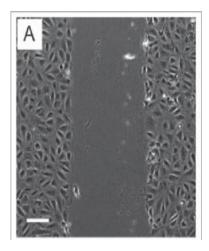
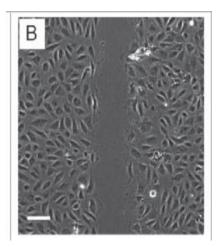
Cell Scratch Assay

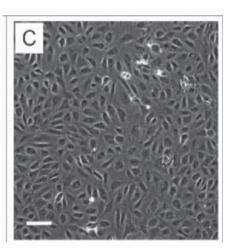
Goal: The goal of this project is to analyse the microscopic image of monlayer cells to find out the rate of cell migration/growth/proliferation using Python programming.

Tools: Python, images from already published papers

Procedures: 3 images were taken from an already published paper. The authors took a monolayer cell and introduced scratching there. Overtime, the cell started proliferating and migrating as such the wound was healed over time. Figure A, B, C were the original image extracted labelled acc to the time. [1]



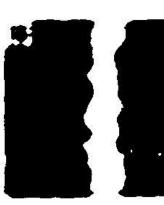




After obtaining the images, entropy filters and "otsu" segmentation was carried out using python to separate the area where cells are absent from the area where cells are still present. The images after segmentation are arranged serially without labelling.

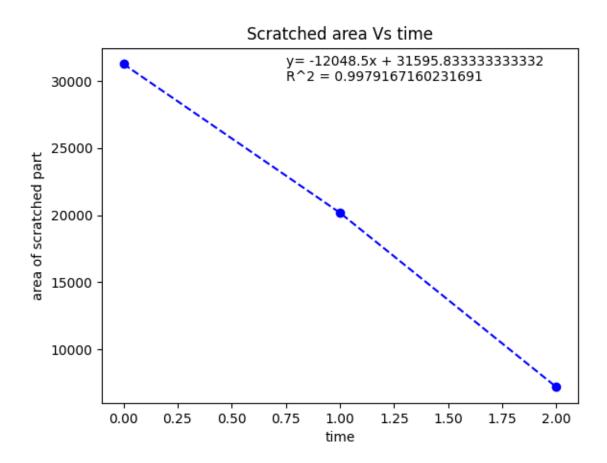








After completing the image processing, the space occupied by the cell-free area over time was analysed and a graph was plotted which showed that the area of cell-free space decreased over time. The slope and r^2 value are embedded with the graph.



Implications: Cell scratch assay is most widely used method to study the coordinated cell movement, as the assay is simple, inexpensive and well-established type of cell migration assay. The cell scratch assay is particularly suitable to study different kinds of cell interactions including the interactions of cells with extracellular matrix (ECM) and cell-cell interactions. This assay is applied to study wound healing and angiogenesis.

```
Python code: *IDE- Pycharm
import matplotlib.pyplot as plt
from skimage import io
from skimage.filters.rank import entropy
from skimage.morphology import disk
from skimage.filters import threshold_otsu
import numpy as np
import glob
#from scipy.stats import linregress
t = 0
t_list= []
area list = []
path = "images/*.*"
for images in glob.glob(path):
  img = io.imread(images, as_gray= True)
  entropy_img = entropy(img,disk(10))
  threshold= threshold_otsu(entropy_img)
  binary = entropy_img<=threshold</pre>
  t_list.append(t)
  t+=1
  area= np.sum(binary == True)
  area_list.append(area)
  print(t, area)
plt.plot(t_list, area_list,'bo--') #plotting the graph with t on x axis and area on y axis, go= green dots
plt.title('Scratched area Vs time')
plt.xlabel('time')
plt.ylabel('area of scratched part')
plt.text(0.75,30000,"y=-12048.5x+31595.8333333333332\nR^2=0.9979167160231691")
plt.show()
#print(linregress(t_list,area_list))
#slope, intercept, r_value, p_value, std_err = linregress(t_list, area_list)
#print("y=" + str(slope)+"x + "+str(intercept))
#print("R^2 value = " + str(pow(r_value, 2)))
```

References:

[1] Jonkman, J. E., Cathcart, J. A., Xu, F., Bartolini, M. E., Amon, J. E., Stevens, K. M., & Colarusso, P. (2014). An introduction to the wound healing assay using live-cell microscopy. *Cell adhesion & migration*, *8*(5), 440-451.