



# Development and evaluation of a 2D-contraction-assay with a gelatine substrate for human dermal fibroblasts

Iris Hube, Linda Leicht, Larissa Walter

School of Applied Chemistry, Reutlingen University, Alteburgstraße 150, 72762 Reutlingen, Germany

## Motivation

### Background

Gelatine consists of several types of collagen. Is it therefore possible to replace collagen type I with gelatine as a substrate for cell-contraction assays? Cells, such like fibroblasts contract during their cell migration by tightening the filaments of their actin-cytoskeleton. This can be measured. [1]

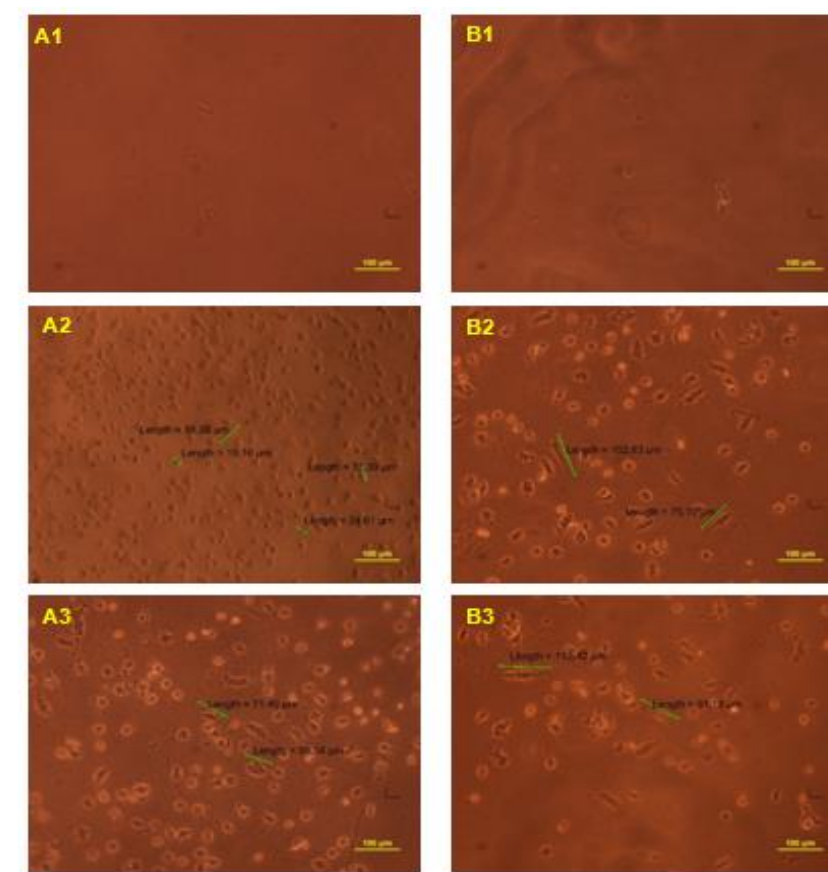
The advantage of gelatine compared with collagen type I are the lower costs. This allows a more extensive experimental set-up.

## References

[1] Peter Friedl, Katharina Wolf, „Tumor-cell invasion and migration: diversity and escape mechanisms“, Nature Reviews Cancer 3, 362-374 (2003)

## Procedure

### Cell behaviour



A1: 5 % gelatine-gel, negative control; A2: 5 % gelatine-gel, specimen, 10.06.2016; A3: 5 % gelatine-gel, specimen, 13.06.2016; B1: 7 % gelatine-gel, negative control; B2: 7 % gelatine-gel, specimen, 10.06.2016; B3: 7 % gelatine-gel, specimen, 13.06.2016

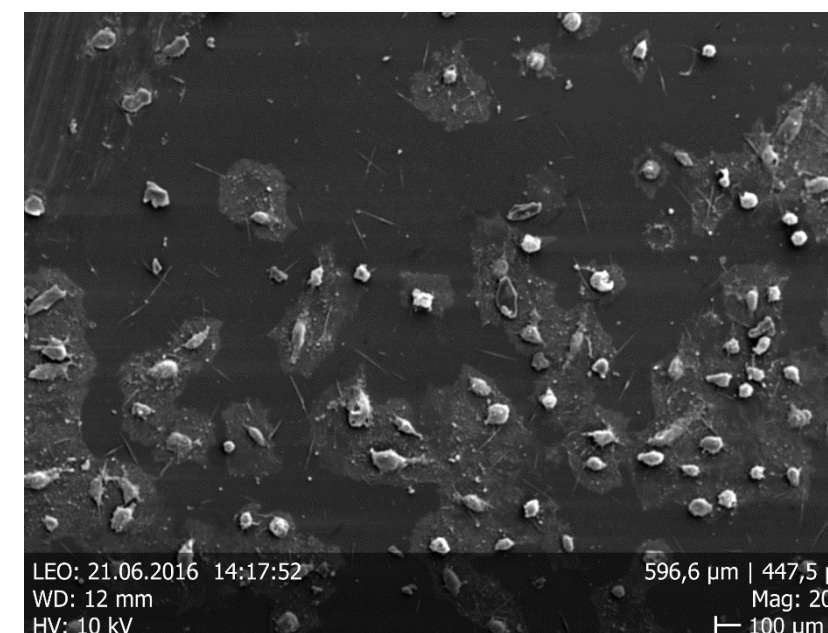
1\*10<sup>5</sup> cells were seeded on each gel.

Cells are incubated on the surface of the gelatine gel with culture media.

### REM-Images



LEO: 21.06.2016 14:03:15 59,66 µm | 447,5 µm  
WD: 12 mm Mag: 2000x  
HV: 10 kV | 10 µm



LEO: 21.06.2016 14:17:52 59,6 µm | 447,5 µm  
WD: 12 mm Mag: 2000x  
HV: 10 kV | 100 µm

7% gelatine gels

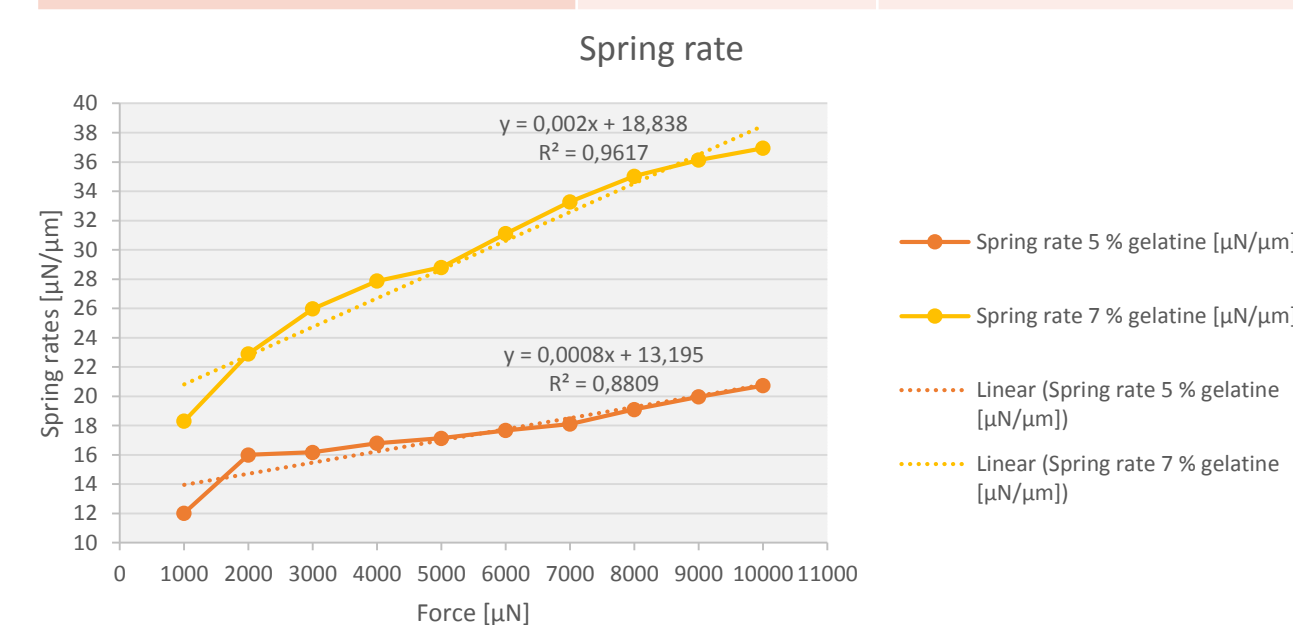
Images with REM were taken after dehydration with ethanol and critical point drying.

For a better conductivity the specimen were sputtered with gold particles.

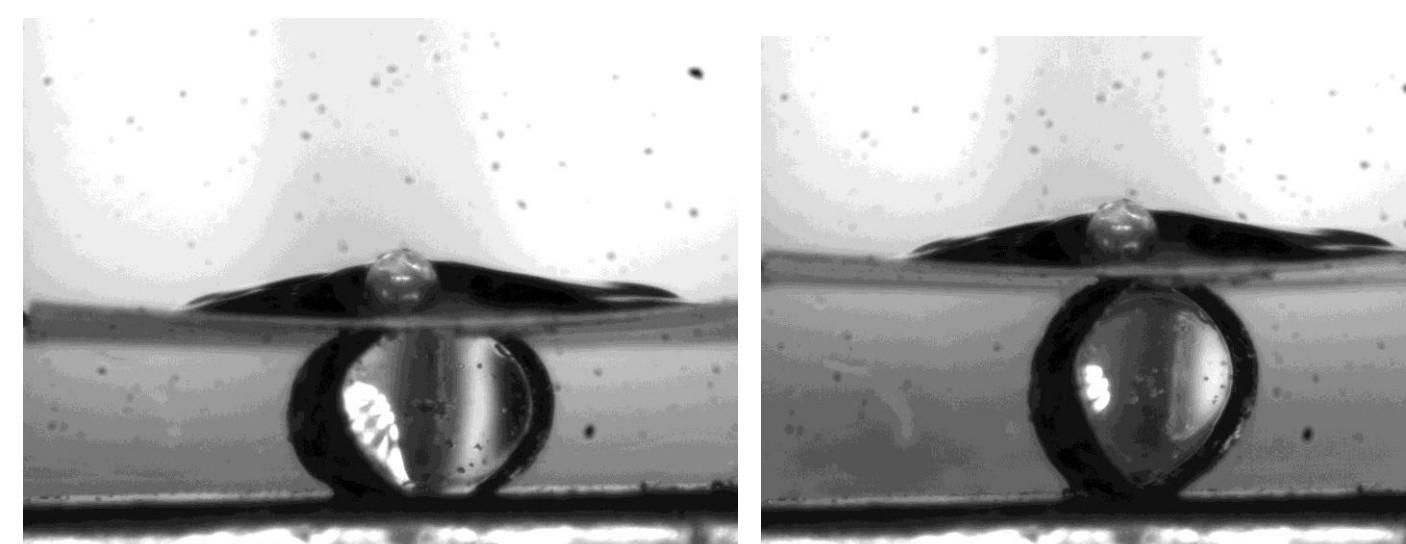
### Mechanical properties of the gelatine gel

- Spring rate
- Swelling behaviour
- Concentration and crosslinking

	Gelatine	Swelling Ratio [%]
Heat, 63 °C	5 %	35,3
	7 %	49,4
Saturated NaCl-solution	5 %	16,7
	7 %	17,4



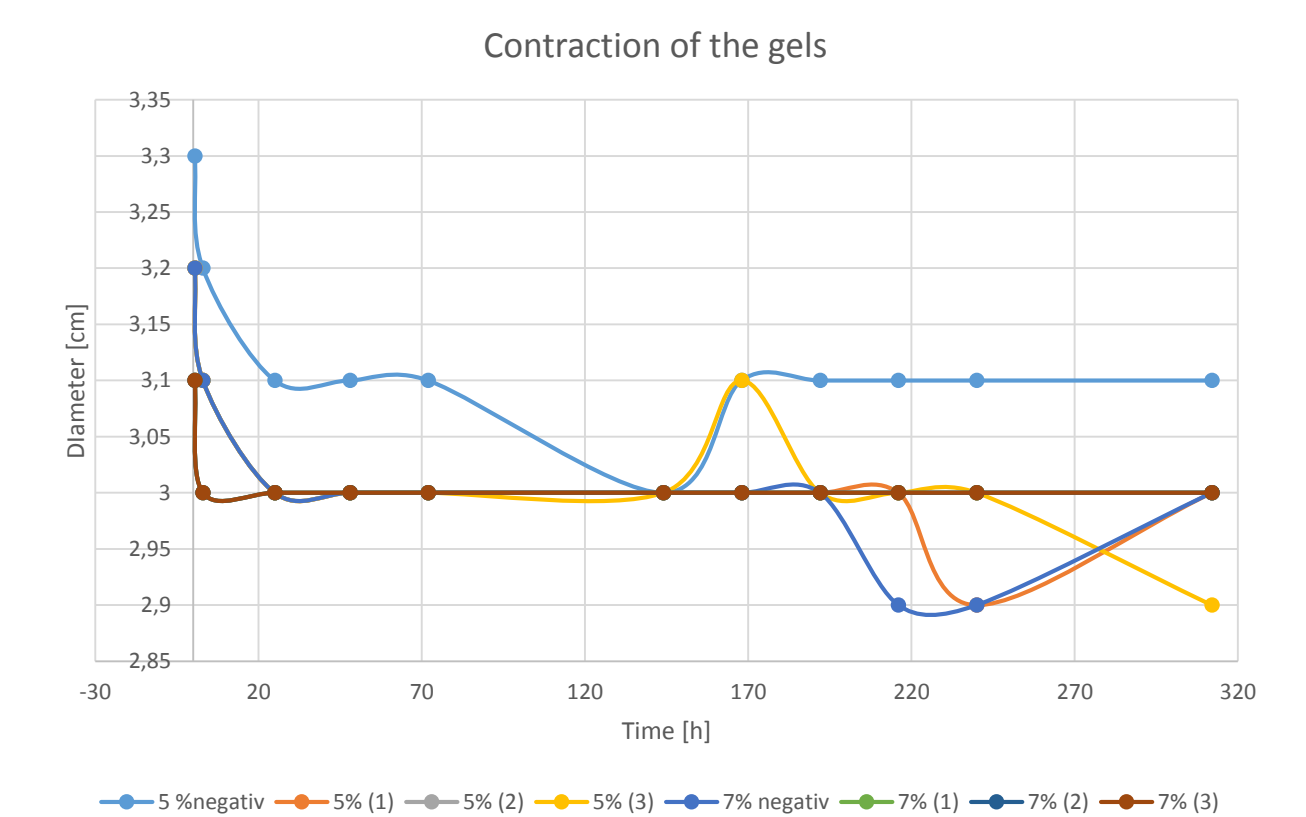
Pictures from Micro-Squisher  
5 % gelatine, squished (left) and relaxed (right)



## Results

### Contraction-assay

Cells were incubated on gels containing 5 % and 7 % gelatine. The contraction was measured from day 0 to day 13 (312 h).



### Conclusion

Pretests showed an ideal gelatine concentration of 5% and 7% for practical mechanical properties. The expansion of the cells on the hydrogel was successful. The cells were not able to contract the hydrogels as expected.