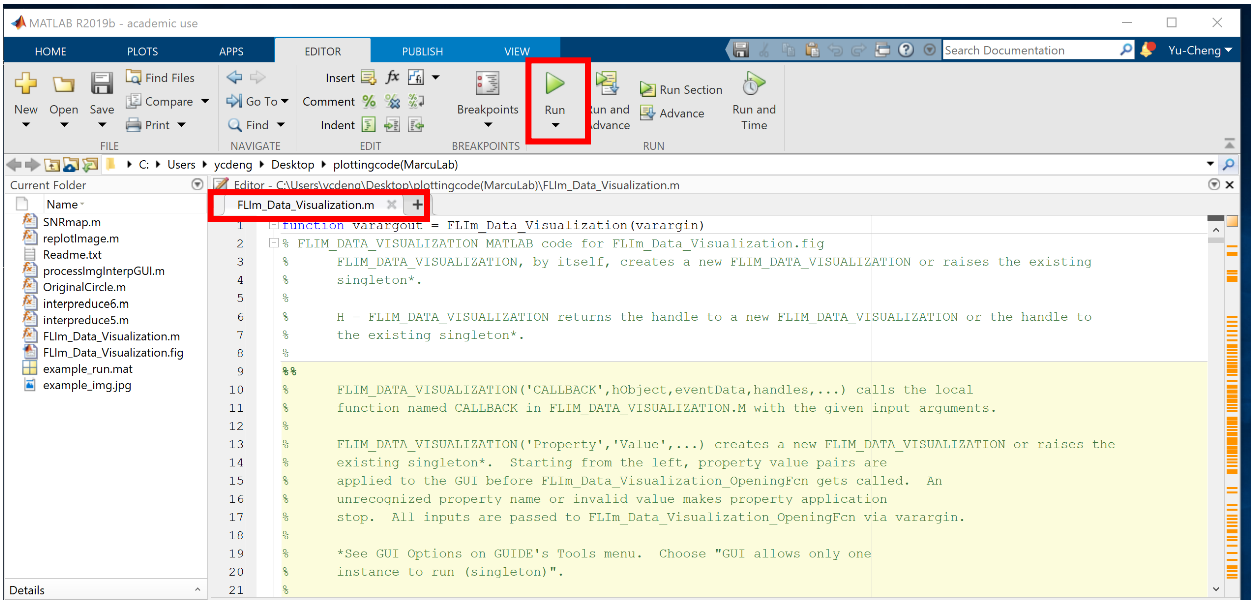


Figure 1. Click the green triangular button, **Run**, to run **FLIm_Data_Visualization.m** to open the GUI.

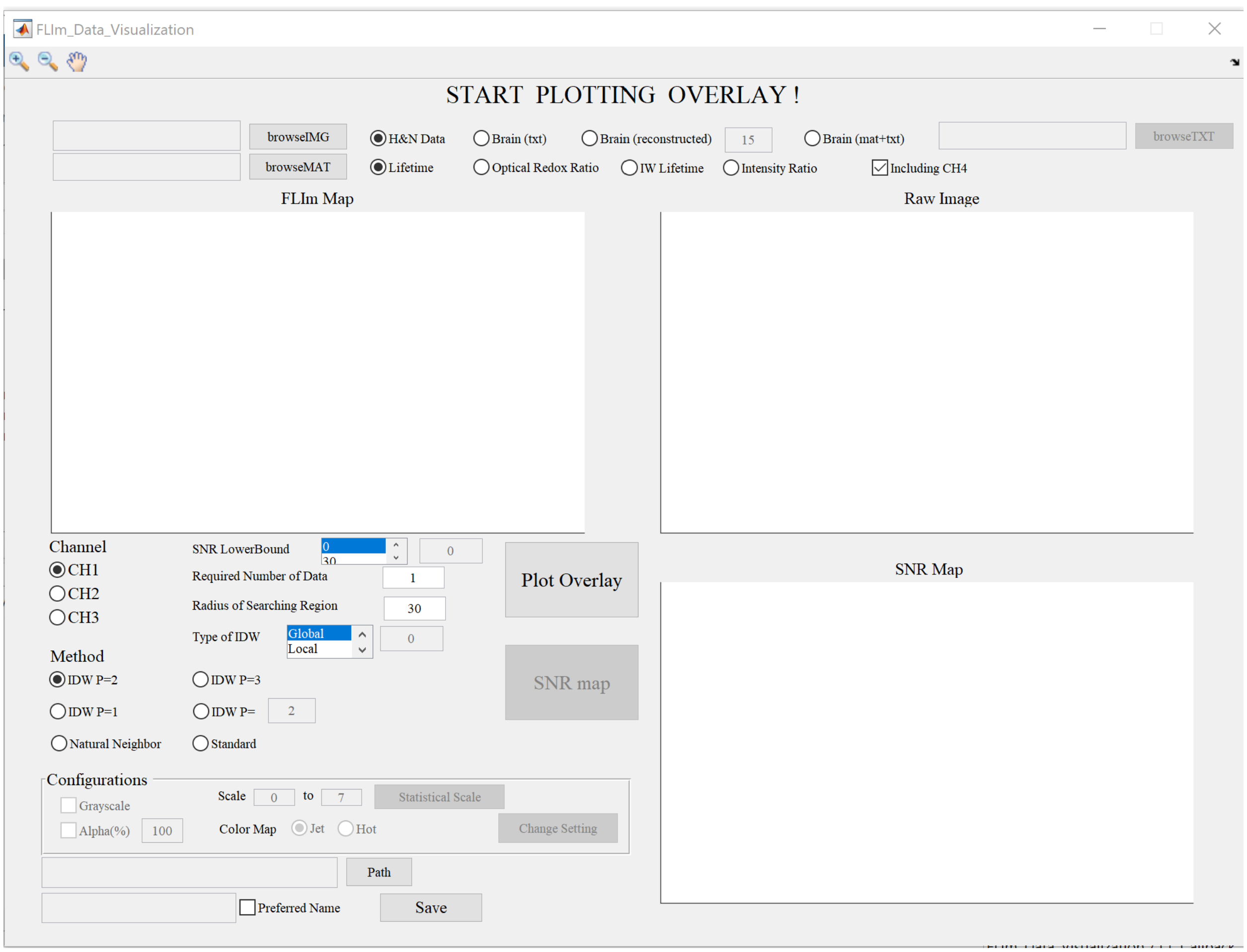


Figure 2. The appearance of the GUI.

In this section, we will introduce the steps for plotting FLIm map successively and respectively explain the role of each box (functionality) in Fig. 3.

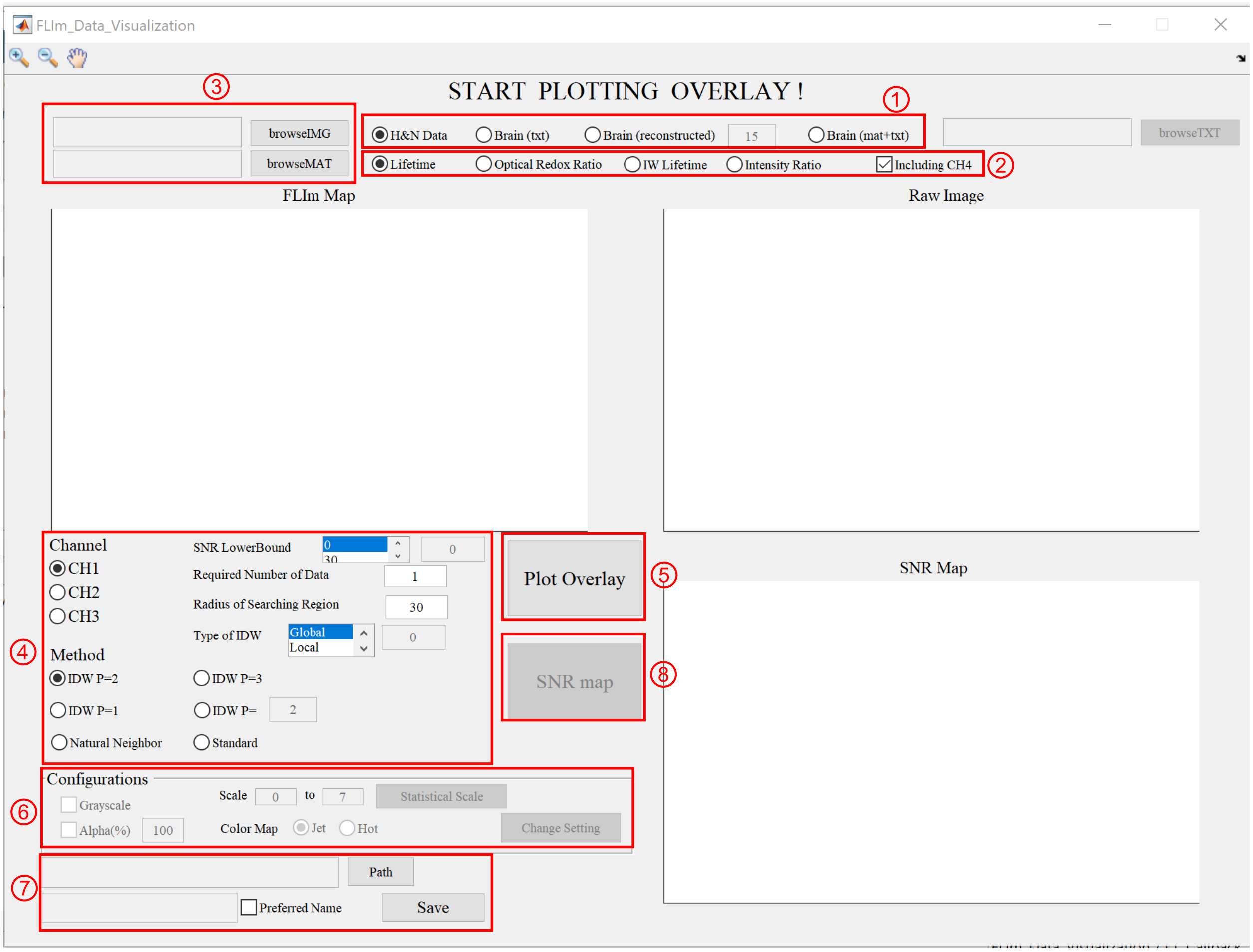


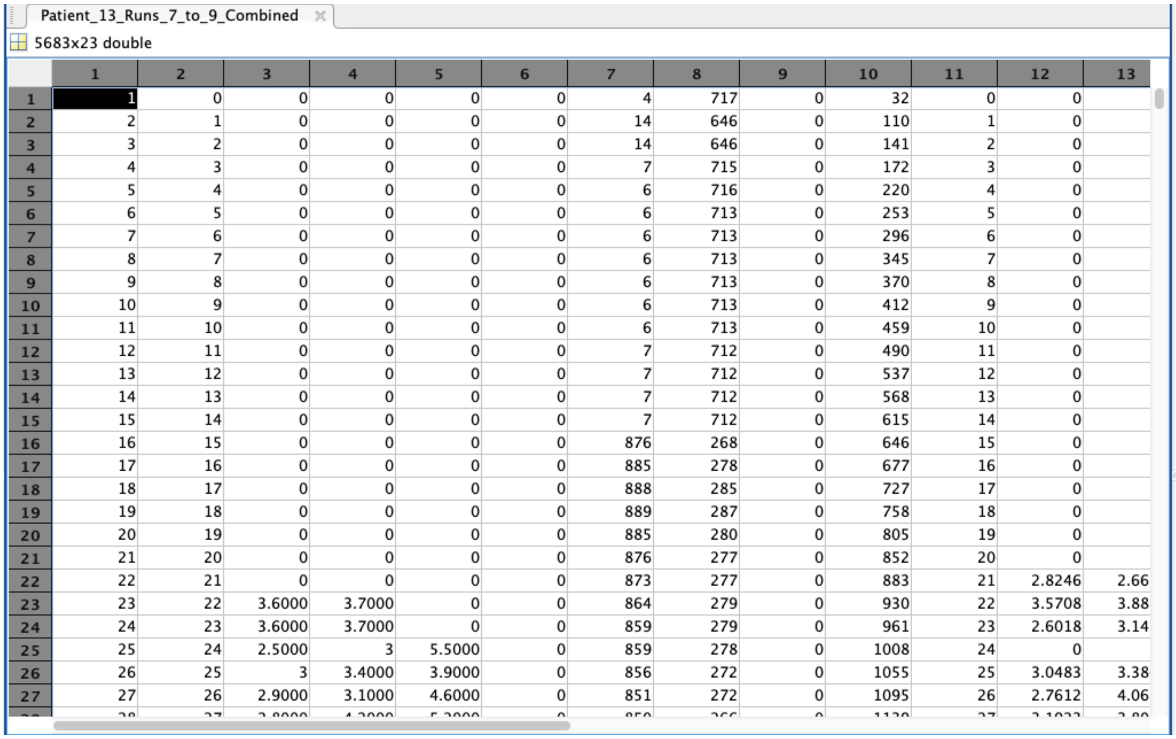
Figure 3. Each box in the GUI has its functionalities.

(1) Choice of dataset

There are four radio buttons in the **box 1** in Fig. 3. In other words, the GUI can process four kinds of formats of dataset majorly utilized in our lab. In this part, we will respectively introduce these four formats for users to choose accurately to avoid errors.

(a) Radio Button: H&N Data

The first button is to process the head and neck (H&N) dataset which typically is the **mat** file (.mat) having **23 columns** as Fig. 4 shows. The meaning of each column can be found in Table. 1. To visualize the map, we only employ column 1, 7, 8, 9, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23.


Patient_13_Runs_7_to_9_Combined

5683x23 double

	1	2	3	4	5	6	7	8	9	10	11	12	13
1	1	0	0	0	0	0	4	717	0	32	0	0	
2	2	1	0	0	0	0	14	646	0	110	1	0	
3	3	2	0	0	0	0	14	646	0	141	2	0	
4	4	3	0	0	0	0	7	715	0	172	3	0	
5	5	4	0	0	0	0	6	716	0	220	4	0	
6	6	5	0	0	0	0	6	713	0	253	5	0	
7	7	6	0	0	0	0	6	713	0	296	6	0	
8	8	7	0	0	0	0	6	713	0	345	7	0	
9	9	8	0	0	0	0	6	713	0	370	8	0	
10	10	9	0	0	0	0	6	713	0	412	9	0	
11	11	10	0	0	0	0	6	713	0	459	10	0	
12	12	11	0	0	0	0	7	712	0	490	11	0	
13	13	12	0	0	0	0	7	712	0	537	12	0	
14	14	13	0	0	0	0	7	712	0	568	13	0	
15	15	14	0	0	0	0	7	712	0	615	14	0	
16	16	15	0	0	0	0	876	268	0	646	15	0	
17	17	16	0	0	0	0	885	278	0	677	16	0	
18	18	17	0	0	0	0	888	285	0	727	17	0	
19	19	18	0	0	0	0	889	287	0	758	18	0	
20	20	19	0	0	0	0	885	280	0	805	19	0	
21	21	20	0	0	0	0	876	277	0	852	20	0	
22	22	21	0	0	0	0	873	277	0	883	21	2.8246	2.66
23	23	22	3.6000	3.7000	0	0	864	279	0	930	22	3.5708	3.88
24	24	23	3.6000	3.7000	0	0	859	279	0	961	23	2.6018	3.14
25	25	24	2.5000	3	5.5000	0	859	278	0	1008	24	0	
26	26	25	3	3.4000	3.9000	0	856	272	0	1055	25	3.0483	3.38
27	27	26	2.9000	3.1000	4.6000	0	851	272	0	1095	26	2.7612	4.06

Figure 4. The H&N dataset format (The example is Patient_13_Runs_7_to_9_Combined.mat).

Column 1	Frame	Column 9	Z-Axis	Column 17	Intensity 2 Deconv
Column 2	~~ID Txt~~	Column 10	~~Unknown~~	Column 18	Intensity 3 Deconv
Column 3	~~Lifetime 1~~	Column 11	~~ID Deconv~~	Column 19	Intensity 4 Deconv
Column 4	~~Lifetime 2~~	Column 12	Lifetime 1 Deconv	Column 20	SNR 1 Deconv
Column 5	~~Lifetime 3~~	Column 13	Lifetime 2 Deconv	Column 21	SNR 2 Deconv
Column 6	~~Lifetime 4~~	Column 14	Lifetime 3 Deconv	Column 22	SNR 3 Deconv
Column 7	Width	Column 15	Lifetime 4 Deconv	Column 23	SNR 4 Deconv
Column 8	Height	Column 16	Intensity 1 Deconv		

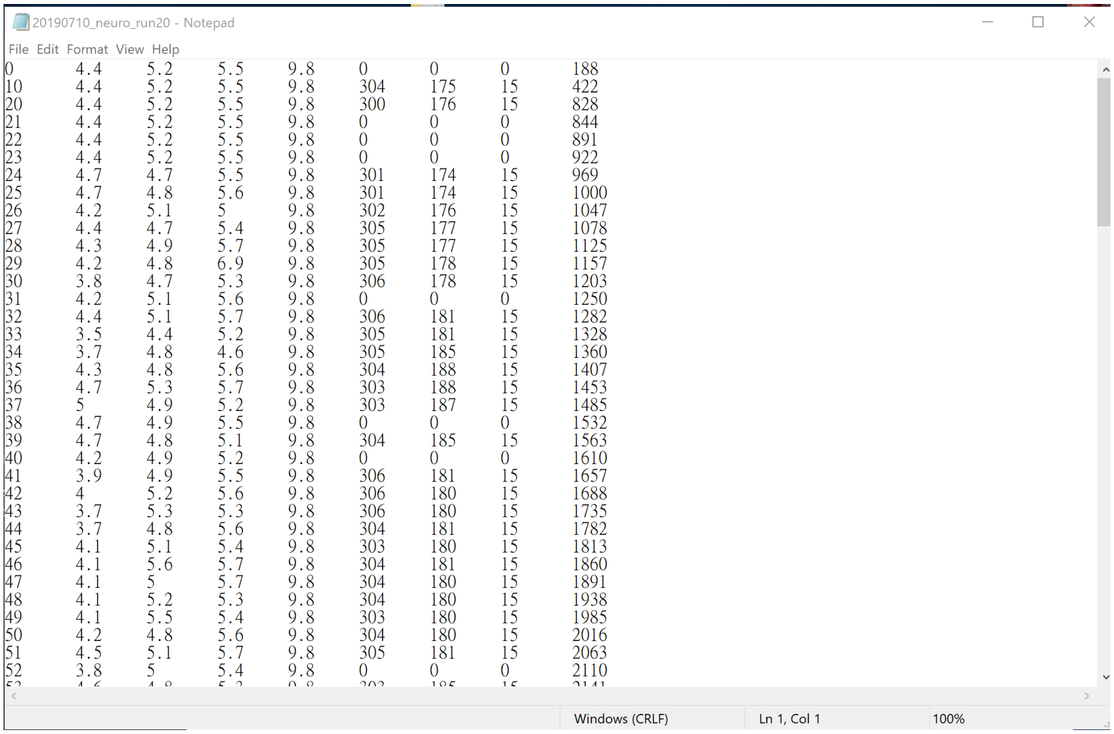
Table 1. The meaning of each column in H&N dataset. The columns with strikethrough are the data we do not use in the GUI.

(b) Radio Button: Brain (txt)

The second button is to process the brain dataset which is in the format of **text** file (.txt) containing **9 columns** as Fig. 5 shows. The meaning of each column can be found in Table 2, while column 9 will not be used for constructing the map.

Column 1	ID	Column 4	Lifetime 3	Column 7	Height
Column 2	Lifetime 1	Column 5	Lifetime 4	Column 8	Radius
Column 3	Lifetime 2	Column 6	Width	Column 9	~~Unknown~~

Table 2. The meaning of each column in the Brain (Txt) dataset. The column with strikethrough is the data we do not use in the execution of GUI.



0	4.4	5.2	5.5	9.8	0	0	0	188
10	4.4	5.2	5.5	9.8	304	175	15	422
20	4.4	5.2	5.5	9.8	300	176	15	828
21	4.4	5.2	5.5	9.8	0	0	0	844
22	4.4	5.2	5.5	9.8	0	0	0	891
23	4.4	5.2	5.5	9.8	0	0	0	922
24	4.7	4.7	5.5	9.8	301	174	15	969
25	4.7	4.8	5.6	9.8	301	174	15	1000
26	4.2	5.1	5	9.8	302	176	15	1047
27	4.4	4.7	5.4	9.8	305	177	15	1078
28	4.3	4.9	5.7	9.8	305	177	15	1125
29	4.2	4.8	6.9	9.8	305	178	15	1157
30	3.8	4.7	5.3	9.8	306	178	15	1203
31	4.2	5.1	5.6	9.8	0	0	0	1250
32	4.4	5.1	5.7	9.8	306	181	15	1282
33	3.5	4.4	5.2	9.8	305	181	15	1328
34	3.7	4.8	4.6	9.8	305	185	15	1360
35	4.3	4.8	5.6	9.8	304	188	15	1407
36	4.7	5.3	5.7	9.8	303	188	15	1453
37	5	4.9	5.2	9.8	303	187	15	1485
38	4.7	4.9	5.5	9.8	0	0	0	1532
39	4.7	4.8	5.1	9.8	304	185	15	1563
40	4.2	4.9	5.2	9.8	0	0	0	1610
41	3.9	4.9	5.5	9.8	306	181	15	1657
42	4	5.2	5.6	9.8	306	180	15	1688
43	3.7	5.3	5.3	9.8	306	180	15	1735
44	3.7	4.8	5.6	9.8	304	181	15	1782
45	4.1	5.1	5.4	9.8	303	180	15	1813
46	4.1	5.6	5.7	9.8	304	181	15	1860
47	4.1	5	5.7	9.8	304	180	15	1891
48	4.1	5.2	5.3	9.8	304	180	15	1938
49	4.1	5.5	5.4	9.8	303	180	15	1985
50	4.2	4.8	5.6	9.8	304	180	15	2016
51	4.5	5.1	5.7	9.8	305	181	15	2063
52	3.8	5	5.4	9.8	0	0	0	2110

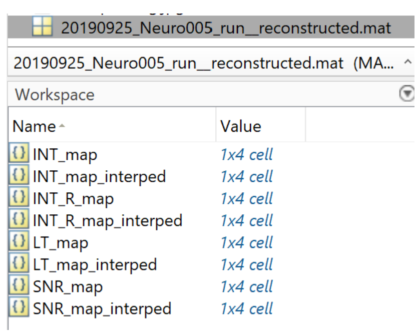
Figure 5. The Brain Txt file format (The example is 20190710_neuro_run20.txt).

(c) Radio Button: Brain (reconstructed)

The third button is to process brain dataset which is in the format of **mat** file (.mat) containing **eight 1x4 cell** with the **suffix "reconstructed"** in its name like Fig 6. shows. Each 1x4 cell includes the different measurements as Table 3 shows. Each cell of each 1x4 cell includes the measurement values of each channel in matrix format corresponding to the raw image.

INT_map	~~Intensity values~~	LT_map	Lifetime values
INT_map_interped	~~Unknown~~	LT_map_interped	~~Unknown~~
INT_R_map	Intensity Ratio values	SNR_map	SNR values
INT_R_map_interped	~~Unknown~~	SNR_map_interped	~~Unknown~~

Table 3. The meaning of each 1x4 cell in the Brain (reconstructed) dataset. The columns with strikethrough are the data we do not use for the execution of constructing the map.

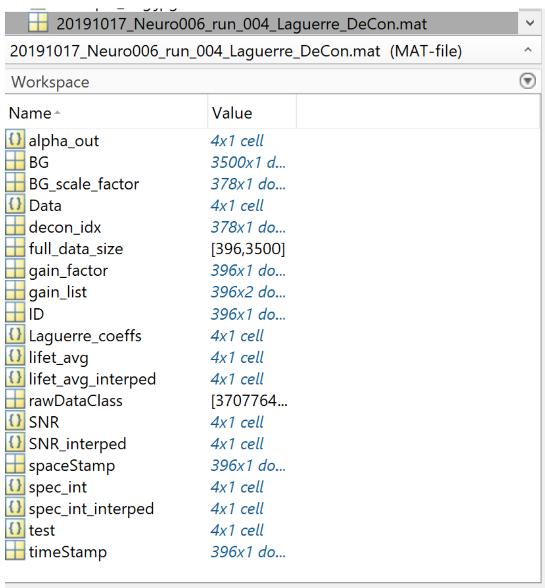


Name	Value
INT_map	1x4 cell
INT_map_interped	1x4 cell
INT_R_map	1x4 cell
INT_R_map_interped	1x4 cell
LT_map	1x4 cell
LT_map_interped	1x4 cell
SNR_map	1x4 cell
SNR_map_interped	1x4 cell

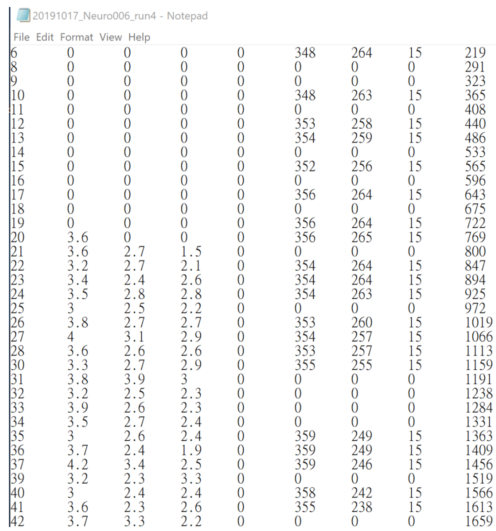
Figure 6. The Brain (reconstructed) file format (example: 20190925_Neuro005_run_reconstruncted.mat)

(d) Radio Button: Brain (mat+txt)

The last button is to process the brain dataset which is in the format of **mat** file (.mat) but with the suffix "**Laguerre_DeCon**" as Fig. 7 shows. However, since such kind of dataset does not include the width and height data, you also need to import its corresponding **txt** file (.txt) to acquire the width and height values in order to successfully process the map generation. To process this kind of dataset, we need to use the "ID", "SNR", "lifet_avg", "spec_int" in the mat file and column 1, 6, 7 in the text file.



(a)



(b)

Figure 6. The mat file example (a) and the text file example (b) of the Brain (mat+txt) format (20191017_Neuro006_run_004_Laguerre_DeCom.mat) (20191017_Neuro006_run_004.txt)

(2) Load the files

The next step is to import the raw image file and the dataset. To input the image file, please click the **browseIMG** radio button in the **box 2** in Fig. 3 and choose the image file you like to input. The imported image will be displayed in the window of the name "raw image". To input dataset, please click the **browseMAT** button and choose the dataset file you want to process. If you choose to process the **brain data (mat+txt)**, you also need to import the text file by simply clicking the **browseTXT** radio button. After inputting the files successfully, the file names will be displayed in the textboxes near the buttons. It is worth noting that the files can be in any folder in your computer, not only restricted to the folder the tool in.

(3) Select the category of the maps

There are four kinds of map you could generate through this tool, lifetime, intensity ratio, intensity weighted lifetime, optical redox ratio. Each category has its corresponding radio button as it shows in the **box 3** in Fig. 3. Here IW Lifetime denotes the intensity weighted lifetime.

When calculating the intensity ratio and the intensity weighted lifetime, you could determine whether you want to include channel 4 to calculate those values by toggling the **Including CH4** checkbox.

(4) Determine the threshold and the plotting method

In the **box 4** in Fig.3, you need to first determine which **channel** you want to plot by selecting the radio button **CH1**, **CH2**, or **CH3**.

After that, you need to set the **SNR lowerbound**. There are three options: (1) **0**, (2) **30**, (3) **others**. By choosing **others**, you can type any positive value you like into the box near the scroll bar of SNR lowerbound.

Then, you need to determine the method for plotting the map. There are three major methods can be used: standard method, natural neighbor interpolation, and inverse distance weighting (IDW). To use standard method, simply select **Standard** radio button. To use natural neighbor interpolation, simply select **Natural Neighbor** radio button. For IDW, you can choose different p (decay parameter) to produce different results. To set the p to be 2, simply select the **IDW P=2** radio button. To set the p to be 3, simply select the **IDW P=3** radio button. To set the p to be 1, simply select the **IDW P=1** radio button. If you want to use customized p value, simply select the **IDW P=** radio button and type the desired value into the box on the right of the IDW P radio button. Moreover, you could decide the type of IDW by changing the option in the scroll bar of **Type of IDW**. To use global IDW, simply click the **Global** in the message box. To use local IDW, simply click the **Local** in the message box and type the local radius value into the text box on the right.

Finally, when using natural neighbor interpolation or inverse distance weighting, you need to decide the required number of data and radius of searching region to visualize the reasonable region of the map. Examples regarding these two parameters are illustrated in the Section 4.

(5) Start plotting map

After selecting the target dataset, loading the related files, and deciding the desired kinds of the maps, you can click the **Plot Overlay** button in the **box 5** in Fig. 3. Following clicking the button, there will be a progress bar popped up as shown in Fig. 7, displaying the progress of the plotting process.

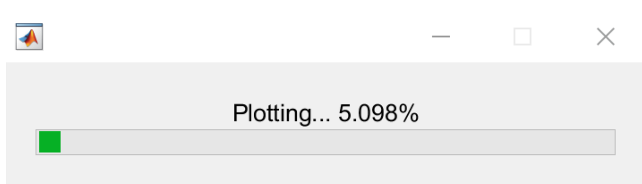


Figure 7. After clicking the "Plot Overlay" button, the progress bar will pop up and show the progress of the generation of the map.

(6) Appearance setting

After plotting successfully, you can modify the appearance of the map in the **box 6**, **Configurations section,** on the bottom left of the GUI in Fig. 3. In this part, you could **(a)** check the **Grayscale** checkbox to convert the background image from RGB image to grayscale image, **(b)** check the **Alpha** checkbox and type in the desired value to change the transparency of the image, **(c)** change the value in **Scale** to change the scale of the color bar, **(d)** choose the colormap you like in **Color map**.

After setting all the values, simply click Change Setting button to change the appearance of the result. Note that the default value of the scale of the color bar is set from $\mu - 2\sigma\ to\ \mu + 2\sigma$ (μ is the mean of the data while σ is the standard deviation of the data). Besides, the "Statistical Scale" button helps you reset the value of the scale to be the default value which is $\mu - 2\sigma \sim \mu + 2\sigma$.

(7) Export the resultant image

After plotting the map, you will be able to export the resultant image to any path with customized name in your computer in the **box 7** in Fig. 3. To select the path where you want to export the image, simply click the **Path** button. To customize the name of image file, simply toggle the **Preferred name** checkbox and type the desired filename into the message box on the right. Note that the default name of the image file will be formed using Method+Channel+LowerBound+Date, for example, Global IDW, P=2, Channel=2, LowerBound=30, (20-05-07).jpg. After choosing the desired path and/or name, simply click the **Save** button on the right bottom of the **box 7**.

(8) SNR map

After plotting the overlay, you could also generate the SNR map by simply clicking the **SNR map** button in **box 8** in Fig. 3. The SNR map is generated by using standard method. The scale of the map is from the lowest SNR to the highest SNR. The constructed SNR map will be automatically stored in the folder with the name SNR.jpg

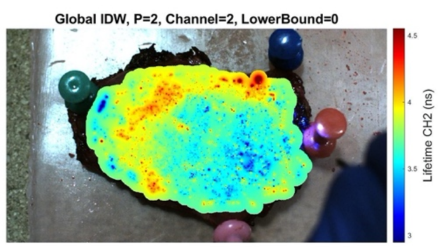
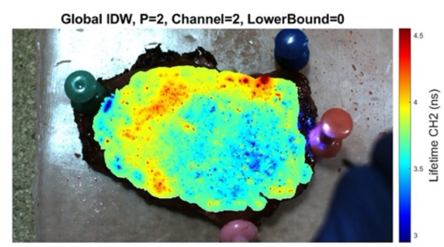
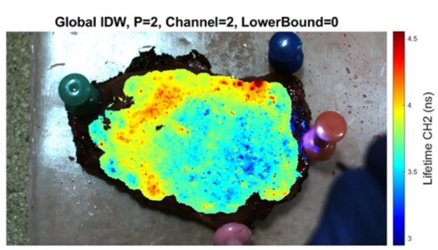
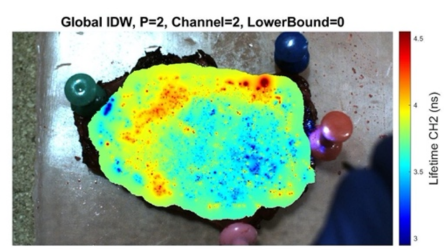
4. Examples and Discussion

In this section, we explain the purpose of the two parameters: (1) number of required data (2) radius of searching region. Then we show the results generated by using different methods in order for user to select the appropriate method to produce the desired appearance of the map.

(1) Required number of data (N) & Radius of searching region (r)

When using spatial interpolation, we need to specify the region/pixels we would like to visualize. Therefore, these two parameters could help you decide the reasonable region to visualize.

For the implementation of the algorithm, we go through every pixel of the raw image and calculate the number of the measurements inside the **searching region of given radius (r)**. If the number of the measurements is larger than or equal to the **required number of data (N)** we set, we will visualize that pixel. If not, we will not visualize that pixel. A good combination for the image of 720x1280 pixels resolution is (r,N) = (30,1). Here we show the resultant images generated by using different combination of r and N.

(r,N) = (30,1)	(r,N) = (30,5)
	
(r,N) = (30,10)	(r,N) = (50,10)
	

(2) Visualization Method

(a) Standard Method

The standard method is the current algorithm used for FLIm visualization. The idea behind the standard method is to visualize the measurements by plotting a set of discs as Fig. 8 shows.

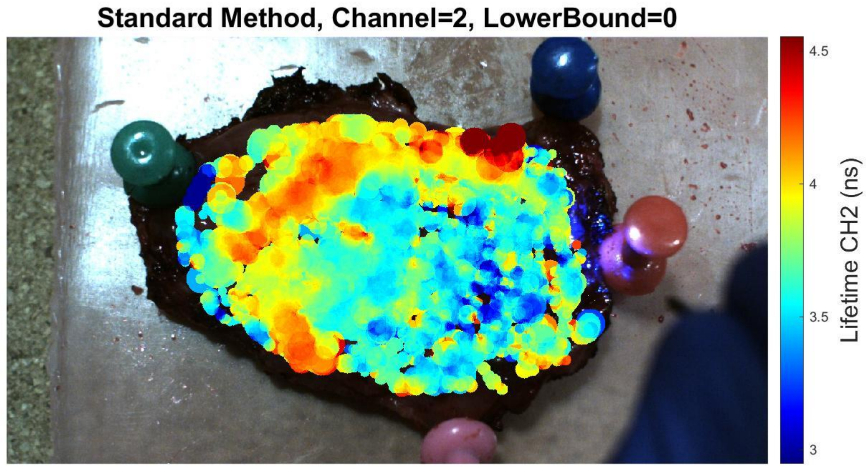


Figure 8. FLIm map constructed by using standard method

(b) Natural Neighbor Interpolation

Natural neighbor interpolation is a spatial interpolation method calculated based on the distribution of the measurement. However, there are usually some polygon shapes on the peripheral region of the map as Fig. 9 shows.

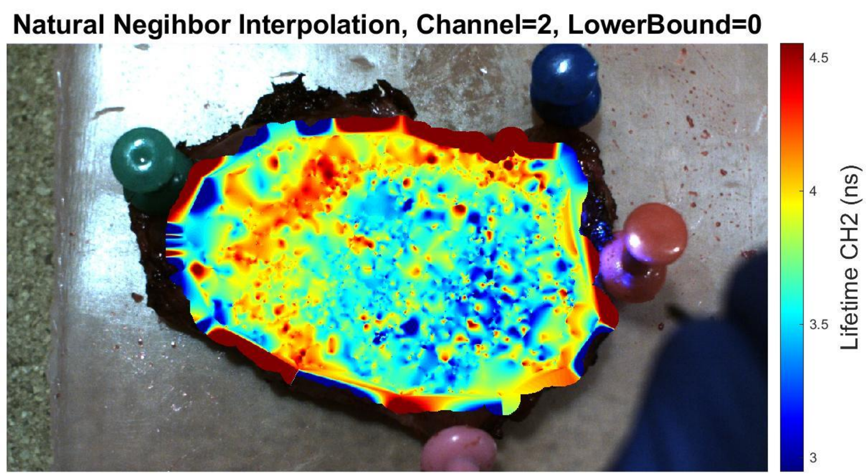


Figure 9. FLIm map constructed by using natural neighbor interpolation

(c) Inverse Distance Weighting (IDW)

Different combination of the parameters of the IDW will produce different results. When the decay parameter p becomes lower, the smoother FLIm map you will get. When the radius of local value becomes lower, the higher contrast FLIm map you will get. Below are the images produced by using different combination of the IDW parameters. A good choice is Local IDW with decay parameter p = 1 and radius value R = 30.

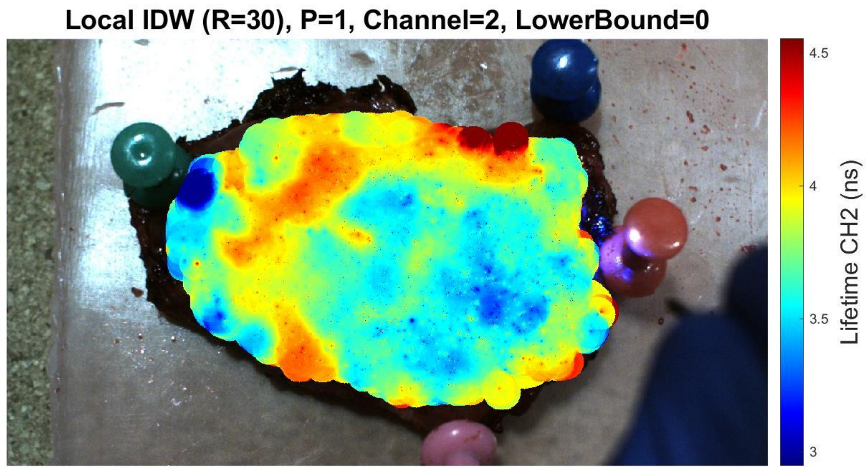


Figure 10. FLIm map constructed by using Local IDW with p=1 and R=30

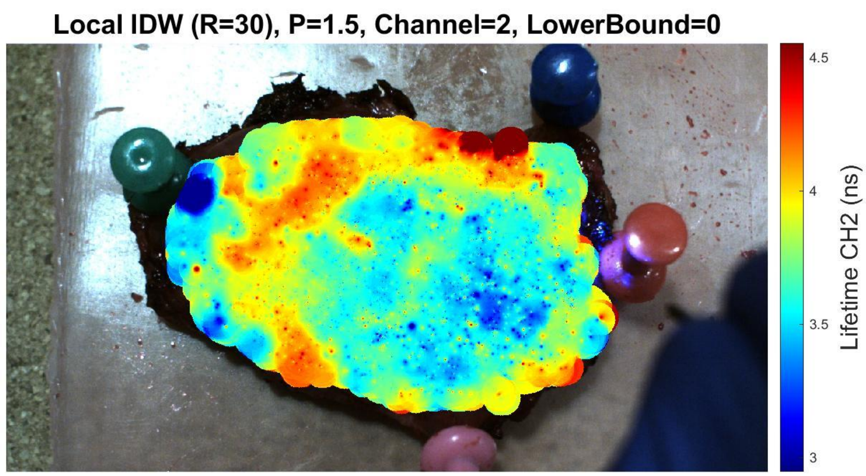


Figure 11. FLIm map constructed by using Local IDW with p=1.5 and R=30

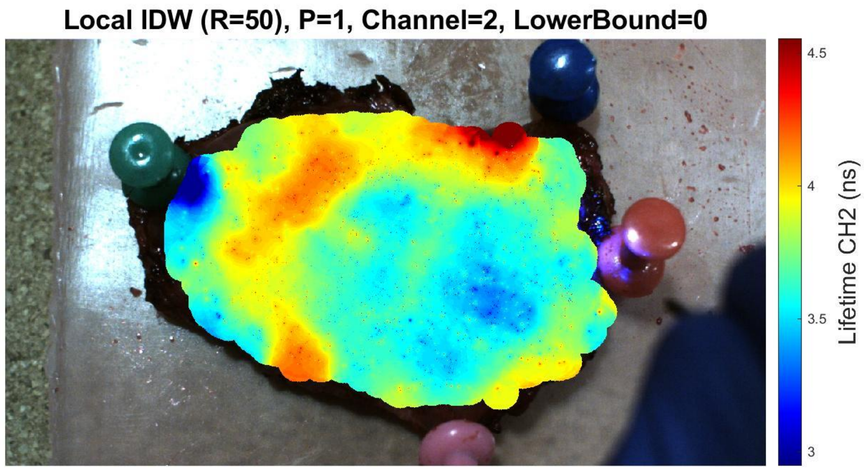


Figure 12. FLIm map constructed by using Local IDW with p=1 and R=50

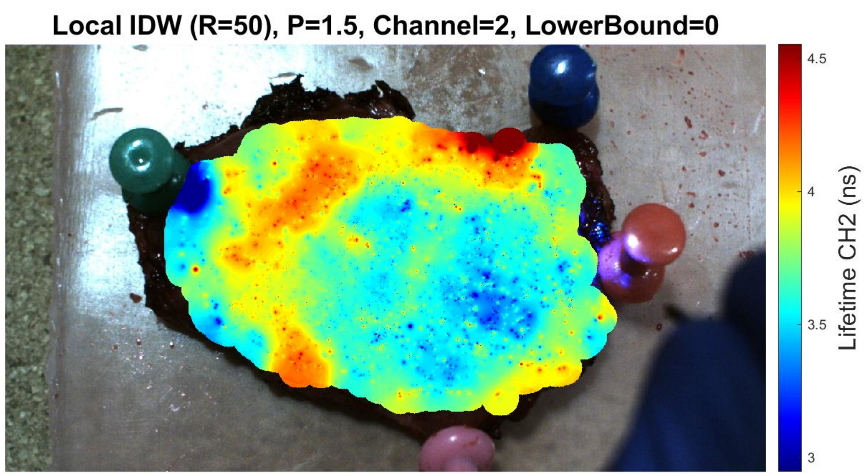


Figure 13. FLIm map constructed by using Local IDW with p=1.5 and R=50

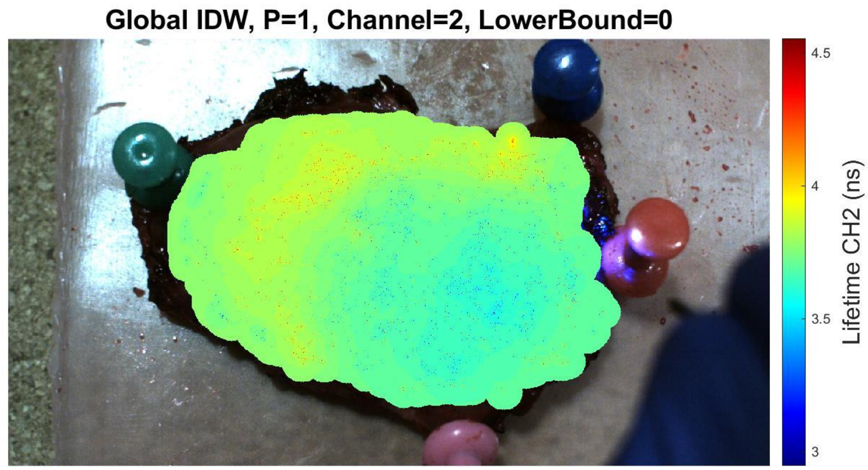


Figure 14. FLIm map constructed by using Global IDW with p=1

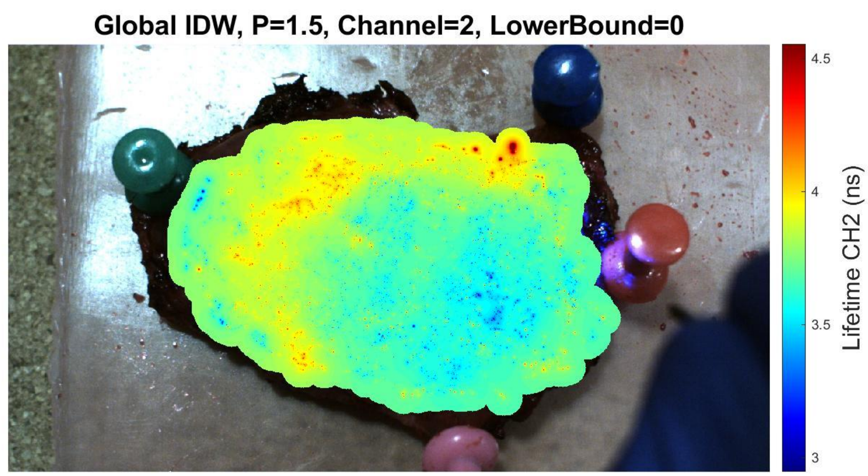


Figure 15. FLIm map constructed by using Global IDW with p=1.5