Foreign Body Response Investigated with an Implanted Biosensor

Floyd B. Karp[1], Erasmo A. Lopez[2], Karl F. Böhringer[3], Buddy. D. Ratner[1]
Departments of [1]Bioengineering, [2]Mechanical Engineering, and [3]Electrical Engineering
University of Washington, Seattle, WA U.S.A.

Introduction:
One of the principal challenges for the long-term implantation of biosensors is that the normal physiological response of the body creates a fibrotic capsule of scar-like tissue surrounding the implanted device (the foreign body response). This growing collagenous capsule increasingly isolates the sensor device from its local environment, causing a time dependent degradation of the sensor signal. We utilize this change to the signal as an indicator of the physiological responses to the implantation of the device. We have chosen a micro-electrode array as the sensor device. We thus track the foreign body response electronically as the basis for this novel assay, which determines in comparable terms, an ongoing measurement of foreign body response (FBR) capsule character. This technique has the potential to become an important analytical method for comparing biomaterials and surface treatments aimed at reducing the FBR.

Materials and Methods:
We analyzed chronological electrical impedance spectroscopy (EIS) data to track changes of the electrical signal behavior over time between micro-electrode arrays which we utilized as the biosensor. We have performed experiments in three environments *in vitro*, *ex ova,* and *in vivo*. *In vitro,* we used a reservoir of phosphate buffered saline into which selected proteins were introduced that adsorb onto the electrode surface. Three proteins were studied, collagen, fibronectin and egg white.

We have investigated the FBR *ex ova* using the chick chorio-allantoic membrane (CAM) model. Previous work has verified that the CAM exhibits a response similar to the mammalian foreign body response. Eggs were incubated for 3.5 days and then opened and deposited into Petri dishes, where they were incubated for an additional three days prior to electrode implantation into the CAM. The FBR was then tracked by daily EIS measurements for as long as eleven days.

For our *in vivo* experimental environment, we implanted the biosensor electrode array into the temporalis muscle of mature Sprague Dawley rats. Mature rats form significantly more scar tissue following wounding than adolescent rats. The healing of the wound site from the traumatic implantation procedure was electronically monitored by EIS measurements for up to six weeks. Histological fibrous capsule measurements were correlated to the electrical data. In different experiments, we plasma-coated the biosensor electrodes with decorin protein and tetraglyme polymer developed in our lab. These coatings are designed to alter the FBR.

Results and Discussion:
In the *in vitro* model, each of the three proteins was found to affect the EIS results very differently. All of the proteins increased the real component of the complex impedance of the system. This *in vitro* result provided the necessary confidence in the biosensor and electronic measurement methods to proceed to the *ex ova* model investigation. In addition, the experimental *in vitro* data was mathematically analyzed to estimate the component values in a model electrical circuit.

When the biosensor micro-electrode array was implanted into the *ex ova* CAM model, it was observed that the electrical signal degrades with tissue growth during the healing and remodeling phase following the traumatic implantation. Histological examination of samples of membrane tissues with implanted probe-tips confirmed that the chick CAM wound healing processes adjacent to the implanted probe are similar to the mammalian FBR. The CAM tissues showed ectoderm hyperplasia and had monocyte activation with giant cell formation adjacent to the implanted probe-tip. Collagen rich extra cellular matrix was observed in 0.7-μm sections stained with 1-% toluidine blue.

These clear results provided confidence to proceed with implants into a live animal model. In addition, the experimental *ex ova* data was mathematically analyzed to estimate the component values in the same model electrical circuit as the *in vitro* data.

We performed intramuscular implantation of the biosensor and tracking of the EIS character in mature Sprage-Dawley rats. Both clean and tetraglyme polymer coated biosensors were implanted. Preliminary results (n = 5) show similar EIS chronological responses as the *ex ova* results. We report that the effects upon implantation in the rat wound healing model are a slow increase in the components of impedance.

Conclusions / Summary:
We have demonstrated that an implantable array of micro-electrodes can be utilized as a biosensor to distinguish the gradual changes in its environment caused by the growth of a foreign body capsule in both an *ex ova* CAM and *in vivo* rat wound healing models. Future work will quantify this biosensor data permitting further comparison to the physiological response.

The development of this novel type of biosensor provides a tool for assessing the long-term biocompatibility of various coatings and surface treatments. Problems common to many *in vivo* sensors could be addressed with this versatile new tool.