FINAL PROJECT.

```
%%%%%%%%%%%%%%%%%%%%%% Initial Data managment %%%%%%%%%%%%%%%%%%%

%this part of the code gives access to tif video.
reader = bfGetReader('pos_1B_20211206S1_spHEK_Mag_60s_90s_5%_5fps_27oC_fiji.tif');
```

```
log4j:WARN No appenders could be found for logger (loci.formats.ClassList).
log4j:WARN Please initialize the log4j system properly.
```

```
% get the size of the video
Fields=reader.getSizeT
```

```
Fields = 1
```

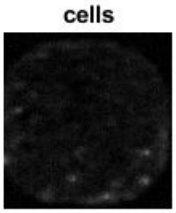
```
Channel=reader.getSizeC
```

```
Channel = 1
```

```
% get the length of the video
Time=reader.getSizeZ
```

```
Time = 700
```

```
%getting acces to the first frame of the video
chan = 1; z = 1; time = 1;
ind = reader.getIndex(z - 1, chan - 1, time - 1) + 1;
img1_change = bfGetPlane(reader,ind);
img1_original = bfGetPlane(reader,ind);

%selecting the first well of illustrated in the frame
img1_original=img1_original(1:110,1:105);

%imaging this first well
figure()
imshow(img1_original,[0 1000])
title('cells')
```

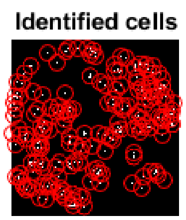



```
%%%%%%%%%%% Identify single of cells %%%%%%%%%%

%for this we will process the previous image to segementate it and identify
```

```
%single cells.

%change the image to black and white.
img1_change = img1_change(1:110,1:105);
img1_change = im2bw(img1_change,0.0016);

% we created the function 'segmentame_esta'
% which will segmentate groups of cells to single cells.
img1_change = segmentame_esta(img1_change);
img1_change = bwpropfilt(img1_change,'Area',[1 10])
```

```
img1_change = 110×105 logical array
   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0···
   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0
   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0
   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0
   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0
   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0
   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0
   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0
   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0
   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0   0
     ⋮
```

```
%then we used region props to identify the centroids in the
img1_centroids= regionprops(img1_change,'centroid');
img1_area= regionprops(img1_change,'area');
fig=can_I_see_the_centroids(img1_centroids,img1_change);
```




```
centroids_set=[];
original_intensity1=[];

%%%%%%%%%%%%% detect the fluoresence change on the centroids %%%%%%%%%%%%

% This for loop will scan the change of intesnsity on the centroids
% initially dectected.

for i=1:length(img1_centroids)
centroids_set(i,:) = round(struct2array(img1_centroids(i)));
check = centroids_set(i,:);
original_intensity1(i,:) = img1_original(check(1),check(2));
end
```

```
centroids_set
```

```
centroids_set = 174×2
     3    36
     2    40
     1    59
     3    44
     2    48
     3    66
     3    86
     4    27
     5    56
     5    87
     ⋮
```

```
original_intensity1
```

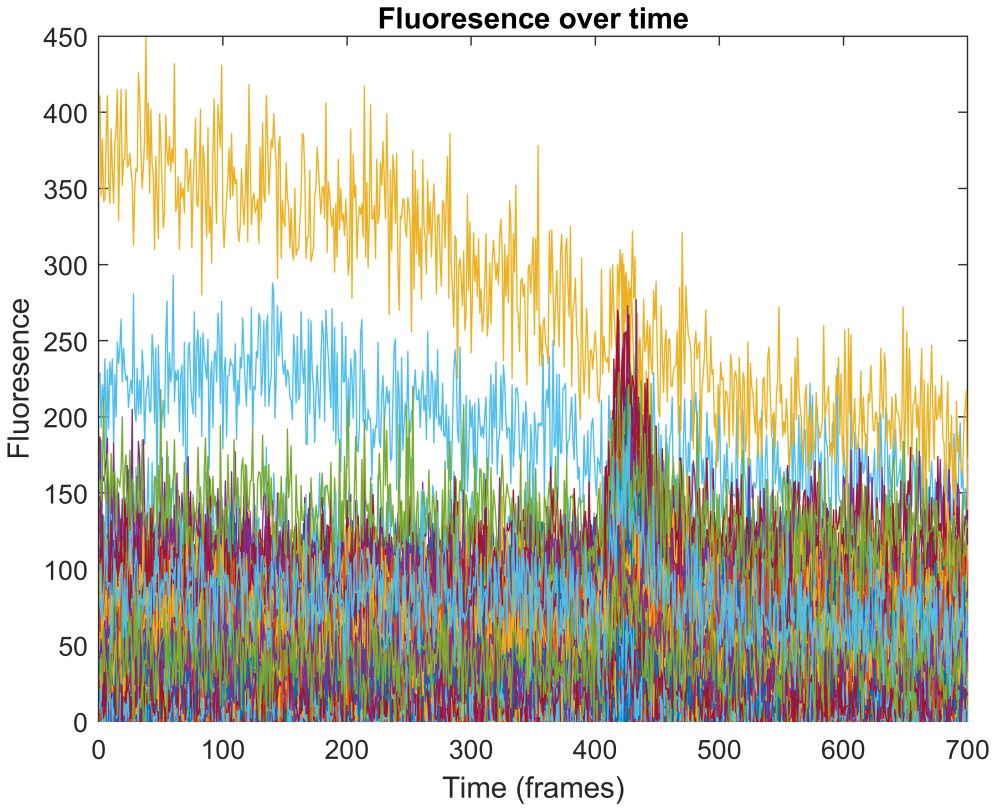
```
original_intensity1 = 174×1
     0
    24
    13
    21
     2
    14
    26
    30
    40
     3
     ⋮
```

```
% original_intensity contains the intensity of a centroid over the 700 time
% points

original_intensity=centroids_overtime(img1_centroids,reader)
```

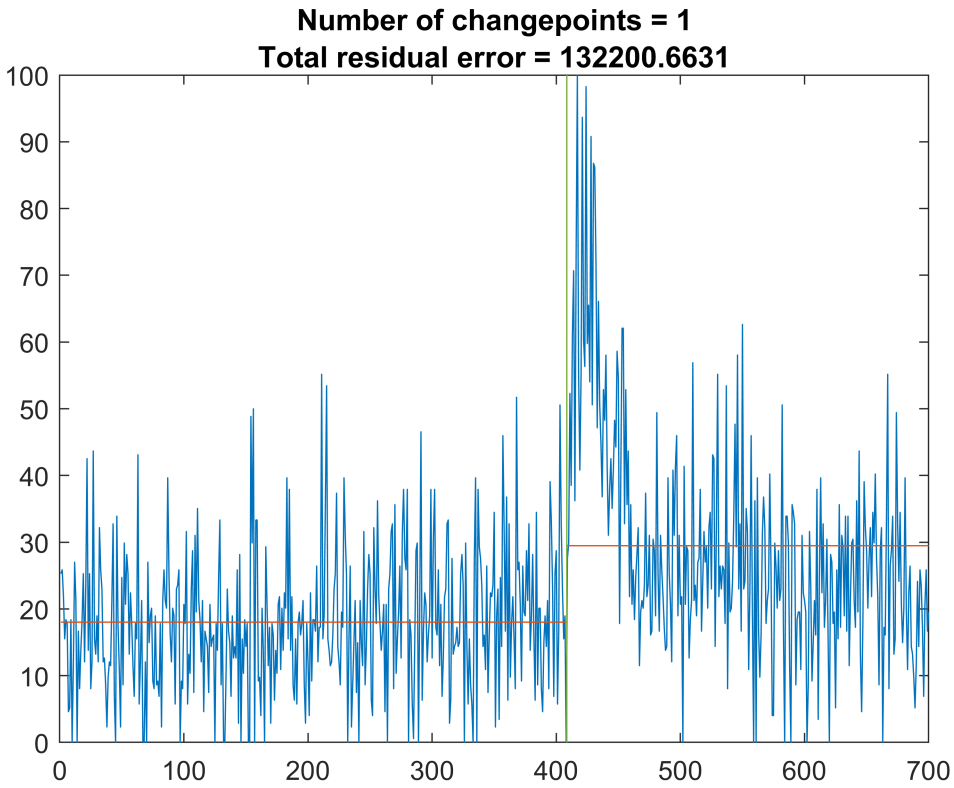
```
original_intensity = 174×700
     0     0     0     7     1     0    40    45     0     0     0    11    37 ···
    24    25    14    23    15     9    13     5    22    43    19    13    24
    13    19     0    44    53    16    16    11     5     0    32     6    26
    21    26    10    29     2    11     0    27    26     6     0     0     4
     2    25    21     0     0    14    27     0     4    14    11     0    17
    14     5    19     5    25    23     8    10     0     0    10     2     6
    26    41    35    54    12    39    31    50    21    27     6    25     2
    30    18    30    19    18     0     8    36    28    15    15    11    14
    40    50    63    47    48    22    41    72    68    41    38    51    25
     3     0     0    21     8     0    15    32     0     3     0    13     0
     ⋮
```

```
%identifyinf the maximum intensity over the time
maximum = max(original_intensity,[],2);

% Average of intesity over the time
New_original_intensity=original_intensity*100./maximum;
```

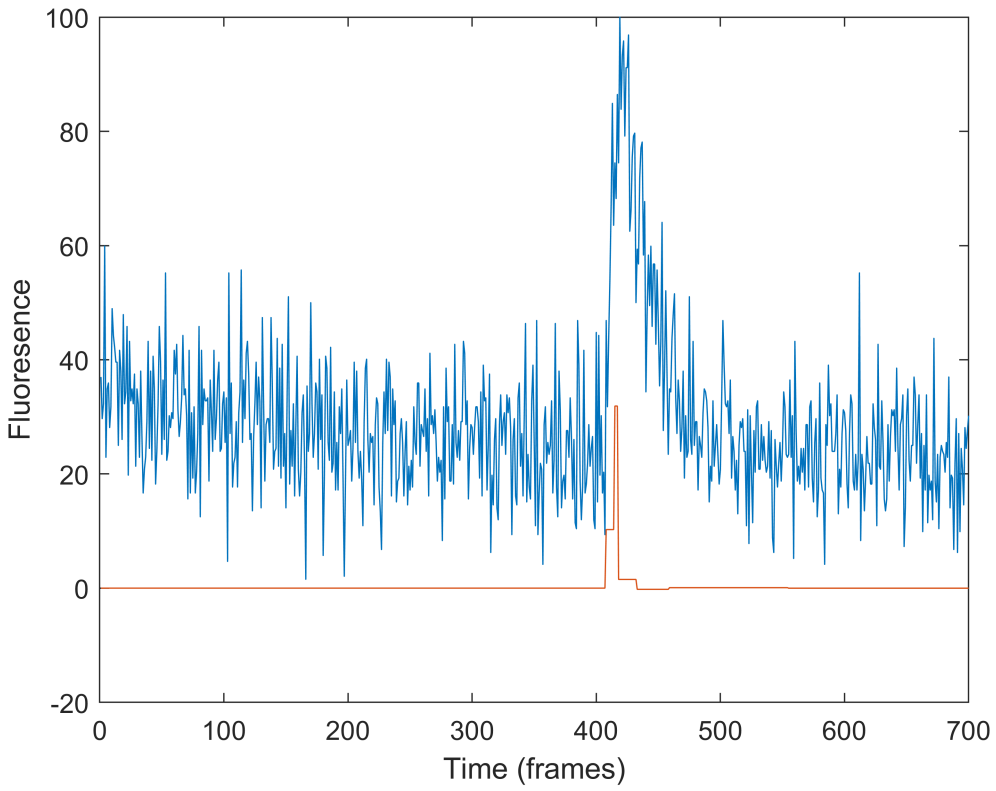
```
%ploting all the singnals found
figure()
plot(1:700,original_intensity)
title('Fluoresence over time')
xlabel('Time (frames)')
ylabel('Fluoresence')
```




```
%Detecting drastic changes on the fluoresence. (try)
findchangepts(New_original_intensity(25,:))
```

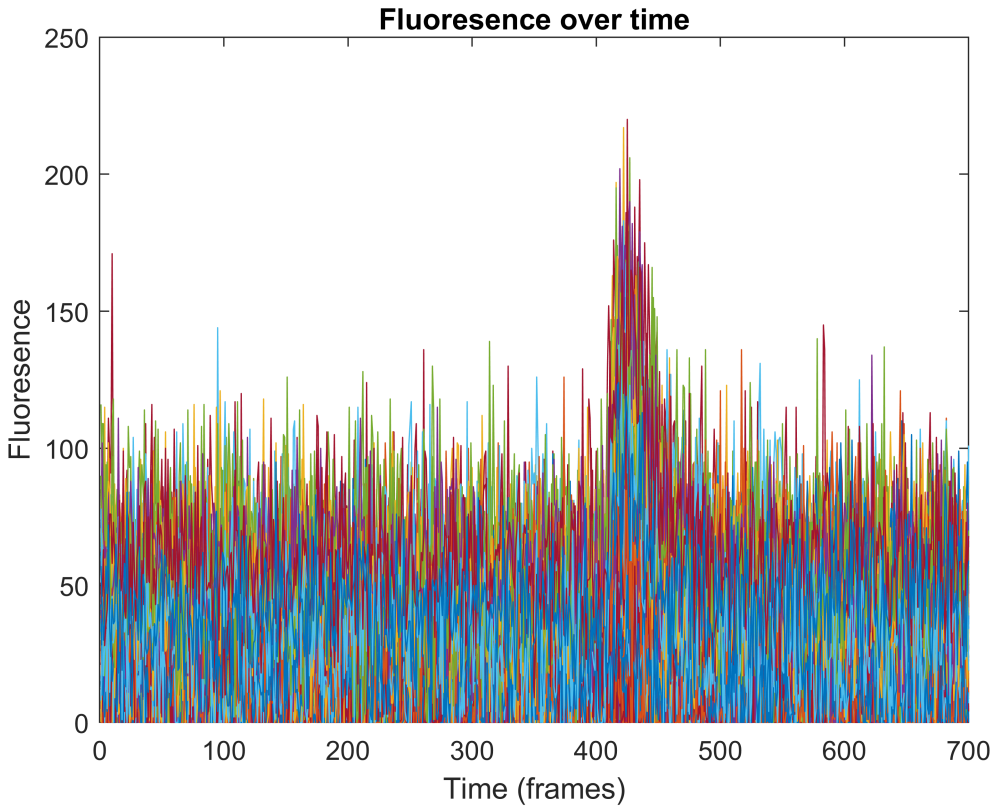



```
%Detecting drastic changes on the fluoresence. (Selected)
[~,S1,~] = ischange(New_original_intensity(25,:),'linear','Threshold',1300);

figure()
plot(New_original_intensity(12,:))
hold on
plot(S1)
xlabel('Time (frames)')
ylabel('Fluoresence')
hold off
```

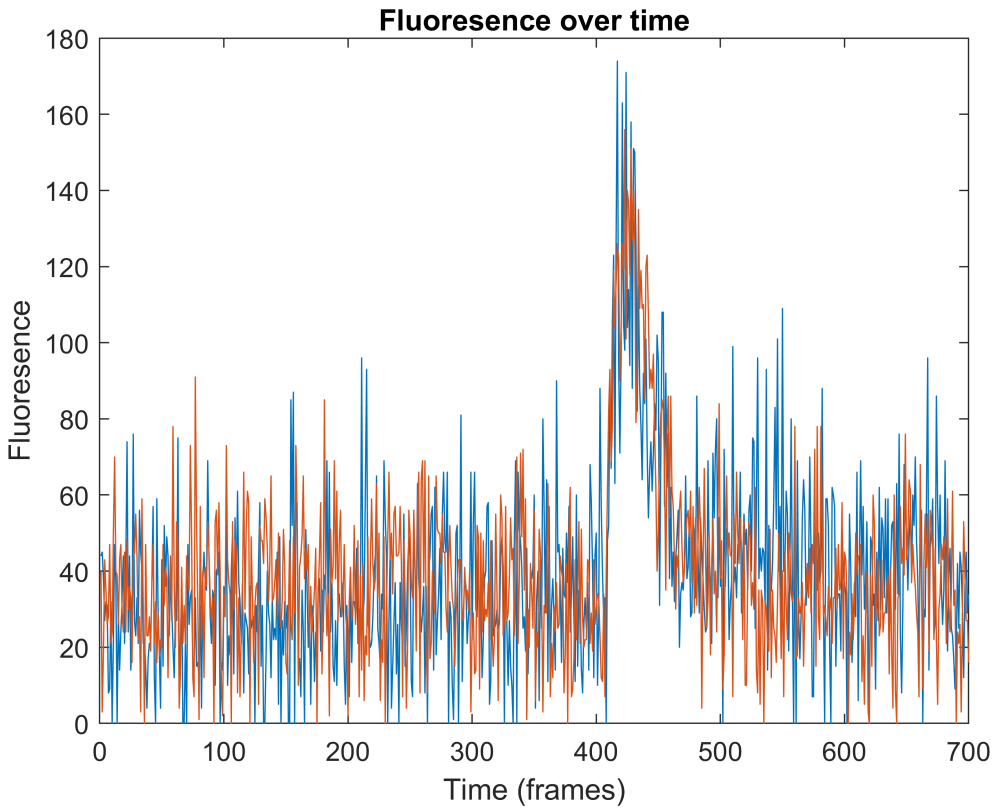



```
% Maximun values of each cell over the time.
maximum = max(original_intensity,[],2);

% Mean intesnity over the first 200 timepoints.
mean_total=  mean(original_intensity(:,1:200),2);

% Percentage of original intensity.
new_original_intensity=original_intensity*100./maximum;


% Selecting just data with a significant change of intensity.
total_data=[];
for i=1:length(mean_total)
    [~,S1,~] = ischange(New_original_intensity(i,:),'linear','Threshold',450);
    if max(S1)+10 > mean((S1(1,1:100)+10)*5) && mean(New_original_intensity(i,1:100))*3 < max(N
       total_data=[total_data;original_intensity(i,:)];
    end
end

total_data;
new_original_intensity=original_intensity*100./maximum;

%refined  data plotted. some of them present a drastic change around the time point 400.
```

```
figure()
plot(1:700,total_data)
title('Fluoresence over time')
xlabel('Time (frames)')
ylabel('Fluoresence')
```




```
%there is some data that doesnt show a drastic change.
figure()
plot(1:700,total_data(19:20,:))
title('Fluoresence over time')
xlabel('Time (frames)')
ylabel('Fluoresence')
```




```
%space to discuss a better aproach to detect changes but the algorithm
%currently is not working which would be a good one?


A=length(original_intensity(:,1))
```

```
A = 174
```

```
B=length(total_data(:,1))
```

```
B = 57
```

```
percentageof_Wrong_data_eliminated = (A - B)*100/A
```

```
percentageof_Wrong_data_eliminated = 67.2414
```

FUNCTIONS

```
function original_intensity=centroids_overtime(img1_centroids,reader)
```

```
centroids_set=[];
original_intensity=[];

for ii = 1:700

    chan = 1; z = 1;
    time=1;
    ind = reader.getIndex(ii - 1, chan - 1, time - 1) + 1;
    img1_original = bfGetPlane(reader,ind);
    img1_original=img1_original(1:110,1:105);


    for i=1:length(img1_centroids)

        centroids_set(i,:) = round(struct2array(img1_centroids(i)));
        check = centroids_set(i,:);
        original_intensity(i,ii) = img1_original(check(1),check(2));

    end


end

end

function fig=can_I_see_the_centroids(stat1B,I1)
%plot centroids
fig=0;
figure();
imshow(I1); hold on; title('Identified cells')
for x = 1: numel(stat1B)
    plot(stat1B(x).Centroid(1),stat1B(x).Centroid(2),'ro');
end

end

function [I1,I2,I3]=gimme_the_images(I,levelr,levelg,levelb)

rmat=I(:,:,1);
gmat=I(:,:,2);
bmat=I(:,:,3);

I1 = im2bw(rmat,levelr);
I2 = im2bw(gmat,levelg);
I3 = im2bw(bmat,levelb);

end

function segmented_image = segmentame_esta(BW2)
    D = 255-uint8(bwdist(~BW2));
    gs=watershed(D);
    ws=gs==0;
    segmented_image = BW2 & ~ws;
end
```