

# CELLULAR STUDIES ON PIEZOELECTRIC POLYVINYLIDENE FLUORIDE NANOFIBERS SUBJECTED TO ULTRASOUNDS STIMULATIONS

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## Introduction

In recent decades, there is an increasing interest in research related to development of the smart materials [1]. Such materials should response to external physical, chemical or mechanical stimuli and behave similar to natural body tissues. An example of smart materials are piezoelectric scaffolds, which can generate electrical signals in response to the applied stress [2]. Furthermore, they can stimulate the signalling pathways and thereby enhance the tissue regeneration at the impaired site. The piezoelectric scaffolds can act as sensitive mechano-electrical transduction systems. It is known that electrical charges are crucial for various activity of cells. The major advantage of piezoelectric scaffolds is that electrical potential can be generated non-invasively under the influence of mechanical field, without the need of using invasive electrodes [3,4].

## Materials and Methods

Polyvinylidene fluoride (PVDF,  $M_w = 400\,000$  g/mol) nanofibers were electrospun from 15% solution of dimethylformamide and acetone (DMF/Ac 4:1 weight ratio) at feed rate 0.2 mL/h (3 mm needle) and collected on drum collector (diameter 40 mm) at a distance between the needle and collector 180 mm:

- PVDF100 – collected at drum rotational speed 100 rpm and linear velocity 2 m/s resulted in random fiber distribution.
- PVDF1000 – collected at drum rotational speed 1000 rpm and linear velocity of 20 m/s resulted in aligned fiber distribution.

After the process, the samples were left for 24 hours for solvent evaporation.

Before *in vitro* studies, the samples were sterilized with UV light for 30 minutes. Fibroblasts L929 cells were cultured on the piezoelectric PVDF scaffolds.

Further, the samples with cultured cells were subjected to ultrasound stimulation for 30 minutes per day, for 7 days. Ultrasounds stimulus with power 20 mW, 80 mW and frequency 1.7 MHz were applied. As a control, piezoelectric PVDF scaffolds without ultrasonic stimulation were used (0 mW). In order to confirm the piezoelectric effect of the PVDF scaffolds on fibroblasts mitochondrial activity, MTT test was used.

The observations of fibers and cell morphology was conducted using Scanning Electron Microscopy (SEM). Results were statistically analyzed using OriginPro v.8.

## Results and Discussion

Scaffolds with random and aligned fibers orientation were produced.

Results of MTT test are presented in FIG. 1.

Mitochondrial activity of cells indicates the positive effect of piezoelectric phenomena on the cells under ultrasound stimulation.

This effect was similarly positive for low and high collector rotational speed. The observations using SEM verified the attachment and morphology of the cells.

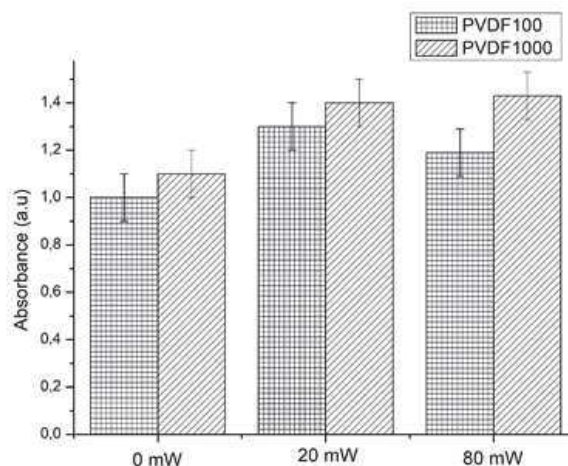


FIG. 1. Viability of fibroblasts L929 cultured on PVDF nonwovens and subjected to ultrasound stimulation (20 mW, 80 mW) after 7 day of cell culture.

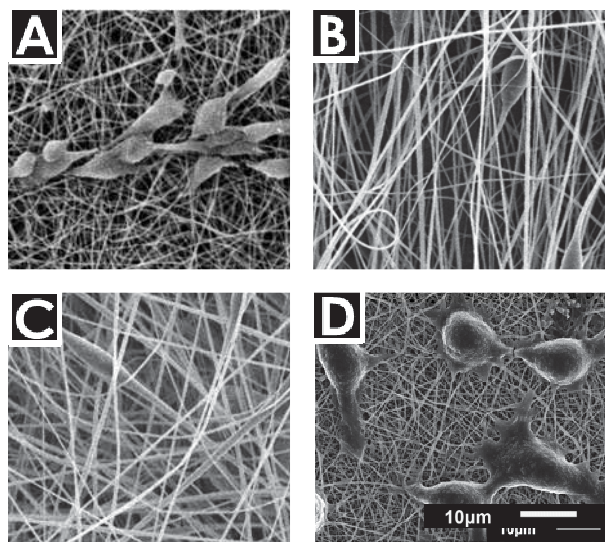


FIG. 2. SEM images of PVDF nanofibers: formed at different rotational speed of collector, 100 rpm (A) and 1000 rpm (B), with fibroblasts L929 (C, D) cell culture on day 7.

## Conclusions

PVDF nonwovens as piezoelectric polymer stimulated by ultrasounds is advantageous for cells activity. The obtained preliminary results are promising from the perspective of tissue engineering applications.

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## References

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