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Rosengren, Agneta; Wallman, Lars; Danielsen, Nils; Laurell, Thomas; Bjursten, Lars Magnus

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Tissue Reactions Evoked by Porous and Plane Surfaces Made Out of Silicon and Titanium

Agneta Rosengren, Lars Wallman*, Nils Danielsen, Thomas Laurell, and Lars Magnus Bjursten

***Abstract*—Square-shaped silicon or titanium implants with plane or porous surfaces surrounded by a rim of silicone were implanted in the rat abdominal wall for evaluation of the tissue response after one, six, or 12 weeks. Cell damage was identified as increased membrane permeability using fluorescence microscopy by injection of propidium iodide prior to the killing of the rats. Capsule thickness and immunohistochemical quantification of macrophages were used as a further measure of the foreign-body reaction. There were no significant differences in capsular cell densities for macrophages, total cells (macrophages, fibroblasts, and other cells), or necrotic cells at the different time points for the four surfaces studied. However, significant differences in the kinetics of the response were found between plane surfaces compared with porous ones. Both types of plane surfaces developed a significant increase in capsule thickness over time in contrast to the porous implants. Porous silicon displayed a significant decrease in total cells in the reactive capsule over time. Furthermore, porous silicon and titanium surfaces displayed a significant decrease in total cell numbers at the implant interface between six and 12 weeks. The present study demonstrated that implanted silicon elicited soft-tissue reactions comparable to that of titanium.**

***Index Terms*—Biocompatibility, capsule formation, macrophages, neural interface, sieve electrodes.**

I. Introduction

SEVERAL previous studies have demonstrated that peripheral axons have the capacity to regenerate through sieve electrodes implanted in nerves and that it is possible to record action potentials from such electrodes [1]–[15]. Usually, these sieve electrodes, which can be regarded as neural interfaces, have been made out of silicon but other materials such as polyimide have been used [13].

Implantation of a sieve electrode into a peripheral nerve trunk can be compared with implantation of any foreign material into other soft tissues, thus, in this aspect sieve electrodes are a biomaterial which in theory may be implanted for life. In previous studies, the main focus has been on achieving regeneration through sieve electrodes and to record nerve signals and not to study the tissue reactions evoked by the implanted material with a few exceptions [11]. This biomaterial aspect has to be considered since excessive fibrous encapsulation, which could hamper necessary electrochemical contact between axons and electrodes, has been suggested to be the most important reason for failure in the clinical use of neural interfaces [16]. Furthermore, little is known about soft-tissue reactions evoked by silicon, one of the most used materials in sieve electrode design. Therefore, this study addresses the cellular reactions in soft tissue induced by silicon compared with titanium (a well-established biomaterial), with either porous, or plane surfaces.

All implanted biomaterials induce an initial host response characterized by an acute inflammatory reaction resembling the normal wound healing process. This initial inflammatory reaction, evoked by the surgical trauma, appears to have a specific cellular reaction pattern, irrespective of the eliciting stimulus [17]. The inserted implant is then a persistent stimulus for the evoked inflammatory response [18]. This foreign-body response is characterized by an inner layer of macrophages and/or foreign-body giant cells with an outside secondary zone of layered fibroblasts and connective tissue [19]. For implanted materials in general, this response has been attributed to factors related to the implant or to the host tissue such as design of the implant, localization, physico-chemical surface properties including surface morphology, state of the host bed, surgical technique, and mechanical loading [17], [18], [20]–[25]. These factors are most probably also important for neural interfaces such as implanted sieve electrodes. It has been demonstrated that the surface topography affects the tissue reactions [26]–[29] and when the surface topography is modified it is conceivable that various factors are altered such as surface chemistry and surface energy [20] which in turn may affect both the molecular and cellular events at the surface. The surface topography seems to be the predominant factor for the induced tissue response when altering both material and topography [30], [31]. Textured, especially porous surfaces are preferable when compared with plane ones [32]–[34]. *In vitro* studies on cell culturing have shown that cell adherence and viability increased using porous silicon compared with planar silicon [35].

In the present study, capsule thickness and immunohistochemical quantification of macrophage subclasses [36]–[39] were used to measure the inflammatory response induced by the implanted silicon or titanium surfaces. Both materials will be oxidized naturally in air or in a biological environment. In addition, cell damage including increased membrane permeability was identified using flourescence microscopy after propidium iodide injection [40].

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A. Rosengren and L. M. Bjursten are with the Department of Experimental Research, University Hospital MAS, SE-205 02 Malmö, Sweden.

*L. Wallman is with the Department of Electrical Measurements, Lund University, Post Box 118, SE-221 00 Lund, Sweden (e-mail: lars.wallman@elmat.lth.se).

N. Danielsen is with the Department of Physiological Sciences, Neuroendocrine Cell Biology, Lund University, SE-221 84 Lund, Sweden.

T. Laurell is with the Department of Electrical Measurements, Lund University, SE-221 00 Lund, Sweden.

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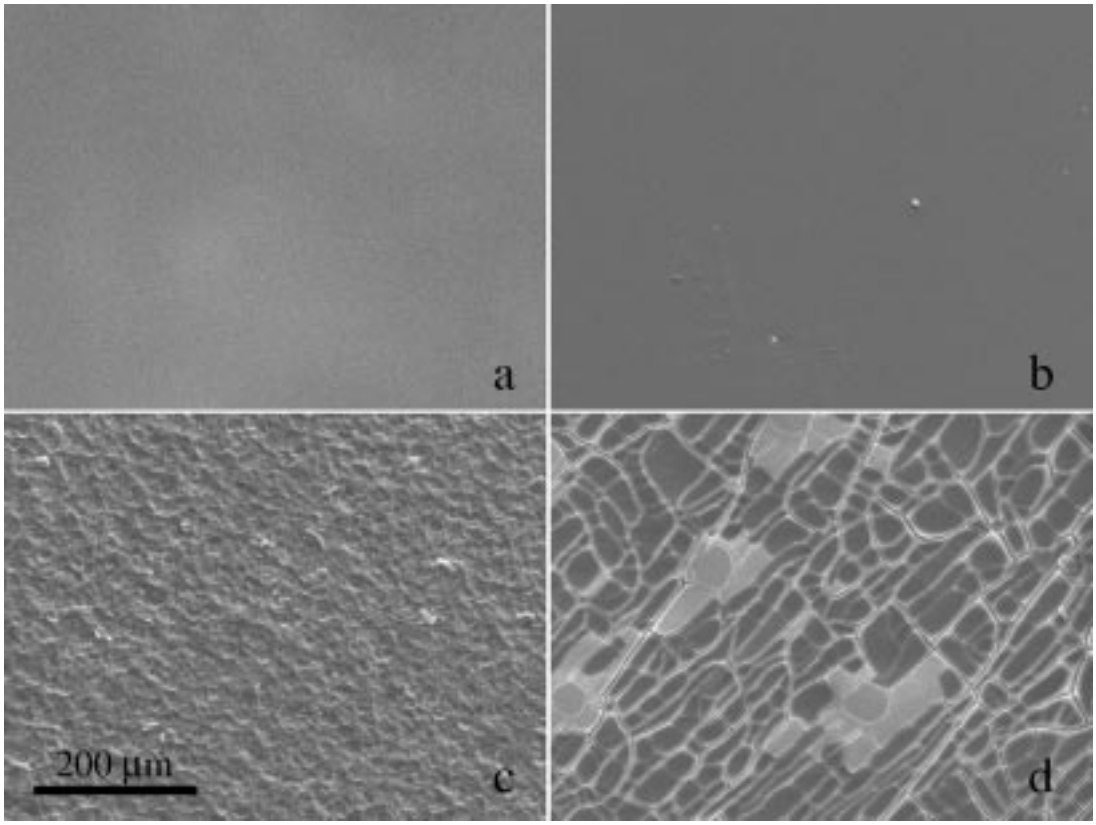

Fig. 1. SEM of the four different implanted surfaces. (a) Titanium with an electropolished plane surface. (b) silicon with a polished plane surface. (c) Titanium with an anodic oxidized porous surface. (d) Silicon with anodized porous surface. The light patches represent cracks induced by the drying process.

II. Material and Methods

A. Animals

Male Sprague-Dawley rats $(n = 24)$, weighing 200–250 g, fed on standard pellets and water ad libitum, were used. The rats were anesthetized by intraperitoneal injections of 1.0-ml/100–g body weight of a solution containing sodium pentobarbital (60 mg/ml) and NaCl (9 mg/ml) in 1:9 volume proportions. The experimental protocol was reviewed and approved by the Animal Ethics Committee in Lund, Sweden.

B. Implants

Implants of titanium and silicon with porous and plane surface structures were used. The surface morphology of the porous silicon was made to a similar degree as the porosity of the experimentally oxidized titanium. Fig. 1 shows SEM (Philips 515) of the different surfaces before implantation. The different implants were fabricated as described below.

1) *Planar Titanium:* The samples were electro polished in a solution of methanol (540 ml), n-butanol (350 ml) and percloric acid (60 ml) for 7 min and with a potential of 22.5 V at -30°C [Fig. 1(a)].
2) *Planar Silicon:* P-doped $(\approx 10\ \Omega\ \text{cm})\langle 111\rangle$ single sided polished silicon was used [Fig. 1(b)].
3) *Porous Titanium:* The titanium was electro oxidized by anodization in 1-M H_2SO_4 at room temperature with a potential of 80 V for 7 min creating a porous oxide layer [Fig. 1(c)].
4) *Porous Silicon:* P-doped $(\approx 10\ \Omega\ \text{cm})\langle 111\rangle$ oriented silicon was anodized in a HF/ethanol solution (equal volumes) at a current of 10 mA/cm^2 for 5 min [Fig. 1(d)].

The implants were cut into squares (4×4 mm, thickness 0.5 mm) and the edges were surrounded by silicone rubber avoiding undesirable tissue trauma from the corners. Before insertion the implants were cleaned and disinfected in 70% ethanol, rinsed and kept in sterile saline until surgery.

C. Implantation Procedure

We chose a well-established implantation model for soft-tissue reactions in order to achieve comparable results with previous studies. Details of this implantation procedure have been described previously [41]. In brief, the rectus abdominis muscle sheath was opened and the muscle moved laterally. All four implant types were placed in the same rat. Two implants were inserted on either side of the linea alba, outside the peritoneum without injuring the peritoneal membrane. The implants had their modified surfaces placed toward the muscle tissue. The rectus abdominis muscle was then slipped back to cover the implant and a suture was placed in the muscle sheath to secure the position of the implant. Eight animals for each implantation time point were used.

D. Tissue Fixation

After one, six, or 12 weeks, respectively, the animals were re-anesthetized and propidium iodide (PI) (0.1-ml/100-g body weight; 2.7-mg/ml saline; Sigma Chemical Co, St Louis, MO)

was injected intravenously. After 5 min, the implants with surrounding tissue were removed en bloc. The specimens were washed in ice-cold phosphate buffered saline (PBS pH 7.4), embedded in Tissue Tek O.C.T. compounds 4583 (Histolab Products AB, Göteborg, Sweden) and snap frozen for 30 s in 2-methylbutane at -70°C. The muscle tissue was sectioned in a cryostat (6 μm in thickness) and collected on chromium-alum treated slides and allowed to air dry. The slides were kept at -70°C until analyzed.

E. Immunohistochemistry

It has been possible to identify subpopulations of macrophages by the use of the monoclonal antibodies ED1 and ED2. The ED1 antibody identifies membrane markers associated primarily with newly recruited blood monocytes and the ED2 antibody identifies mature tissue macrophages [36], [37]. This staining procedures for ED1 and ED2 macrophages were done as described previously [38]. In brief, the staining procedure was as follows:

After removal of the endogenous peroxidase activity and blocking of unspecific bindings, the slides were incubated with primary antibodies. The primary antibodies used were mouse anti-rat ED1 and ED2 (Serotec Ltd., Oxford, U.K.). The primary antibodies were exposed to a biotinylated horse anti-mouse IgG secondary antibody. The sections were then incubated with Vectastain ABC peroxidase standard PK-4000 (Vector Lab. Inc., Burlingame, CA). The presence of peroxidase was detected using 3-Amino-9-Ethyl-Carbazole (Sigma Chemical Co, St. Louis, MO). Thereafter, the sections were counterstained in Mayers HTX. For the control sections, either the primary antibodies were excluded or mouse monoclonal antibodies directed to human cell surface antigens were used as primary antibody.

F. Image Processing

All histological images were obtained using a Kodak DSC-200 digital camera (Rochester, NY), mounted on a Nikon FXA (Tokyo, Japan) microscope using bright field or fluorescence microscopy. The images were retrieved and labeled using Photoshop 5.5 software (Adobe Photoshop, Mountain View, CA) on a Macintosh PowerPC computer (Cupertino, CA).

G. Morphometry

Sections from the four types of implant interfaces representing each material and each surface modification were evaluated for each implantation time. All sections were coded and evaluated by the same person (A. R). The numbers of cells were determined by manual counting of positive cells on sections stained for ED1 and ED2, respectively ($n = 8$ for each evaluated biological parameter at each time point). Furthermore, the numbers of cell nuclei (counterstained cells) were used as a measure of the total numbers of cells, which, thus, includes macrophages, fibroblasts and other cells. The quantification was done in a Nikon FXA microscope in bright field mode at $20\times$ magnification. A 10×10 ocular square grid where each square covered a 40×40 μm large area was superimposed at the center along the tissue border adjacent to the implant surface. The thickness of the reactive capsule was determined using the grid and was defined as the distance between the tissue border adjacent to the implant and the muscle border. The number of cells in the measured capsule was manually counted in five orthogonal rows of squares from the implant surface to the border of the muscle. The cell numbers are given in numbers per square millimeter (mm^2) based on the actually counted area. In order to quantify the tissue reactions immediate to the implants, the number of cells in the interfacial area was defined as the row of five squares along the implant surface. Within these squares the numbers of cells were counted as described for the capsule. Increased membrane permeability of the damaged cells was evaluated on sections that were mounted and directly examined for PI-stained nuclei using the FITC filter set of the fluorescence microscope under EPI-illumination. The procedure for the count of the PI-positive cells was similar to that for the interfacial ED1/ED2-positive cells with the exception of the squares counted. The cells were counted in the two rows of ten squares each, close to the interface.

H. Statistics

Nonparametric statistics was used in this study (Statview 4.5 for the Macintosh, Abacus Concepts, Berkeley, CA.). Material and surface differences were analyzed using the nonparametric Friedmans test. The Kruskal–Wallis test (K. W.) and the Kendall Rank correlation test (K. R.) were used for evaluating the effects over time. Since nonparametric statistics were used all values are presented as median values with indication of the 25 and 75 percentile (in the form of box plots).

III. Results

At all time points, irrespective of surface topography, the foreign-body reaction consisted of an inner cell rich zone where monocytes/macrophages with ED1 immunoreactivity predominated [Fig. 2(a)–(d)]. This inner cell rich zone also contained a few ED2 positive cells and other cells, most likely mainly fibroblasts (not shown). More distant from the implant surface the ED2 positive cells (not shown) were more abundant as compared with the ED1 cells. The PI-positive cells were mainly located close to the interfacial border and very rarely these cells were seen outside this area (not shown).

No significant difference in capsule thickness could be detected between the four different implants at each individual evaluation time (Fig. 3). Neither were there any significant differences in capsular cell densities for the ED1 positive (Fig. 4), ED2 positive (not shown), nor for the total cells (Fig. 5); or in the cell densities in the interfacial region (not shown except for total cells Fig. 6). There was, however, a significant difference in the kinetics of the response for the plane surfaces compared with the porous ones. Thus, the capsule thickness increased significantly over time for both plane silicon and plane titanium surfaces, whereas the porous surfaces had essentially the same thickness at the different time points (Fig. 3). The porous silicon and titanium surfaces further showed a significant decrease in total cell numbers at the interface (Fig. 6) between six and 12 weeks. In addition, the porous silicon implants also displayed

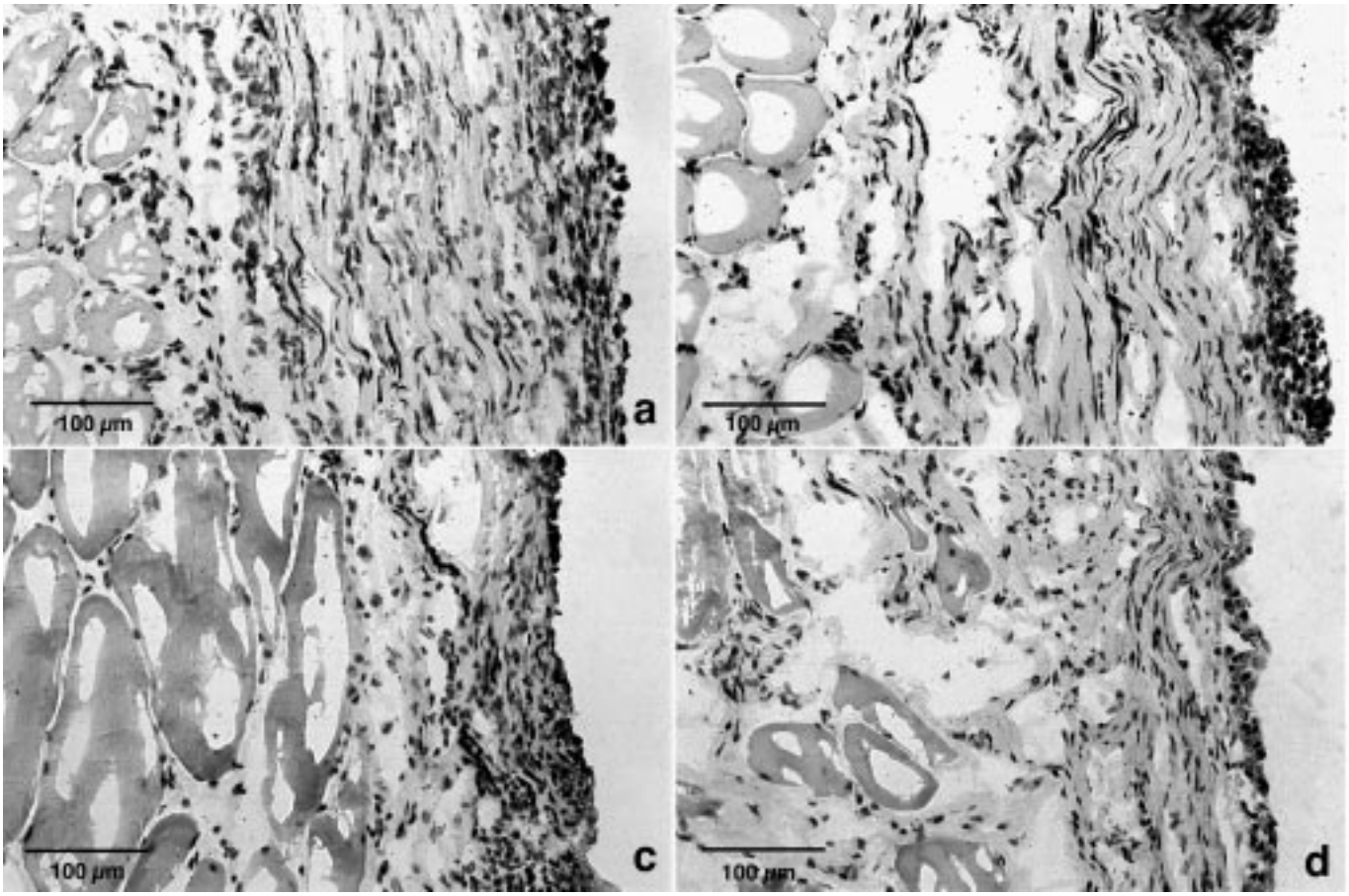

Fig. 2. Microphotographs of immunohistochemically stained tissue for ED1 positive macrophages adjacent to the different implants surfaces inserted in rat abdominal wall for 12 weeks. Note the capsule thickness. (a) Titanium with an electropolished plane surface. (b) Silicon with a polished plane surface, c) titanium with an anodic oxidized porous surface. (d) Silicon with anodized porous surface.

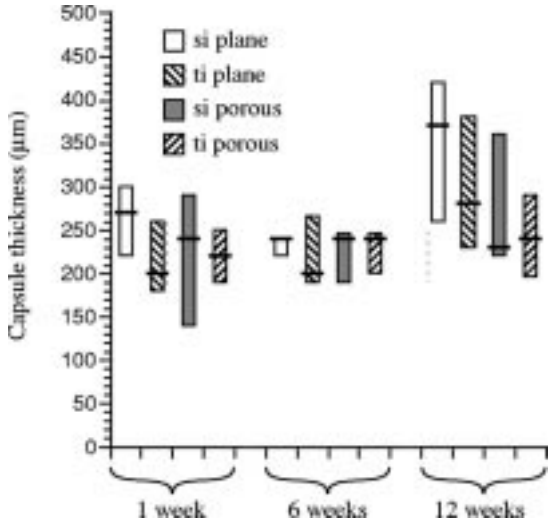

Fig. 3. Box plot of the capsule thickness of planar and porous silicon, respectively, titanium after one, six, or 12 weeks implantation in rat abdominal wall ($n = 8$ for each time point and implant type). The box represents the 25 and 75 percentiles and the median value is indicated by the horizontal line within the box. Capsule thickness increased significantly over time for both planar silicon and planar titanium (K. R. $p < 0.0098$; $p < 0.0272$).

a significant decrease in total cells at interface over time. At all surfaces in both the capsule (Fig. 4) and the interfacial area (not shown), a decrease in ED1 positive cells were seen between six and 12 weeks. At all time points, there were few PI-positive cells in the interfacial zone and no obvious difference could be revealed between the different implant types. At one week, all four implants were surrounded by larger numbers of PI positive cells as compared with later time points (not shown). However, this was only statistically significant for the porous silicon (K. W. $p < 0.0254$) and the porous titanium implants (K. W. $p < 0.0451$) which also displayed a significance over time (K. R. $p < 0.0310$).

A. Discussion

The aim of the present study was to evaluate the biocompatibility of silicon which is a well-known material for micromachining and it has been used as a neural interface, e.g., in a number of sieve electrode experiments. In this study, two different silicon surfaces that are possible candidates for neural interfaces were compared with the corresponding titanium surfaces. Thus, planar and porous silicon and titanium surfaces were chosen to correspond as closely as possible. Titanium was used as a reference material since it has a very well-documented biocompatibility both in hard and soft tissues [18], [42] and has also been used in soft-tissue animal models [39], [41], [43]. Implanted silicon-induced tissue reactions comparable to those observed for titanium. There were no significant differences in capsular cell densities for macrophages, total cells, or necrotic

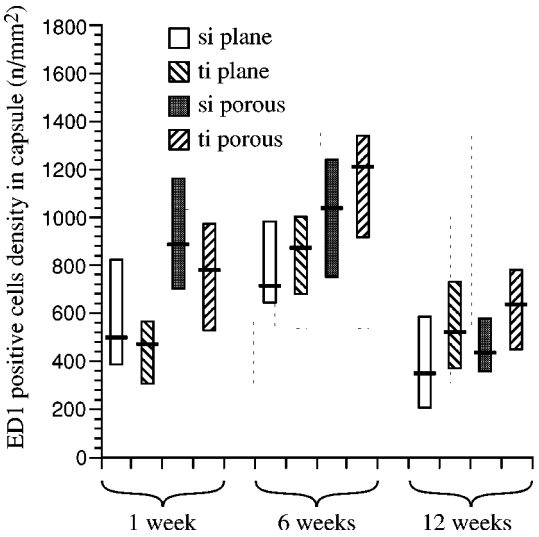

Fig. 4. Box plot of the ED1 positive cell density in the reactive capsule after one, six, or 12 weeks implantation in rat abdominal wall ($n = 8$ for each time point and implant type). The box represents the 25 and 75 percentiles and the median value is indicated by the horizontal line within the box.

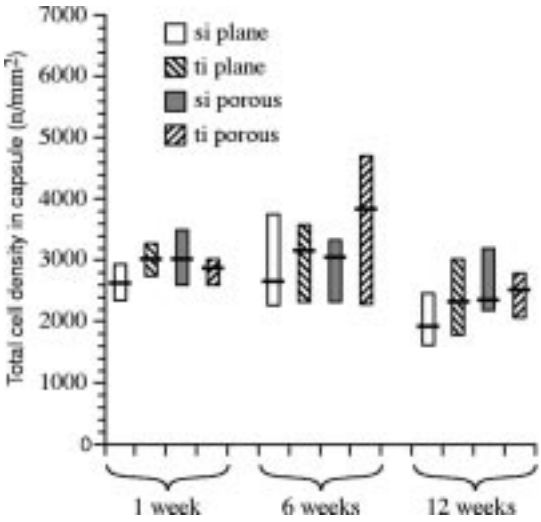

Fig. 5. Box plot of the total cell density (macrophages, fibroblasts, and other cells) in the reactive capsule after one, six, or 12 weeks implantation in rat abdominal wall ($n = 8$ for each time point and implant type). The box represents the 25 and 75 percentiles and the median value is indicated by the horizontal line within the box. Porous silicon implants showed a significant decrease in total cells over time (K. R. $p < 0.0031$).

cells between the four different implants at each individual time point. Significant differences were only seen over time (kinetics of the response) between the different surface morphologies. Porous silicon displayed a significant decrease in total cells in the reactive capsule over time and both porous surfaces also showed a significant decrease in total cell numbers at the implant interface between six and 12 weeks. Both porous surfaces displayed a constant capsule thickness over time, whereas the plane surfaces showed an increase in capsule thickness. This indicates that the surface morphology seems to be more important for the kinetics of the foreign-body reaction rather than the chemistry of the base material. In a chronic implant situation, such as a neural interface, findings indicating a stable

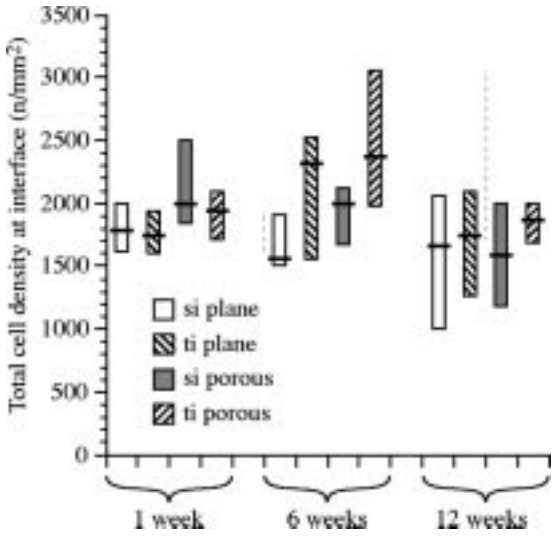

Fig. 6. Box plot of the total cell density (macrophages, fibroblasts, and other cells) at interface after one, six, or 12 weeks implantation in rat abdominal wall ($n = 8$ for each time point and implant type). The box represents the 25 and 75 percentiles and the median value is indicated by the horizontal line within the box. The porous and titanium surfaces showed a significant decrease in total cell numbers between six and 12 weeks (K. W. $p < 0.0359$; K. W. $p < 0.0223$).

capsule formation over time could be advantageous as compared with implants where the capsule increases. This is in accordance with previous studies showing that when both material and surface texture are varied, the surface texture appear to be the predominant factor relative to the induced tissue response [30], [31]. There are several reports indicating that structured and/or porous implant surfaces are favorable as compared with planar ones [29], [34], [40], [44]–[49]. It is conceivable that the different surface structures of the implants elicit different macrophage responses, which in turn may induce different tissue reactions by producing different secretion products such as: chemotactic agents for other cells, growth factors which stimulate the collagen production by the fibroblasts and neutral proteases which may affect the implant surface [50], [51] leading to different foreign-body formation including the thickness of the reactive capsule.

The over time capsule formation was reflected in the interfacial total cell counts. Thus, both types of porous implants had a significant decrease in the interfacial total cell numbers between six- and 12-week implantation periods, whereas the planar ones did not display any significant changes over time. However, the relationship between the cellular response and the capsule formation is still unclear. There are reports showing both a larger and a smaller number of inflammatory cells adjacent to porous implants as compared with planar ones. Our findings of a decreasing cellular density adjacent to the porous implants and thereby a lower number of inflammatory cells as compared with the planar ones are in concordance to some earlier studies [29], [30], [34], [39]. However, in some other studies, textured implants elicited a larger macrophage response as compared with smooth ones [51], [52].

The present study has demonstrated that porous and planar silicon implants elicited a foreign-body reaction with a magnitude comparable to that observed for corresponding titanium surfaces. Thus, in this aspect silicon could be considered to

be a useful biomaterial with an acceptable biocompatbility. This study may serve as a baseline for future biocompatibility studies regarding planar and porous silicon in more specialized applications such as implantation in the nervous system. In our opinion, it was necessary to first establish the foreign-body reaction evoked by silicon in a more general animal model and in comparison to a well-established biomaterial such as titanium. Further, this study also indicated that porous surfaces have a favorable kinetic response as compared with planar ones. It is not inconceivable that porous surfaces have a better initial implantation stability in the host bed (i.e., less micromovements between implant and the tissue caused by shear forces) and there are several reports supporting this theory [31], [49], [53]–[56]. The fact that fibroblasts has been shown to insert obliquely to textured implants, while they align parallel to almost planar implants [27], [28] is emphasizing this view. Further, it has been demonstrated that larger pore size is favorable in respect to neovascularization around implants and since it is known that inhibition of oxygen and nutrients to the cells adjacent to an implant would limit its possibilities for integration [49].

The question is how to extrapolate the findings from the present study into the design of neural interfaces in general and sieve electrode design in particular. As discussed above, initial implantation stability seems to be an important factor and, therefore, porous surfaces may have an advantage in long term applications. In the specific sieve electrode design, implant stability is usually achieved by enclosing the sieve electrode in a nerve regeneration chamber and securing the two nerve stumps the chamber wall [4], [6], [9], [11]. Unstable implant design relative to the ends of the transected nerve has been reported to induce damage to the nerve ends during the movements of the host [3], [55]. Even if the sieve electrode implant design is stable one could expect epineural and/or endoneural scarring within the nerve regeneration chamber. All peripheral nerve repair methods will induce such scarring including the silicone chamber model [57]. Peripheral nerves always heal with some fibrosis. Therefore, it is of importance to chose the material for the sieve electrode so it will evoke as little tissue reactions as possible. It should not jeopardize the purpose of the sieve electrode, i.e., to record nerve signals over a long time period. Any sieve electrode material that will elicit epineural and/or endoneural scarring to an unwanted degree could hamper the necessary electrochemical coupling between axons and electrode. In this aspect, porous silicon is a theoretically favorable alternative or any other structured material. Furthermore, by increasing the actual area of the recording electrode, by for example making it porous (e.g., porous silicon) the signal coupling increases [58]. However, there are reports indicating that porous silicon is degraded in a biological environment [59]. We, therefore, examined our tissue samples for traces of titanium or silicon using energy dispersed X-ray (EDX) microanalysis LINK ISIS connected to a Philips 515 SEM. The only findings were elements normal for biological tissue such as carbon and sodium. If, despite of these findings, porous silicon is found to corrode in a biological environment there are methods to stabilize it [59].

In conclusion, silicon evoked tissue reactions comparable to that of titanium, a well-established biomaterial.

REFERENCES

[1] A. Mannard, R. B. Stein, and D. Charles, "Regeneration electrode units: Implants for recording from single peripheral nerve fibers in freely moving animals," *Science*, vol. 183, pp. 547–549, 1974.

[2] G. E. Loeb, M. J. Bak, and J. Duysens, "Long-term unit recording from somatosensory neurons in the spinal ganglia of the freely walking cat," *Science*, vol. 16, pp. 1192–1194, 1977.

[3] D. J. Edell, "A peripheral nerve information transducer for amputees: Long term multichannel recordings from rabit peripheral nerves," *IEEE Trans. Biomed. Eng.*, vol. BME-33, pp. 203–213, 1986.

[4] J. Rosen, M. Grosser, and V. Hentz, "Preliminary experiments in nerve regeneration through laser-drilled holes in slicon chips," *Restor. Neurol. Neurosci.*, vol. 2, pp. 89–102, 1990.

[5] R. M. Bradley, R. H. Smoke, T. Akin, and K. Najafi, "Functional regeneration of glossopharyngeal nerve through micromachined sieve electrode arrays," *Brain Res.*, vol. 594, pp. 84–90, 1992.

[6] G. T. A. Kovacs, C. W. Storment, and J. M. Rosen, "Regeneration microelectrode array for peripheral nerve recording and stimulation," *IEEE Trans. Biomed. Eng.*, vol. 39, pp. 893–902, Sept. 1992.

[7] T. Akin, K. Najafi, R. H. Smoke, and R. M. Bradley, "A micromachined silicon sieve electrode for nerve regeneration applications," *IEEE Trans. Biomed. Eng.*, vol. 41, pp. 305–313, Apr. 1994.

[8] G. T. A. Kovacs, C. W. Storment, M. Halks-Miller, J. C. R. Belczynski, C. C. Della Santina, E. R. Lewis, and N. I. Maluf, "Silicon-substrate microelectrode arrays for parallel recording of neural activity in peripheral and cranial nerves," *IEEE Trans. Biomed. Eng.*, vol. 41, pp. 567–577, June 1994.

[9] X. Navarro, S. Calvet, M. Butí, N. Gómez, E. Cabruja, P. Garrido, R. Villa, and E. Valderama, "Peripheral nerve regeneration through microelectrode arrays based on silicon technology," *Restor. Neurol. Neurosci.*, vol. 9, pp. 151–160, 1996.

[10] R. M. Bradley, X. Cao, T. Akin, and K. Najafi, "Long term chronic recordings from peripheral sensory fibers using a sieve electrode array," *J. Neurosci. Meth.*, vol. 73, pp. 177–186, 1997.

[11] Q. Zhao, J. Drott, T. Laurell, L. Wallman, K. Lindström, L. M. Bjursten, G. Lundborg, L. Montelius, and N. Danielsen, "Rat sciatic nerve regeneration through a micromachined silicon chip," *Biomaterials*, vol. 18, pp. 75–80, 1997.

[12] P. Dario, P. Garzella, M. Toro, S. Micera, M. Alavi, J. U. Meyer, E. Valderrama, L. Sebastiani, B. Ghelarducci, and P. Pastacaldi, "A microfabricated interface for neural recording and stimulation," *J. Micromech. Microeng.*, vol. 7, pp. 233–236, 1997.

[13] X. Navarro, S. Calvet, F. J. Rodríguez, T. Stieglitz, C. Blau, M. Butí, E. Valderrama, and J.-U. Meyer, "Stimulation and recording from regenerated peripheral nerves through polyimide sieve electrodes," *J. Peripheral Nervous Syst.*, vol. 3, pp. 91–101, 1998.

[14] P. Heiduschka and S. Thanos, "Implantable bioelectric interfaces for lost nerve functions," *Prog. Neurobiol.*, vol. 55, pp. 433–461, 1998.

[15] L. Wallman, A. Levinsson, J. Schouenborg, H. Holmberg, L. Montelius, N. Danielsen, and T. Laurell, "Perforated silicon nerve chips with doped registration electrodes: *In vitro* performance and *in vivo* operation," *IEEE Trans. Biomed. Eng.*, vol. 46, pp. 1065–1073, Sept. 1999.

[16] P. J. Rousche and R. A. Norrman, "Chronic recording capability of the Utah intracortical electrode array in cat sensory cortex," *J. Neurosci. Meth.*, vol. 82, pp. 1–15, 1998.

[17] P. Thomsen and L. E. Ericson, "Inflammatory cell response to bone implant surfaces," in *The Bone-Biomaterial Interface*, J. E. Davies, Ed. Toronto, Canada: Univ. Toronto Press, 1991, pp. 153–163.

[18] P.-I Brånemark, J. Lausmaa, L. E. Ericson, P. Thomsen, R. Brånemark, and R. Skalak, "Anatomy of osseointegration and the transfer of load," in *Reconstructive Preprosthetic Oral and Maxillofacial Surgery*, R. J. Foneseca and W. H. Davis, Eds. Philadelphia, PA: Saunders, 1995, pp. 165–224.

[19] J. M. Anderson, "Inflammatory response to implants," *Trans Amer. Soc. Artif. Intern. Organs*, vol. 34, pp. 101–107, 1988.

[20] B. D. Ratner, "Biomaterial surfaces," *J. Biomed. Mater. Res.; Appl.. Biomater.*, vol. 21, pp. 59–90, 1987.

[21] T. Albrektsson, P. I. Brånemark, H. A. Hansson, and J. Lindström, "Osseointegrated implants requirements for ensuring a long-lasting, direct bone-to-implant anchorage in man," *Acta. Orthop. Scand.*, vol. 52, pp. 55–170, 1981.

[22] B. Kasemo and J. Lausmaa, "Biomaterial and implant surfaces: On the role of cleanliness, contamination, and preparation procedures," *J. Biomed. Mater. Res.*, vol. 22, pp. 145–158, 1988.

[23] R. Baier and A. Meyer, "Future directions in surface preparation of dental implant," *J. Dent. Educ.*, vol. 52, pp. 788–791, 1988.

[24] D. C. Smith, R. M. Pilliar, and R. Chernecky, "Dental implant materials 1. Ssome effects of preparative procedures on surface topography," *J. Biomed. Mater. Res.*, vol. 25, pp. 1045–1068, 1991.
[25] D. Muster, B. Demri, and M. H. Ali, "Physicochemical characterization of surface and interface on biomaterials and coatings," in *Encyclopedic Handbook of Biomaterials and Bioengineering Part A: Materials*, D. L. Wise, D. J. Trantolo, D. E. Altobelli, M. J. Yaszemski, J. D. Gresser, and E. R. Schwartz, Eds. New York: Marcel Dekker, 1995, vol. 1, pp. 785–812.
[26] D. M. Brunette, B. Chehroudi, and T. R. L. Gould, "Electron microscopic observations on the effects of surface topography on the behavior of cells attached to percutaneous and subcutaneous implants," in *Tissue Integration in Oral, Orthophedic and Maxillofacial Reconstruction Proceedings of the Second International Congress on Tissue Intergration in Oral, Orthopedic, and Maxillofacial Reconstruction*, W. R. Laney and D. E. Tolman, Eds, IL: Quintessence, 1990, pp. 21–31.
[27] B. Chehroudi, T. R. L. Gould, and D. M. Brunette, "Titanium-coated micromachined grooves of different dimensions affect epithelial and connective-tissue cells differently *in vivo*," *J. Biomed. Mater. Res.*, vol. 24, pp. 1203–1219, 1990.
[28] ——, "A light and electron microscopic study of the effects of surface topography on the behavior of cells attached to titanium-coated percutaneous implants," *J. Biomed. Mater. Res.*, vol. 25, pp. 387–405, 1991.
[29] A. Rosengren, N. Danielsen, H. Persson, M. Kober, and L. M. Bjursten, "Tissue reactions to polyethylene implants with different surface topography," *J. Mater. Sci. Mater. Med.*, vol. 10, pp. 75–82, 1999.
[30] C. A. Behling and M. Spector, "Quantitative term implants of selected polymers," *J Biomed Mater Res*, vol. 20, pp. 653–666, 1986.
[31] Y. C. Paquaya, J. E. Ruijter, J. P. van der Waerden, and J. A. Jansen, "Tissue reaction to dacron velor and titanium fiber mesh used for anchorage of percutaneous devices," *Biomaterials*, vol. 17, pp. 1251–1256, 1996.
[32] G. J. Picha and R. F. Drake, "Pillar-surface microstructure and soft tissue implants: Effect of implant site and fixation," *J. Biomed. Mater. Res.*, vol. 30, pp. 305–312, 1996.
[33] L. Hakelius and L. Ohlsén, "A clinical comparison of the tendency to capsular contracture between smooth and textured gel-filled silicone mammary implants," *Plast. Reconstr. Surg.*, vol. 90, pp. 247–254, 1992.
[34] M. Batra, S. Bernard, and G. Picha, "Histologic comparison of breast implant shells with smooth, foam, and pillar microstructuring in a rat model from 1 to 6 months," *Plast. Reconstr. Surg.*, vol. 95, pp. 354–363, 1995.
[35] S. C. Bayliss, L. D. Buckberry, P. J. Harris, and M. Tobin, "Nature of the silicon–animal cell interface," *J. Porous Mater.*, vol. 7, pp. 191–195, 2000.
[36] C. D. Dijkstra, E. A. Döpp, P. Joling, and G. Kraal, "The heterogeneity of mononuclear phagocytes in lymphoid organs: Distinct macrophage subpopulations in the rat recognized by monoclonal antibodies ED1, ED2 and ED3," *Immunology*, vol. 54, pp. 589–599, 1985.
[37] C. D. Dijkstra, E. Van Vliet, E. A. Döpp, A. van der Lelij, and G. Kraal, "Marginal zone macrophages identified by a monoclonal antibody: Characterization of immuno- and enzyme-histochemical properties and functional capacities," *Immunology*, vol. 55, pp. 23–30, 1985.
[38] A. Rosengren, L. M. Bjursten, and N. Danielsen, "Inflammatory reaction dependence on implant localization in rat soft tissue models," *Biomaterials*, vol. 18, pp. 979–987, 1997.
[39] A. Rosengren, N. Danielsen, and L. M. Bjursten, "Analysis of the inflammatory response to titanium and PTFE implants in soft tissue by macrophage phenotype quantification," *J. Mater. Sci. Mater. Med.*, vol. 9, pp. 415–420, 1998.
[40] A. Rosengren, N. Danielsen, and L. M. Bjursten, "Reactive capsule formation around soft-tissue implants is related to cell necrosis," *J. Biomed. Mater. Res.*, vol. 46, pp. 458–464, 1999.
[41] P. Thomsen, L. M. Bjursten, and L. E. Ericson, "Implants in the abdominal wall of the rat," *Scand. J. Plast. Reconstr. Surg.*, vol. 20, pp. 173–182, 1986.
[42] K. M. Holgers, P. Thomsen, A. Tjellström, and L. E. Ericson, "Electron-microscopic observations on the soft tissue around clinical long-term percutaneous titanium implants," *Biomaterials*, vol. 16, pp. 83–90, 1995.
[43] A. Rosengren, B. R. Johansson, P. Thomsen, and L. E. Ericson, "A method for immunolocalization of extracellular proteins in association with the implant-soft tissue interface," *Biomaterials*, vol. 15, pp. 17–24, 1994.
[44] Y.-Z. Zhang, L. M. Bjursten, C. Freij-Larsson, M. Kober, and B. Wesslén, "Tissue response to commercial silicone and polyurethane elastomers after different sterilization procedures," *Biomaterials*, vol. 17, pp. 2265–2272, 1996.
[45] R. M. Brohim, P. A. Foresman, P. K. Hildebrandt, and G. T. Rodeheaver, "Early tissue reaction to textured breast implant surfaces," *Ann. Plast. Surg.*, vol. 28, pp. 354–362, 1992.
[46] G. J. Picha, R. F. Drake, and K. Mayhan, "Soft tissue response to ion textured surfaces," *Artif. Organs*, vol. 14, pp. 32–37, 1990.
[47] S. R. Taylor and D. F. Gibbons, "Effect of surface texture on the soft tissue response to polymer implants," *J. Biomed. Mater. Res.*, vol. 17, pp. 205–227, 1983.
[48] J. Smahel, P. Hurwitz, and N. Hurwitz, "Soft tissue response to textured silicone implants in an animal experiment," *Plast. Reconstr. Surg.*, vol. 92, pp. 474–479, 1993.
[49] J. H. Brauker, V. E. Carr-Brendel, L. A. Martinson, J. Crudele, W. D. Johnston, and R. C. Johnson, "Neovascularization of synthetic membranes directed by membrane microarchitecture," *J. Biomed. Mater. Res.*, vol. 29, pp. 1517–1524, 1995.
[50] J. M. Jacob-LaBarre, M. Assouline, T. Byrt, and M. McDonald, "Synthetic scleral reinforcement materials: I. Development and *in vivo* tissue biocompatibility response," *J. Biomed. Mater. Res.*, vol. 28, pp. 699–712, 1994.
[51] C.-S. Zhu, K. Ohsaki, K. Li, Q. Ye, Y. H. Tran, Y. Ohba, and K. Moriyama, "Long-term observation of subcutaneous tissue reaction to synthetic auditory ossicle (Bioceram) in rats," *J. Med. Invest.*, vol. 46, pp. 97–103, 1999.
[52] J. A. Hunt, P. J. Doherty, D. Annis, and D. F. Williams, "Evaluation of soft tissue response to a poly(urethane urea)," *Biomaterials*, vol. 13, pp. 651–656, 1992.
[53] J. D. Andrade, "Interfacial phenomena and biomaterials," *Med. Instrum.*, vol. 7, pp. 110–120, 1973.
[54] A. F. von Recum, "New aspects of biocompability: Motion at the interface," in *Clinical Implant Materials, Advances in Biomaterials*, G. Heimke, U. Soltész, and A. J. C. Lee, Eds. Amsterdam, The Netherlands: Elsevier Science, 1990, vol. 9, pp. 297–302.
[55] D. Green, L. Pruitt, and C. S. Maas, "Biomechanical effects of e-PTFE implant structure on soft tissue implantation stability: A study in the porcine model," *Laryngoscope*, vol. 107, pp. 957–962, 1997.
[56] C. S. Maas, T. Eriksson, T. McCalmont, D. Mabry, D. Cooke, and R. Schindler, "Evaluation of expanded polytetrafluoroethylene as a soft-tissue filling substance: An analysis of design-related implant behavior using the porcine skin model," *Plast. Reconstr. Surg.*, vol. 101, pp. 1307–14, 1998.
[57] N. Danielsen, "Regeneration of the rat sciatic nerve in the silicone chamber model," *Restor. Neurol. Neurosci.*, vol. 1, pp. 253–259, 1990.
[58] L. Wallman, "Silicon neural interfaces—Design and biomedical aspects," Ph.D.dissertation, Dept. Elect. Meas., Lund Univ., Lund, Sweden, 2001.
[59] L. Canham, M. Stewart, J. Buriak, C. Reeves, M. Anderson, E. Squire, P. Allcock, and P. Snow, "Derivatized porous silicon mirrors: Implantable optical components with slow resorbability," *Phys. Stat. Sol. (a)*, vol. 182, pp. 521–525, 2000.

Agneta Rosengren received the Licentiate degree in anatomy and cell biology from Institute of Anatomy and Cell Biology, Göteborg University, Göteborg, Sweden, in 1995 and the Ph.D. degree from Department of Experimental Research, Lund University, Lund, Sweden, in 1997.

Her Present research investigates soft-tissue reactions to implanted biomaterials, focusing the mechanisms in foreign-body induction.

Lars Wallman was born in Vallentuna, Sweden, in 1971. He received the M.S. degree in electrical engineering and the Ph.D. degree from Department of Electrical Measurements at Lund Institute of Technology, Lund, Sweden in 1994 and 2001, respectively

His current research focuses on micromachined neural interfaces for neural prosthesis control and microcomponents for liquid sampling handling in chemical microsystems.

Nils Danielsen was born in Nora, Sweden, in 1956. He received the M.D. degree from the Medical Faculty, Göteborg University, Göteborg, Sweden, in 1982 . He received the Ph.D. degree from the Department of Anatomy, Göteborg University in 1986.

He was a Post-doctoral fellow at the Department of Biology, University of California, San Diego between 1986 and 1987. After his return to Sweden, he has held research positions at the Department of Anatomy, Göteborg University and at the Department of Hand Surgery, Malmö. Since 1995, he is an Associate Professor with the Department of Physiological Sciences, Lund University, Lund, Sweden. His current research interests are in peripheral nerve regeneration, neuropeptides, biomaterials and *in vivo* recordings from peripheral nerve fibers. He has been the scientific and financial coordinator of the Artificial Hand Project since year 2000.

Thomas Laurell received the M.S. degree in electrical engineering and the Ph.D. degree from the Department of Electrical Measurements, Lund Institute of Technology, Lund University in 1986 and 1995, respectively.

He became an Associate Professor of Electrical Measurements in 1998 and was appointed Professor of Medical and Chemical Microsensors at the Department of Electrical Measurements, Lund Institute of Technology in 2000. He currently leads the microstructure technology group at the Department of Electrical Measurements. His research focuses on microbiomedical and Lab-On-A-Chip technologies, e.g., microstructured components for biomedical monitoring/analysis, implantable neural electrodes, and microcomponents for liquid/cell sample handling in chemical microsystems, i.e., dispensing, injecting, pumping, and sampling of picoliter volumes. In the past years, a strong focus has been is put on the development of new microsystem technology for proteomics research.

Lars Magnus Bjursten received the certificate as physician in 1975 and the Ph.D. degree in clinical immunology from Göteborg University, Göteborg, Sweden, in 1987.

In 1989, he became Specialist in Internal Medicine and was Professor of Bioimplant Research at Lund University, Lund, Sweden. He has, apart from his clinical activities, been closely associated with the research connected with titanium as an implant material. Since 2000, he has held an appointment as Adjunct Professor in the Department of Bioengineering, University of California, San Diego. His current research is focused on determining which design parameters in biomaterials have the most significance for the biological host response when used as implants. The main focus of this research is on mechanical factors at the interface between materials and tissue as well as the microstructure of the biomaterial.

In 1987, Dr. Bjursten was board certified specialist in clinical immunology and transplantation immunology