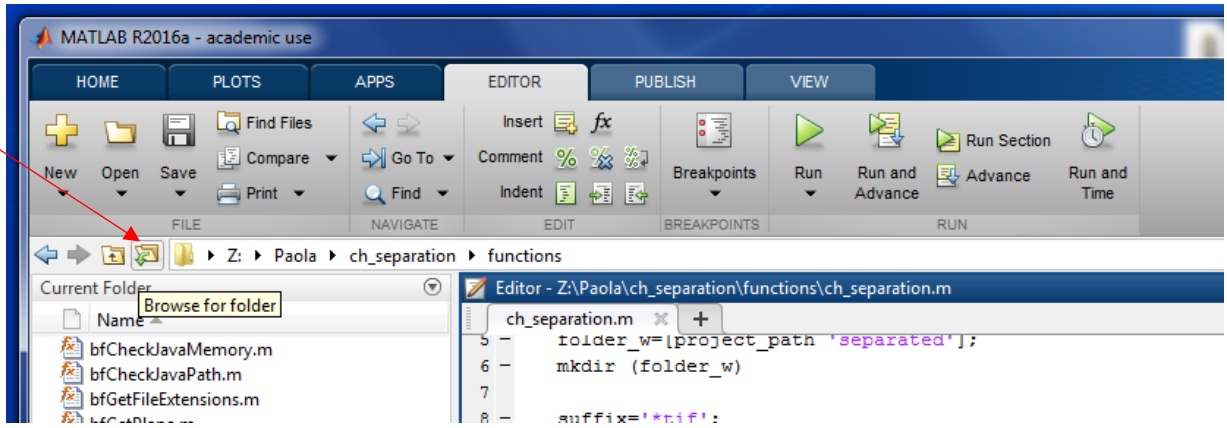
PROTOCOL: CHANNELS SEPARATION + OMEFILES (DATA FROM SPINNING DISK)

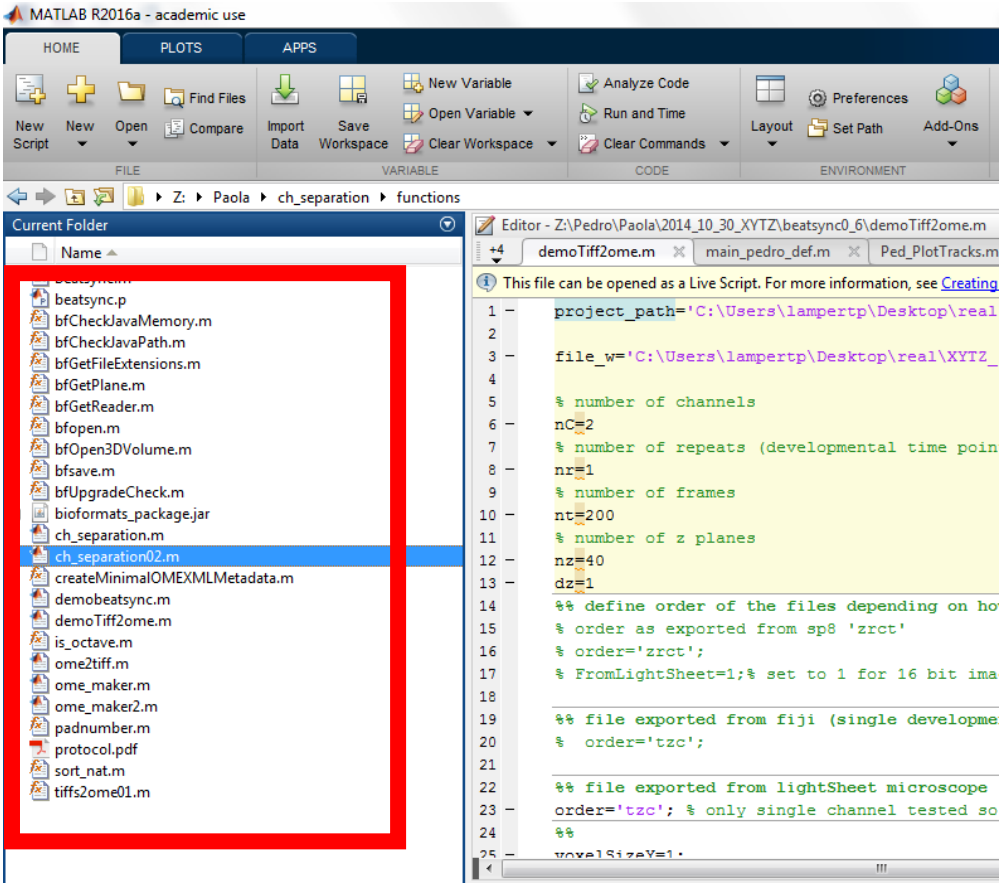
The following protocol uses the functions ch_separation02.m and tiffs2ome01.m that are in 'labo5\paola\ch_separation\functions'. Use this protocol and these functions if:

- **YOUR DATA ARE FROM THE SPINNING DISK**
- **THE CHANNELS ARE NOT SPLITTED**
- **YOU HAVE ONLY 1 DEVELOPMENTAL TIME (NO TIMELAPSES)**

Start Matlab
Copy the folder Labo5\Paola\ch_separation\functions on your desktop
Choose the folder you created as Matlab directory, by clicking here:

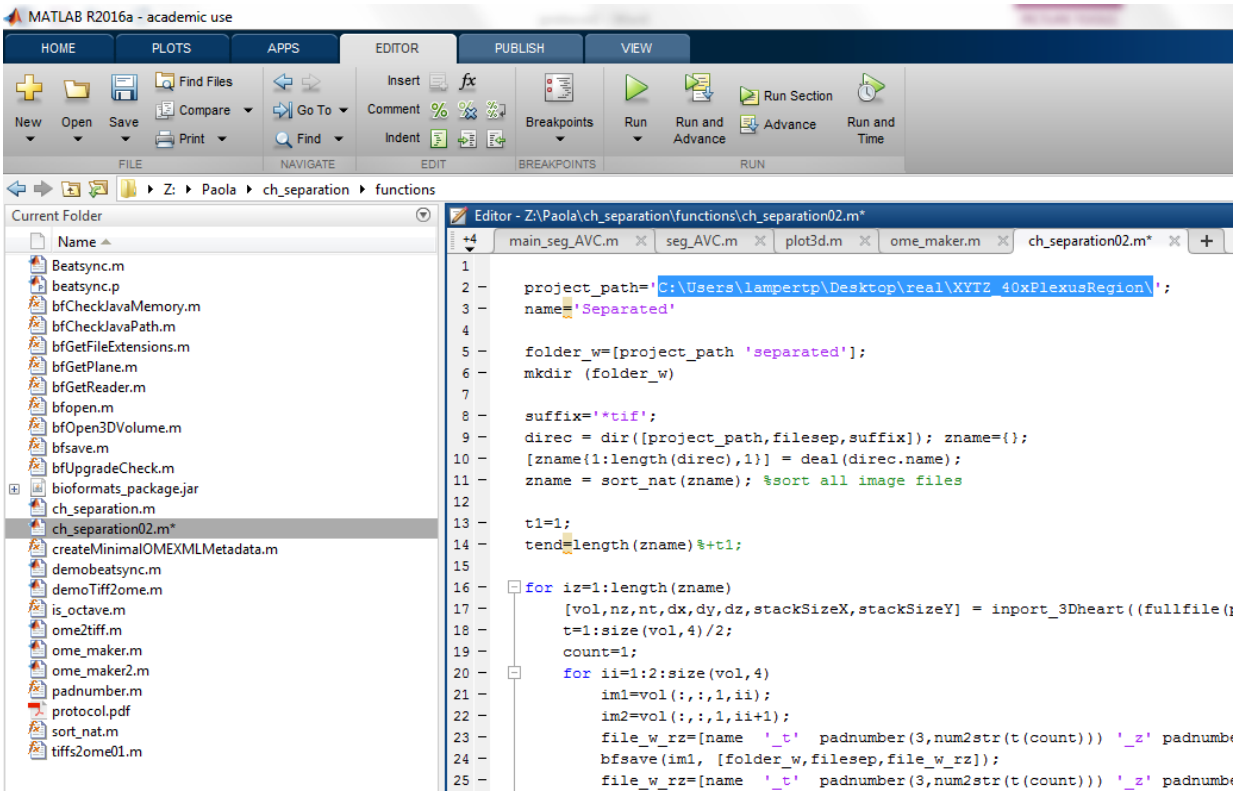


And choosing the right folder.
A list of functions will appear.



Double click on ch_separation02.m

Define as project_path the path of the folder where the data from the spinning disk are saved (ONLY ONE EXPERIMENT IN ONE FOLDER!).
The path has to be between ‘ and \’;



Define the number of channel as nC, the number of frames as nt and the number of z planes as nz.
Don’t change nr!

```
% number of channels
nC=2
% number of repeats (developmental time points)
nr=1
% number of frames
nt=200
% number of z planes
nz=201
dz=1
%% define order of the files depending on how they were exported
% order as exported from sp8 'zrct'
% order='zrct';
% FromLightSheet=1;% set to 1 for 16 bit images

%% file exported from fiji (single developmental point) exported from sp8 'zrct'
%  order='tzc';

%% file exported from lightSheet microscope (single developmental point) exported from sp8 'zrc
order='tzc'; % only single channel tested so far
%%
voxelSizeY=1;
```

Run the code.
A folder named Separated will appear in the same folder where the data are saved.
Inside a list of images is saved in the order tzc.

A folder named Omefiles will appear in the folder Separated.
Inside a list of ome files is saved in the order rz.
The ome files are the files you need for the realignment!