

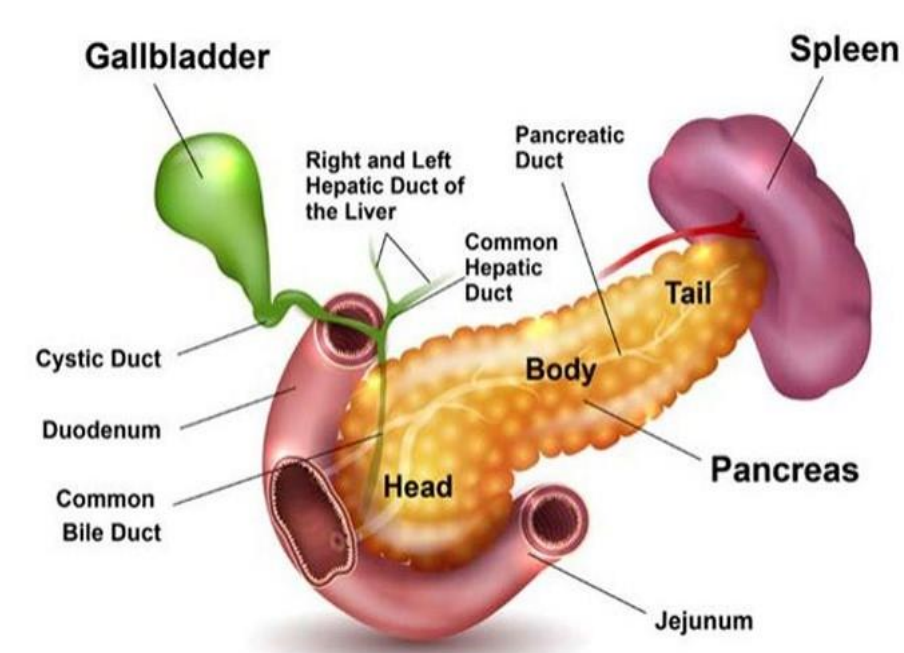
# The Effect of Nerve Growth Factor Beta (NGFβ) on Cell motility in Pancreatic Cancer Cells

Jamal Bonner<sup>1,2</sup> Steven Scahill<sup>2</sup>, Harry Gould, III, Kelly Jean Sherman, Dennis Paul  
 St. Augustine High School<sup>1</sup>  
 Louisiana State University Health Sciences Center<sup>2</sup>

## Introduction

Pancreatic cancer is now the fourth or fifth leading cause of cancer death in the Western world, with a 5-year recovery rate of 10% to 25% after the tumor is removed. Perineural invasion (PNI) that goes all the way to the extra pancreatic nerve bundle is a morphological trait of pancreatic cancer. It leads to retro pancreatic tumor extension, makes curative resection impossible, and affects local recurrence after the tumor has been removed. The fact that 84% to 100% of pancreatic cancer cells get into brain tissue shows how important this is. Nerve growth factor (NGF) is a beneficial protein that helps many types of nerve cells in the peripheral nervous system live and grow. Experiments in our lab have revealed that NGF has a stimulatory effect on tumor growth, invasion, and metastasis development. In this experiment we test the hypothesis that the neurotrophic factor NGFβ will increase the motility rate of Panc-1 cells in vitro. We will use a wound healing assay to measure the effect of NGFβ on Panc-1 cells, a highly metastatic cancer line. The wound healing assay is a complicated biochemical and cellular process that is needed to fix broken tissue. It includes dynamic interactions and crosstalk between different types of cells, interactions with molecules in the extracellular matrix, and the controlled release of soluble mediators and cytokines. Cells are seeded in a six-well plate and incubated overnight with 0.2568ng/ml NGFβ. Next a linear thin scratch “wound” is placed in a confluent cell monolayer, data acquisition through microscopic image capturing, gap measurement at three time points, T=0, T=6 and T=24.

## Background

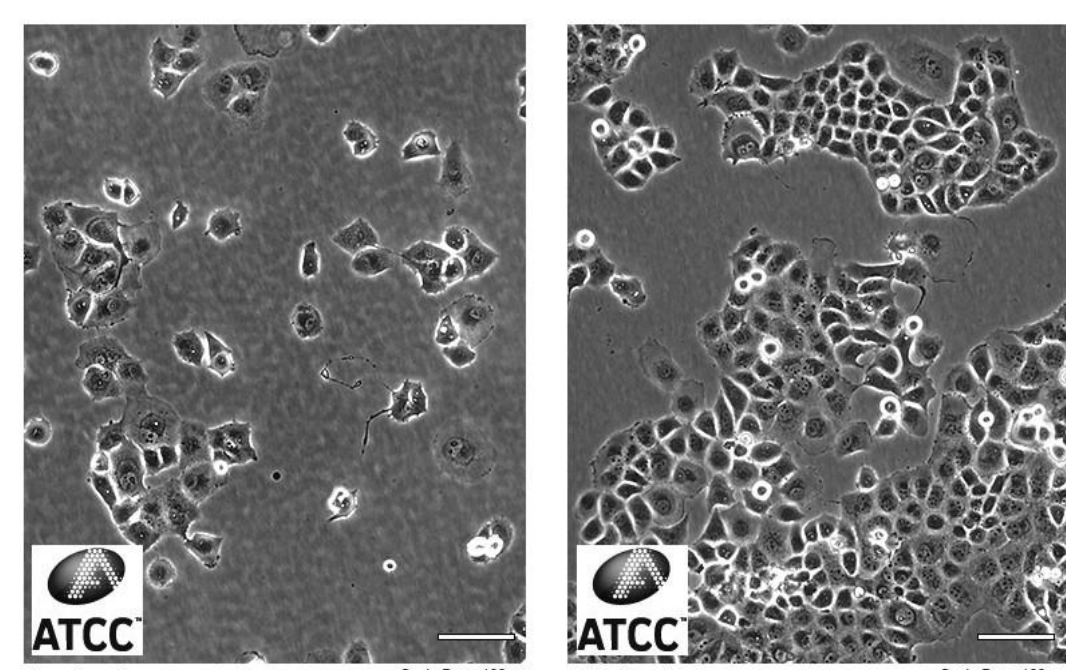


### Pancreas - functions

- maintain blood glucose levels
- produces digestive enzymes

### Pancreatic cancer – risk factors

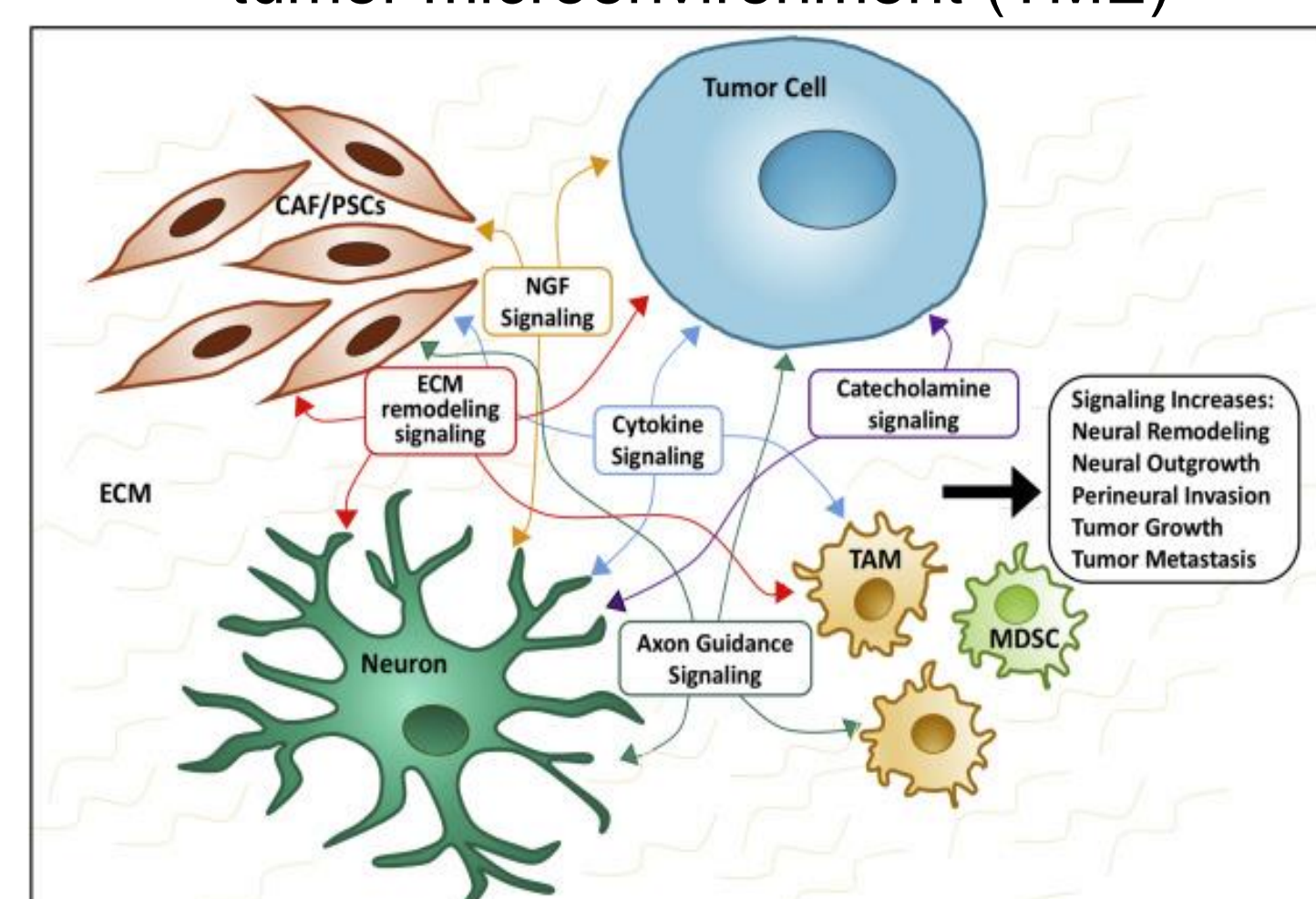
- chronic pancreatitis
- smoking
- diabetes



### Panc-1 cells

- human pancreatic cancer cell
- highly metastatic
- PANC-1 cells have a tendency to clump

### tumor microenvironment (TME)

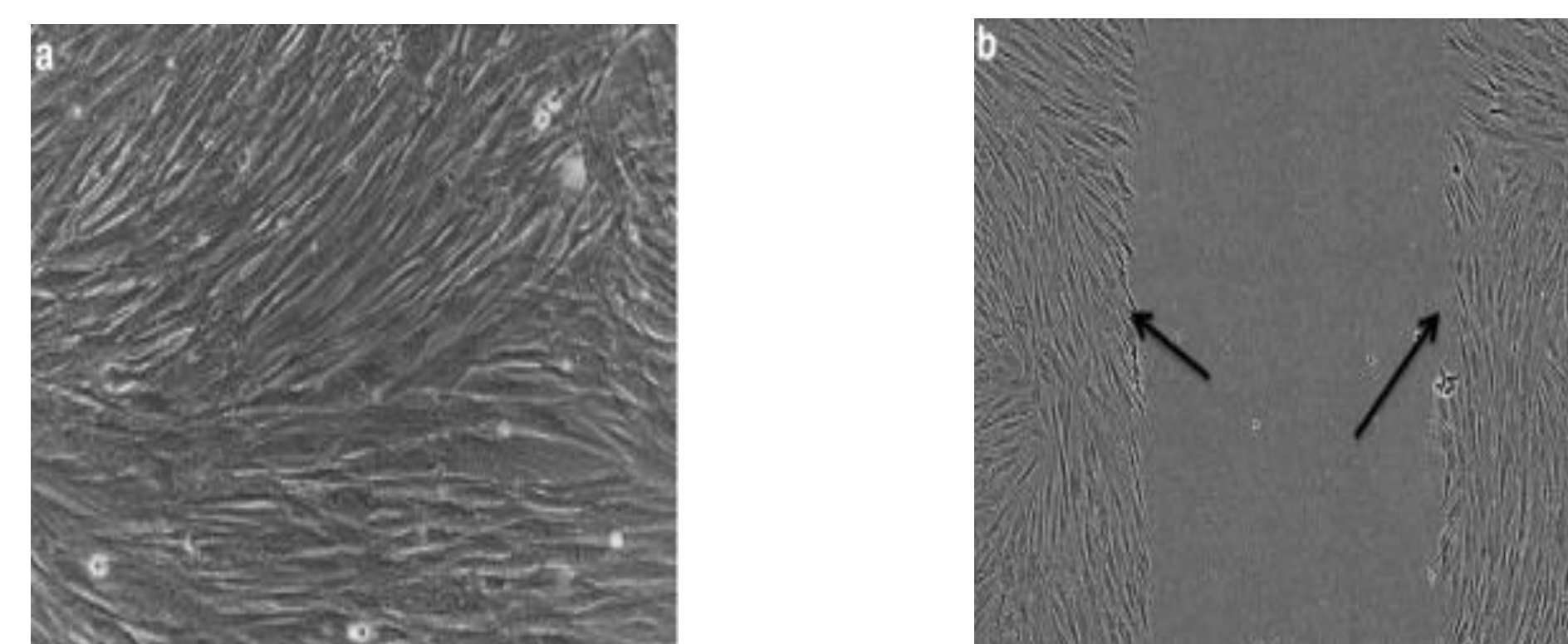


### NGF beta

- NGF beta
- Tumor microenvironment (TME)

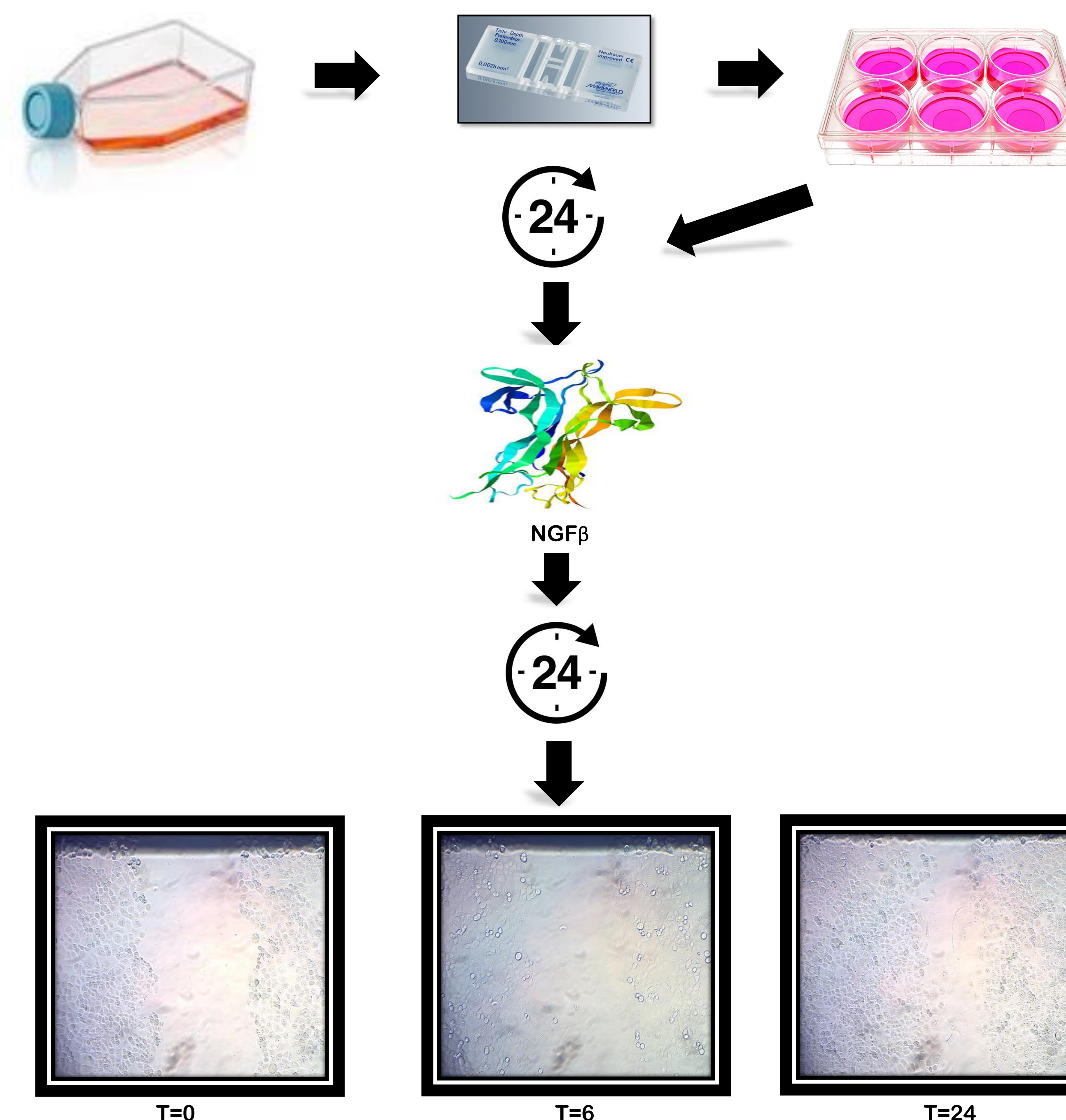
## Wound Healing Assay

- Motility assays in vitro are used to study the progression of cancer
- In this assay, we measured cell motility of cancer cells, panc-1.
- The technique involves basic steps applicable to almost all cell types: Cell seeding and preparation, making a linear thin scratch “wound” in a confluent cell monolayer, data acquisition through microscopic image capturing, gap measurement at each time point, and data analysis.



Research Techniques Made Simple: Analysis of Collective Cell Migration Using the Wound Healing Assay: Ayman Grada<sup>1</sup>, Marta Otero-Vinas<sup>1,2</sup>, Francisco Prieto-Castrillo<sup>2</sup>, Zaidal Obagi<sup>1</sup> and Vincent Falanga<sup>1</sup>

## Experimental Design



$$R_m = \frac{W_i - W_f}{t}$$

$R_m$  = Rate of cell migration ( $\mu\text{m}/\text{h}$ )  
 $W_i$  = Initial wound width ( $\mu\text{m}$ )  
 $W_f$  = final wound width  
 $t$  = duration of migration

## Results

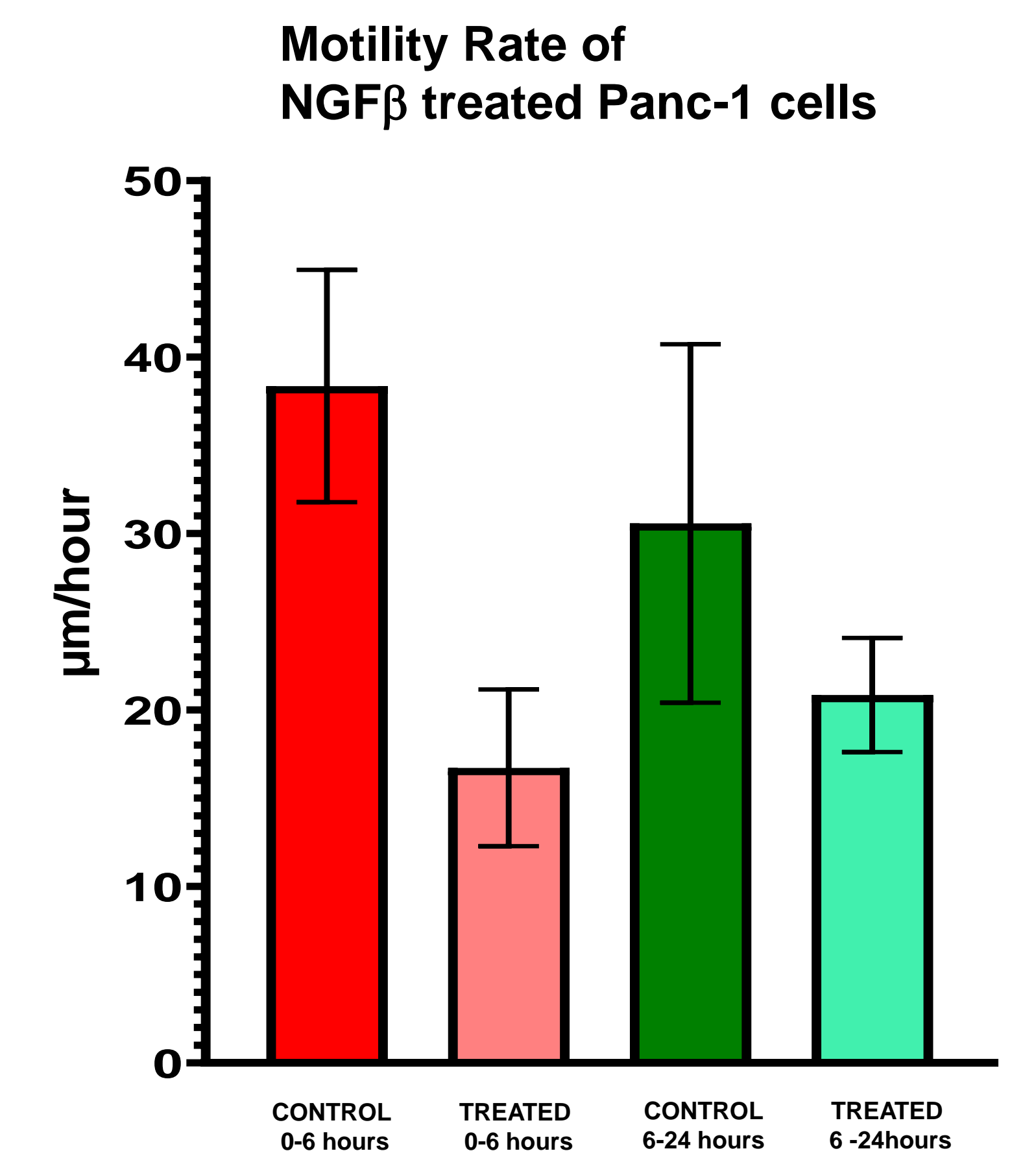


Figure 1. Shows motility rate of NGFβ [0.256 ng/ml] treated compared non-treated for 0-6 and 6-24 hours

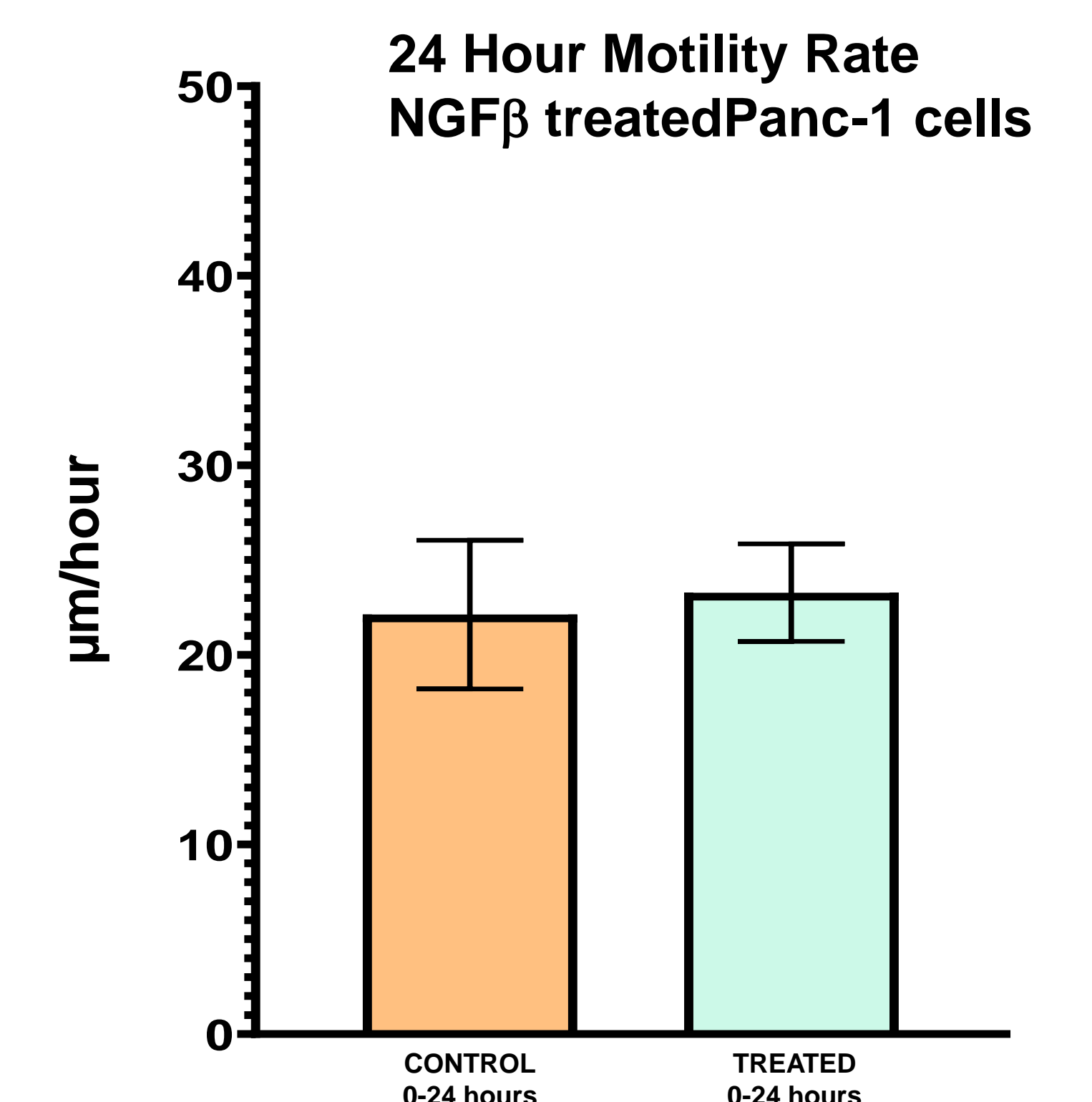


Figure 2. Shows motility rate of NGFβ [0.256 ng/ml] treated compared to non-treated for 24 hours

## Conclusion

Our results show no significant difference of NGFβ treated PANC-1 cells compared to untreated when all time points were compared. The Panc-1 cells, which were seeded at 1 million cells per well, were overly confluent and showed signs of clumping rather than motility, which we believed brought an error into the data. In further experiments we will edit either the time points in which we measure the motility of the cells, the concentration of cells we put into each well, or we will perform a cell curve test.

## Future Experiments

In future experiments we will determine the proper concentration between 1 million cells and 500,000 cells per well, determine which concentration is appropriate to see the cell motility, and use experiments such as the concentration effect curve.