Electroactive Biomaterial Solutions for Tissue Engineering

Amy Gelmi[1,2]

Michael J. Higgins[2] , Gordon G. Wallace[2]

Edwin Jager[1] , Mehrdad Rafat[3]

[1] Biosensors and Bioelectronics, IFM, Linköping University , Linköping 58183, Sweden

[2] Australian Research Council Centre of Excellence for Electromaterials Science (ACES), Intelligent Polymer Research Institute (IPRI), AIIM Facility, Innovation Campus, University of Wollongong, NSW 2522, Australia

[3] Integrative Regenerative Medicine (IGEN) Centre, Department of Clinical and Experimental Medicine, Linköping University , Linköping 58185, Sweden

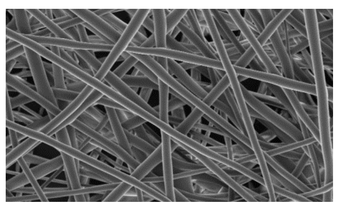
Tissue engineering is a field growing in prominence with advances for medical treatments and devices. The increasing need for treatments involving tissue grafting and organ transplantations is juxtaposed with a limited supply of organ donors. The ability to replace or recreate tissues and function in the body without the need for donor tissue is an area of research vital to improved medical treatment.

In order to control and direct tissue growth, we can use materials with specific properties to induce desired cellular responses. We can develop biomaterials that have multi-pronged approaches to cell control; electrical conductivity for electrical cues and stimulation, mechanical actuation for physical stimulation, biomolecular recognition, and morphological effects such as topographical guidance.

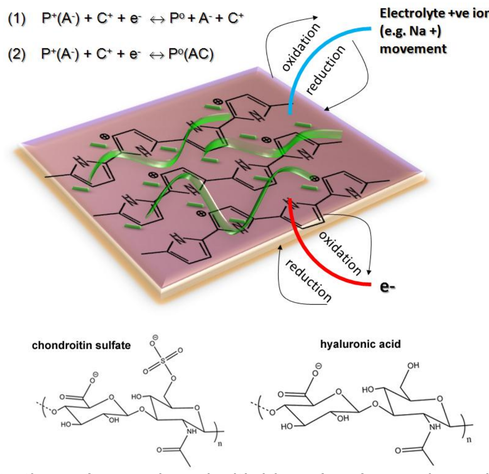
One class of electroactive biomaterials, Organic Conducting Polymers (OCPs), is generating significant interest due to their inherent compatiblity with biological systems of similar composition. The material is electrically conductive, mechanically actuatable, and its physical properties can be fine tuned during synthesis. OCPs, such as Polypyrrole (PPy), can also be doped during synthesis with biocompatible molecules to improve cellular response.

A Multi-pronged Approach

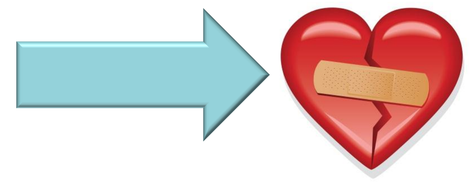
Combining biocompatible fibres as 3D scaffolds with an electroactive biomaterial.



Topographical 3D structure,
electrospun PLGA
Controllable porosity and fibre diameter.



Conductive polymer, doped with biomolecules such as chondroitin sulfate. Electrically conductive, mechanically actuatable, appropriate physical properties



The topographical, electrical, and mechanical stimulation are aimed to stimulate mesenchymal stem cells to differentiate into cardiomyocytes .
The ultimate goal is to replace or repair myocardial infarction scar tissue which impairs cardiac function.

Results

Biomolecular Recognition in Doped Conductive Polymers

Fibronectin was covalently bound to AFM probes, illustrated in Figure 1.

The probes were then used in force spectroscopy on both as-grown and electrically stimulated polypyrrole films.

The rupture lengths and forces were analysed to determine the 'corrected binding distance', i.e. where along the protein was the binding position (Figure 2).

Specific binding of FN to glycoaminoglycan and sulfonated dopants of polypyrrole occurs via heparin binding domains of the protein which corresponds to this length along the protein, as shown in Figure 3 (marked by the asterix).

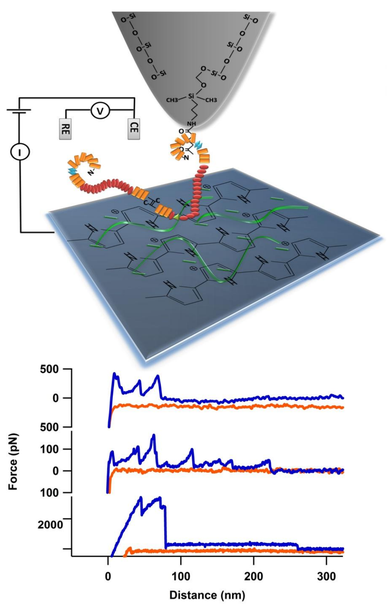


Figure 1: Modified tip-polymer interaction. The protein is bound at the N terminal to the cross-linking agent on the tip. Protein binding events; as the probe pulls the protein off the surface we can observe different events occuring in the force-distance feedback.

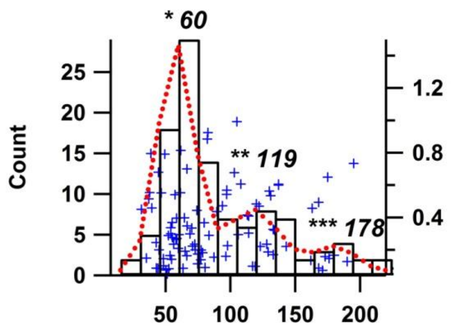


Figure 2: Histogram of protein length for Ppy/CS.

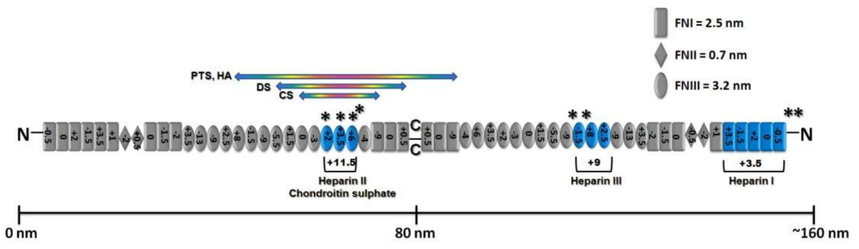


Figure 3: Fibronectin structure by domains, marked with individual domain charge. Domains of interest are marked in blue.

Stimulation of the polymer via cyclic voltammetry results in a reversible change in protein adhesion, as displayed in Figure 4. As the polymer is oxidised there is a significant increase in the total adhesion of FN, (current-voltage curve - red); (adhesion force – black).

This type of interaction is considered to be non-specific (Figure 5B). In contrast, the interaction with the non-stimulated polymers, as shown above in Figures 2 and 3, involves only a few bio-specific binding groups (Figure 5A) and forces an order of magnitude lower.

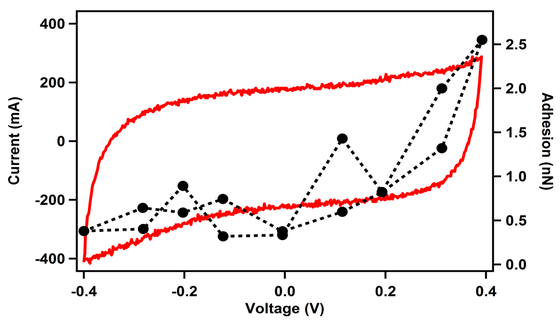


Figure 4: Cyclic voltammogram overlaid on average total adhesion force of FN on Ppy/CS. Total three scans performed with over 50 individual force curve measurements, over a period of 300 seconds.

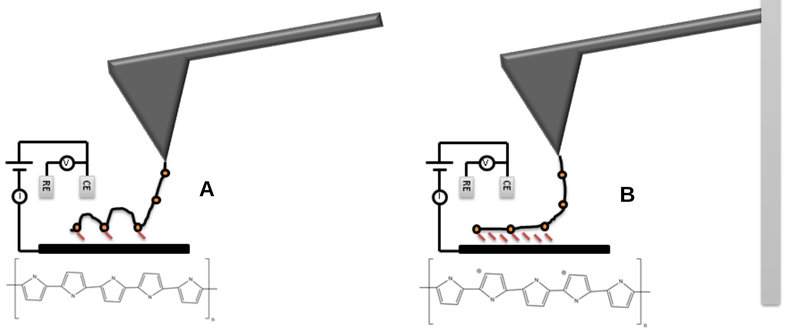
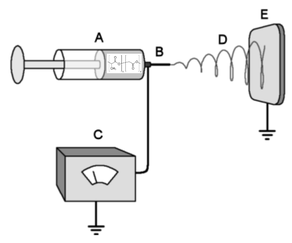
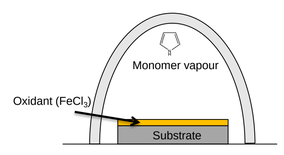


Figure 5: A) Specific interaction between the FN and polymer surface on charge-balanced PPy. B) Non-specific binding due to electrostatic forces across the entire PPy surface, leading to much higher adhesion forces.

Conductive Polymer Coating of PLGA Scaffolds



Poly(lactic-*co*-glycolic acid) (PLGA) was electrospun into fibrous mats.



VPP

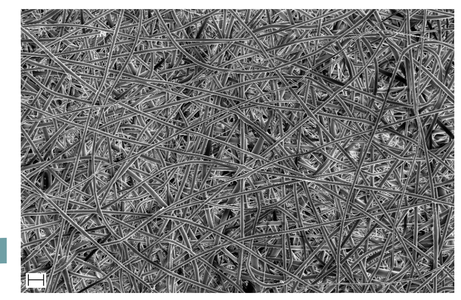


Figure 6: Bare PLGA 50:50 fibres. Scale bar is 10 µm.

PLGA provides the topographical scaffold but is non-conductive. The fibres are initially polymerised using vapour phase polymerisation.

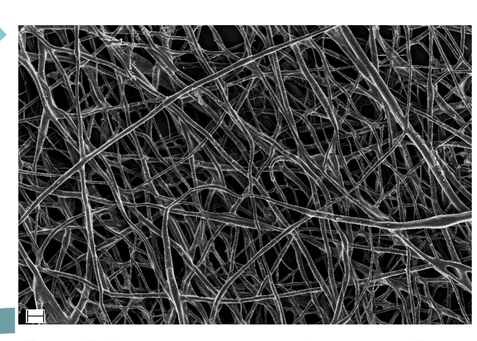
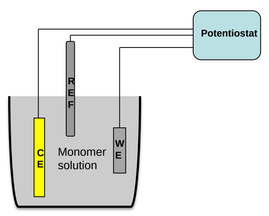


Figure 7: Vapour phase coated fibres, using Ppy and $FeCl_3$ as the oxidant. Scale bar is 10 µm.



ECP

Once they have a thin conductive layer, the fibres are coated with the biomolecularly doped PPy.

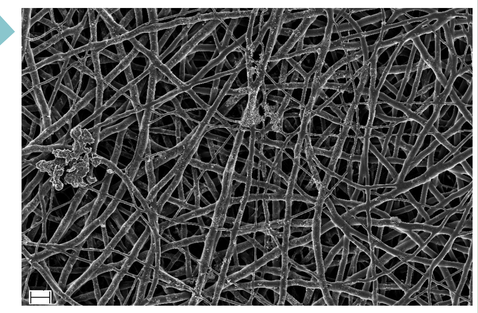


Figure 8: Electrochemically polymerised coated fibres, using Ppy/pTS. Scale bar is 10 µm.

Now we have electrocative, biocompatible 3D scaffolds.

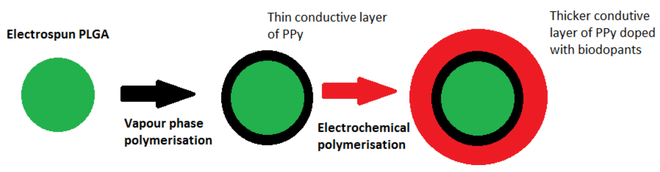
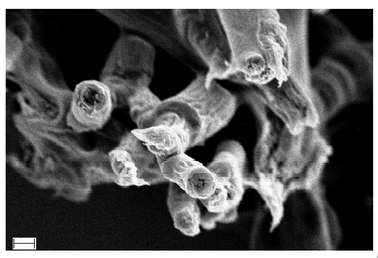


Figure 9: Polymer coating scheme. SEM of coated fibre cross section. Scale bar is 1 µm.



Future Work

The electroactive scaffolds are currently being tested using HUVEC and cardiovascular progenitor cells. The next step is to electrically stimulate the scaffolds, both *in situ* to measure mechanical actuation and *in vitro* to assess the influence on the living cells.

A. Gelmi, M. Higgins, G.G. Wallace, *Physical surface and electromechanical properties of doped polypyrrole biomaterials*, Biomaterials 31(8), 2010

A. Gelmi, M. J. Higgins, G. G. Wallace, *Attractive and Repulsive Interactions Originating from Lateral Nanometer Variations in Surface Charge/Energy of Hyaluronic Acid and Chondroitin Sulfate Doped Polypyrrole Observed using Atomic Force Microscopy,* Journal of Physical Chemistry B 2012, 116, 13498.

A. Gelmi, M. J. Higgins, G. G. Wallace, *Resolving Sub-Molecular Binding and Electrical Switching Mechanisms of Single Proteins at Electroactive Conducting Polymers,* Small 2013, 9, 393

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