

# Methods for Video Analysis in Bio-medical Images (an ImageJ based tutorial)

A few links : <http://gibbs.engr.cuny.cuny.edu/technical/Tracking/RoachTrack.php>

for MATLAB fans or <http://fiji.sc/Fiji> or <http://icy.bioimageanalysis.org/> or <http://www.ipol.im/>

I chose Fiji.

**Resources:** <http://www.math-info.univ-paris5.fr/~lomn/Cours/CV/Material/>

(file *TPVideo.pdf* and software *fiji-linux64.Core.tar.gz* and Data)(For the lecture see </Cours/CV/BME/>)

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## Part 1 : Optical Flow

- Start by applying a **small** rigid transform to an image like the *blobs.gif* one. **Menu Transform/Interactive Rigid (exists in Fiji : rotate 5 degrees and translate 5 pixels if not available)**
  - Save *blobs.gif* as *blobs1.gif* and the transformed one as *blobs2.gif*.
  - Open the two images and make a stack out of both of them. **Menu Images/Images to Stack (or so)**
- a stack is like a video sequence for us ? Can it be like a 3D image ?
- Apply the plugin **Optical Flow/Gaussian approach and check the Display color map box.** Analyze the results. (exists in Fiji in the menu Plugins/Optic Flow)
- What does it mean Gaussian here ?
- Do it again after applying an **Integral Image Filter** like the **Mean**.
- Are the results very different ? Do you know the difference between Mean and Median filtering ?
- From *books*, create an image sequence by importation of the folder *books (Images to Track)*. Then apply image processing on it for pre-processing and then use the **Optical Flow** menus.
- Then compute various optical flows with **bij/FlowJ** (install the plugin from <http://bij.isi.uu.nl/> and <http://webscreen.ophth.uiowa.edu/bij/flowj.htm> or my website) (exists in Fiji in the menu Analyze/Optic Flow)
- Lukas and Kanade is the extended algorithm based on the one described in the lecture.
- From two medical images from a sequence like *t420.tif* and *t421.tif* (from the ISBI 2015 challenge [http://www.codesolorzano.com/celltrackingchallenge/Cell\\_Tracking\\_Challenge/Datasets.html](http://www.codesolorzano.com/celltrackingchallenge/Cell_Tracking_Challenge/Datasets.html)) find **differences, SIFT points of correspondences, flow etc.**
- what kind of images are they?
- With *t026.tif* et *t027.tif* create a stack, binarize by threshold adjustment, remove outliers, filter with a mean and process the gaussian optical flow. Try the **Lucas-Kanade** one in **Analyze/Optic Flow**
- is it adapted to this sequence ?

## Part 2 : ffmpeg to manipulate video sequence

It is a tool very useful for video editing (linket to the codev library lib-av). Find below examples :

```
ffmpeg -i input.avi -c:a aac -b:a 128k -c:v libx264 -crf 23 output.mp4
```

```
ffprobe -show_streams -i "file.mp4"
```

```
mediainfo Dream.House.sample.mkv
```

```
avprobe -show_streams file.mp4
```

```
ffmpeg -i slow.mp4 -s 320x240 -c:a copy smallslow.mp4
```

```
ffmpeg -i video.avi -an -vcodec rawvideo -y video2.avi
```

```
ffmpeg -i redcar.mp4 -i redcareverse.mp4 -filter_complex "blend=all_mode='overlay':all_opacity=1.0" output3.mp4
```

You need to distinguish between the container mp4 and the coding scheme like h264 bitstream see an example from <https://stackoverflow.com/questions/7333232/concatenate-two-mp4-files-using-ffmpeg>)

```
ffmpeg -i input1.mp4 -c copy -bsf:v h264_mp4toannexb -f mpegts input1.ts
```

```
ffmpeg -i input2.mp4 -c copy -bsf:v h264_mp4toannexb -f mpegts input2.ts
```

```
ffmpeg -i "concat:input1.ts|input2.ts" -c copy output.mp4
```

- Explain why some *avi* videos in the tutorial folders cannot be read by Fiji.
- Find a solution
- Please create the videos *redcaroverlay.mp4* video.

## Part 3 : Tracking

- Open the image *Track\_for\_TrakMate* from the samples menu.
- Project the stacks on one image with max intensity (Menu **Image/Stack/ZProject**)
- Threshold it with Adjust threshold.
- Try to use the TrackMate plugin ([https://imagej.net/Getting\\_started\\_with\\_TrackMate](https://imagej.net/Getting_started_with_TrackMate) and <https://imagej.net/TrackMate> )
- Then launch the tracker **MTrack2** (<http://fiji.sc/MTrack2> and <http://www.imagescience.org/meijering/software/mtrackj/> )
- Redo it with another threshold to remove or add noise.
- Which tracks to keep ?

To go beyond (Fiji is always evolving by plugin additions in labs : yourself soon :-)) :

- The **ToAST** plugin (Tool for Automated Sporozoite Tracking or more generally to adress the questions of motility directionality <http://fiji.sc/ToAST>) with image *Malaria Sporozoites*
- The **MOSAIC** plugin to efficiently track multiple targets.  
[http://courses.washington.edu/me333afe/ImageJ\\_tutorial.html](http://courses.washington.edu/me333afe/ImageJ_tutorial.html)  
<http://mosaic.mpi-cbg.de/ParticleTracker/> and [http://fiji.sc/Particle\\_Tracker](http://fiji.sc/Particle_Tracker)  
<http://mosaic.mpi-cbg.de/?q=downloads/imageJ>
- Explore the Python examples in the Repertory Python (check if cv2 is installed)  
*\$python mon\_programme.py [argument1]*  
Other programmes here also :  
<http://www.math-info.univ-paris5.fr/~lomn/Cours/CV/SeqVideo/Material/TPVideoPython/>