

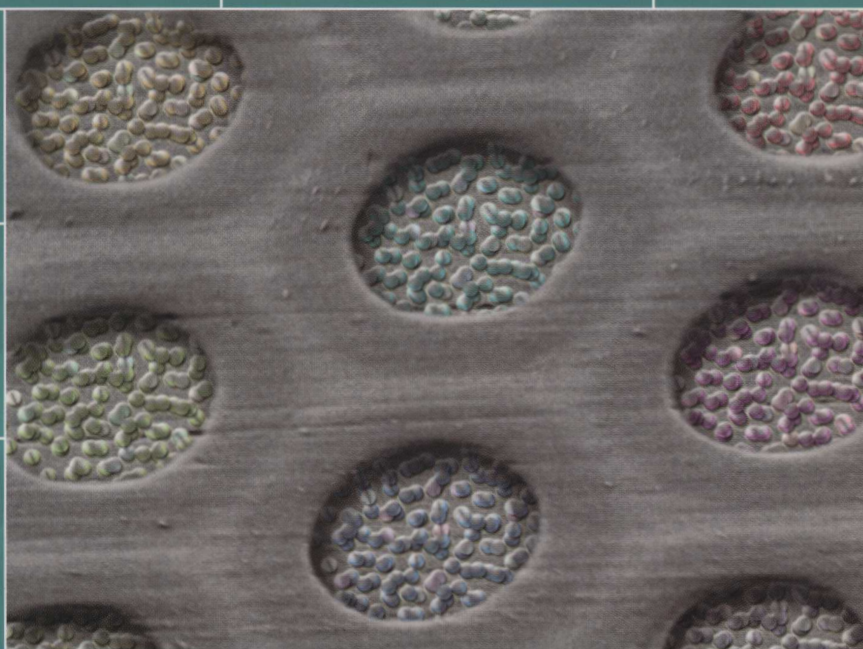
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The cover image is adapted from Figure 1 of the review by David R Walt 'Imaging optical sensor arrays' (pp 689–695). It shows a scanning electron micrograph of an optical imaging fibre array. The coloured objects have been electronically added as a representation of individual cells, or bead-based sensors – two materials that may be used in these arrays.

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Implantable chemical sensors for real-time clinical monitoring: progress and challenges

Megan C Frost and Mark E Meyerhoff*

Recently, progress has been made in the development of implantable chemical sensors capable of real-time monitoring of clinically important species such as PO_2 , PCO_2 , pH, glucose and lactate. The need for developing truly biocompatible materials for sensor fabrication remains the most significant challenge for achieving robust and reliable sensors capable of monitoring the real-time physiological status of patients.

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Introduction

The development of miniaturized, implantable chemical sensors that can be employed for real-time monitoring of clinically important species (e.g. blood-gas measurements such as pH, PO_2 , and PCO_2 ; electrolytes such as Na^+ , K^+ and Ca^{2+} ; glucose; lactate; and oxygen saturation (SaO_2)) remains one of the great challenges in analytical science. Academic and industrial research toward this goal has been ongoing for more than four decades [1]. Although significant advances in the designs and chemistries employed to prepare intravascular and subcutaneous devices have been achieved [1–7], the biological response of the living system toward such sensors can have a dramatic impact on the analytical accuracy of such indwelling probes [2], and this has prevented widespread use of implantable sensors in clinical practice. Herein we examine the current status of research and development in this area and provide our views on the remaining obstacles that must be overcome before indwelling chemical sensors can be employed with confidence to monitor the physiological state of human patients.

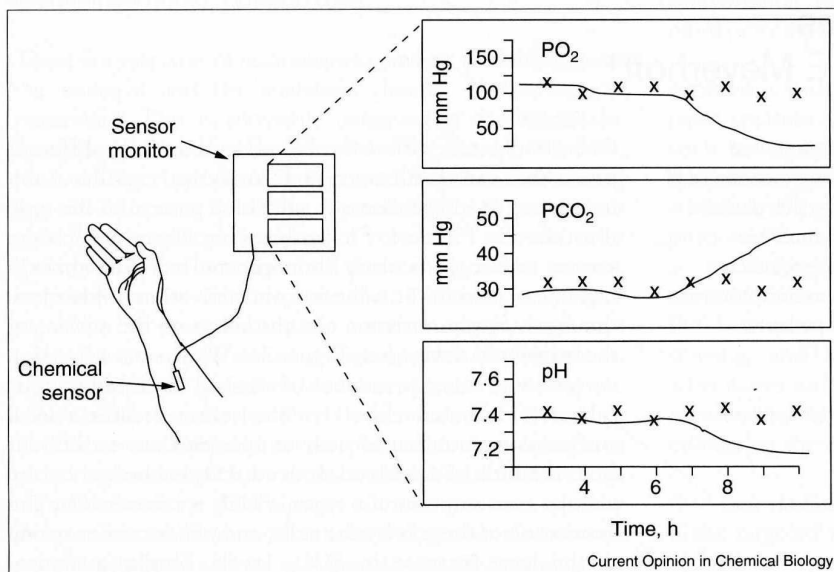
Relevant background

Analytical sensors that can be implanted within human blood vessels are an especially difficult challenge, given the very small dimensions of the required sensing catheters (< 0.6 mm outer diameter [o.d.]) [3] and the overall complexity of a whole blood sample matrix. Nonetheless, measuring blood gases as a means to monitor the cardiopulmonary function/status of critically ill patients mandates the measurement of pH, PCO_2 and PO_2 in flowing arterial blood, ideally via a catheter-type sensor placed within the radial artery of hospitalized patients (see Figure 1), and there have been enormous research and development efforts over the past 15 years aimed at achieving this goal.

Once implanted within the blood stream, three different processes can lead to errant analytical results from indwelling blood gas sensors, all with a pattern of the type illustrated in Figure 1. First, adsorption of proteins on the sensors surface, particularly fibrinogen and von Willebrand's factor, can lead to the adhesion and activation of platelets, the first steps in initiation of a thrombus on the surface of the implanted device (see Figure 2a). While sensor function is retained, the presence of highly metabolic cells (platelets) adhered closely to the sensor creates a local surface concentration of analyte species that is different from the bulk of the blood. Indeed, PO_2 is lowered by the cellular consumption of oxygen, PCO_2 is increased by the generation of this gas by the cells, and pH decreases owing to the local increase in PCO_2 levels. Similar analytical errors can be induced by the physical location of the sensor within the artery. If the chemical sensing region of the implanted device is at the distal end of the catheter, this region can end up touching the wall of blood vessel. Hence, the localized concentrations of analyte species will once again be altered by the metabolic activity of the endothelial cells that line the lumen of the vessel, yielding an error pattern identical to that observed by the adhesion and activation of platelets (see Figure 1) — the so called 'wall effect' [8]. The third process that can induce analytical errors of the type illustrated in Figure 1 is a dramatic reduction in blood flow at the implant site because of vasoconstriction of the blood vessel in which the catheter is inserted (Figure 2c). Pooling of the blood will enhance bulk metabolic changes in its content that will mimic what occurs because of cell adhesion as well as the wall effect.

These issues are less prevalent when implanting sensors within much larger blood vessels, for example the pulmonary artery, where greater blood flow coupled with the wider vessel diameter reduces the potential for these problems. However, monitoring in this particular environment is typically restricted to the most critically ill patients with heart and or lung problems, and usually involves *in situ* measurements of SvO_2 (fraction of total hemoglobin that is oxygenated in mixed venous blood) via fibre optic probes [9] mounted in pulmonary artery catheters that also quantitate cardiac output. Oxygen saturation can be determined by measuring the reflective absorption of the blood at two or three appropriately selected wavelengths [7]. However, the need for implanted catheter-type sensors to perform this function has been reduced substantially by the availability of non-invasive pulse-oximeters, which make the same measurement through the skin [7]. Hence, recent advances in indwelling SaO_2 measurement catheters will not be detailed herein.

Figure 1



Typical deviation in analytical response of catheter-type sensor for blood gases placed within radial artery of patient. Crosses represent measurements of discrete blood samples *in vitro*.

The real-time measurement of blood gas levels and oxygen saturation is only one area where indwelling sensors could dramatically enhance the quality of health care. There is also an obvious need to monitor continuously the values of critical electrolytes in blood, as changes in these levels can often signal the onset of potentially life-threatening problems (e.g. elevated potassium levels can dramatically affect heart function, etc.). *In vitro*, such measurements are carried out nearly exclusively in discrete whole blood samples by modern polymeric membrane-based ion-selective electrode technology [7,10], and thus miniaturization of such devices in catheter designs can provide the basis for preparation of indwelling electrolyte sensors.

An even greater demand exists for the design of implantable analytical sensors that can provide continuous readings of blood glucose levels to manage diabetic patients, so that a fully automated feedback control of subcutaneous insulin delivery can be achieved by newly devised, highly portable insulin pumps. Since such sensors need to function within patients outside the hospital setting, and for extended time periods (days, weeks), placement of such devices subcutaneously, rather than intravascularly, represents a more likely route to success [11–13]. Indeed, several studies have now shown that the levels of glucose in interstitial fluid track blood concentrations closely, although there is some time delay in achieving identical values in the two sample environments [13,14]. However, implanting sensors subcutaneously does not eliminate the biological response issues that can affect analytical performance. Although thrombus formation is not a problem in subcutaneous fluids, indwelling subcutaneous probes are subject to an inflammatory response that leads to encapsulation of the devices within a sheath of

leukocytes, macrophages, fibroblasts, etc., which can alter mass transport of the analyte to the surface (changing the calibration curve), and/or alter surface concentrations [13]. Indeed, simulations by Jablecki and Gough [15] predict that such changes in mass transfer can also greatly affect the lag time of the sensors response to varying glucose levels.

Several excellent reviews have been published in the past decade that summarize the history and state-of-the-art for implantable blood gas, electrolyte, oxygen saturation and glucose sensor research [1–7,11–13]. Mahutte's review [2] is particularly noteworthy, as it provides a comprehensive overview of the clinical benefits of continuous blood gas measurements, discusses details of various optical sensor designs suitable for such measurements, and points to the aforementioned need to achieve reliability, accuracy and consistency in analytical results for a large population of patients before this technology will be accepted clinically. Gerritsen *et al.* [13] offer a good synopsis of the state of subcutaneous glucose sensing as of 1999, with proper emphasis on the biological response to sensors in such an environment. The present article summarizes recent research, with a greater focus on the basic science aspects that could ultimately solve lingering analytical performance issues of implantable chemical sensors.

Measurement technologies

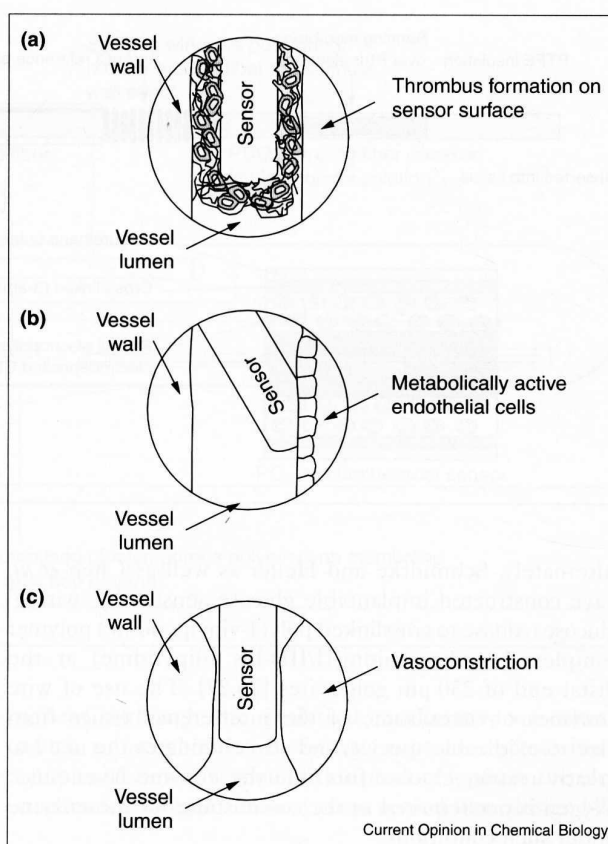
The two main analytical transduction methods that have been applied in the design of implantable chemical sensors are electrochemical and optical techniques. Miniaturizing electrochemical devices in catheter designs is an attractive approach, since almost all instruments used in hospitals to measure blood gases, electrolytes and metabolites (glucose, lactate, urea and creatinine) in discrete undiluted blood

samples employ electrochemical sensors [7,10], and thus the accuracy of this sensing technology for whole blood measurements is now well established. Implantable electrochemical oxygen sensors are usually based on a classical Clarke-style amperometric design in which oxygen is reduced at a micro-platinum, silver or gold working electrode confined (along with Ag/AgCl reference) within a narrow diameter gas-permeable catheter tube made of given polymer [1], with the resulting current being proportional to the partial pressure of oxygen in the medium surrounding the catheter. A novel potentiometric catheter design has also been proposed recently, based on measuring the corrosion potential (EMF) of a cobalt working electrode inserted within a similar narrow diameter tubing [16]. Indwelling electrochemical pH and PCO_2 sensors are typically potentiometric devices, based on either polymer membrane pH electrode technology or the use of solid-state metal-oxide-based pH sensors [4]. Incorporation of lipophilic proton ionophores (e.g. tridodecylamine) within the walls of plastic tubing provides a convenient means to prepare a novel dual lumen pH/ PCO_2 sensing design that has been demonstrated to provide accurate *in vivo* results in animal experiments [17].

Miniaturization of electrode designs that would enable several sensors to be bundled into a single implantable device, however, remains a significant engineering challenge. For this reason, over the past 15 years, many of the efforts aimed at developing commercially viable intravascular blood gas sensors for monitoring pH, PCO_2 and PO_2 simultaneously have employed modern optical fibre-based technology. As the diameter of available fibres continues to decrease, it is possible to bundle three or more separate chemically sensitive fibres in a single catheter with the required dimensions of $<600\text{ }\mu\text{m}$ o.d. [3] for implantation within human radial arteries, without dampening the pressure waveform detected by a microelectronic pressure transducer within the arterial line. Absorbance-, fluorescence- and phosphorescence-based chemistries have all been investigated in the design of sensors with suitable selectivity and calibration stability. Appropriate indicators are usually immobilized on the distal ends of the fibres, although alternate configurations/locations have also been proposed [3]. Most optical oxygen measurements are made with indicators whose luminescence is quenched in the presence of oxygen (either polycyclic aromatic hydrocarbons, metal-ligand complexes (e.g. 8-hydroxyquinolin-5-sulfonic acid with Mg(II), ruthenium(II) tris(bipyridine)), whereas pH sensors are prepared by immobilizing pH indicators (e.g. phenol red) in hydrogel-type films [18]. Optical sensors for PCO_2 can be easily prepared from the same pH sensors by incorporating bicarbonate salt in the hydrogel layer and then covering this layer with a gas-permeable polymeric film (usually silicone rubber material).

Implantable glucose (and lactate) sensors have been based, almost exclusively, on electrochemical transducers. Gough and co-workers [19,20] proposed the use of dual oxygen sensors in a single catheter design for indwelling blood

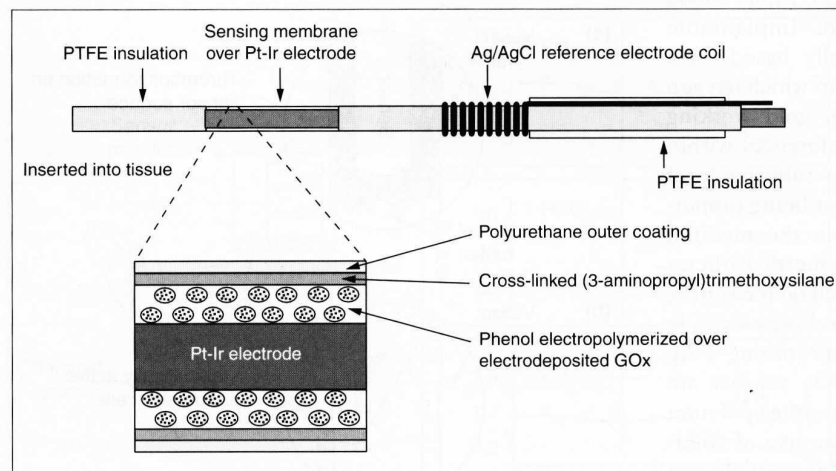
Figure 2



Analytical performance of intravascular sensors can be influenced by (a) thrombus formation on the sensor surface, (b) 'wall effect' caused by positioning the sensor near metabolically active endothelial cells and (c) vasoconstriction around the sensor.

glucose and lactate measurements. One oxygen sensor with immobilized glucose or lactate oxidase provides response to glucose levels (decreasing surface oxygen levels in response to increase in glucose or lactate), whereas the second matched oxygen sensor, without enzyme, is able to correct for unknown and varying levels of endogenous PO_2 . Others have focused on the design of probes with amperometric detection of the hydrogen peroxide (via oxidation) at an underlying platinum anode [21–23,24*,25]. In such designs, use of outer polymer films to restrict glucose (or lactate) diffusion relative to oxygen is critical to achieve a linear electrochemical response to glucose from normal (5 mM) to elevated levels found in diabetic patients (to 50 mM). Further, certain inner polymeric films are also necessary to ensure that positive interferences from other electro-oxidizable species (e.g. ascorbate, uric acid, acetaminophen) are eliminated. Such sensors have been suggested for both blood and subcutaneous glucose and lactate measurements. Figure 3 illustrates the design pioneered by Wilson and co-workers [24*], in which the needle-type probe is constructed by multiple membrane coatings and electrodeposition of the glucose oxidase layer.

Figure 3



Schematic of subcutaneous glucose sensor designed by Wilson *et al.* which utilizes electrodeposited glucose oxidase on a platinum wire covered with polymer coating. Figure adapted from [23,24^{*}] with permission. © 2002 Elsevier Science.

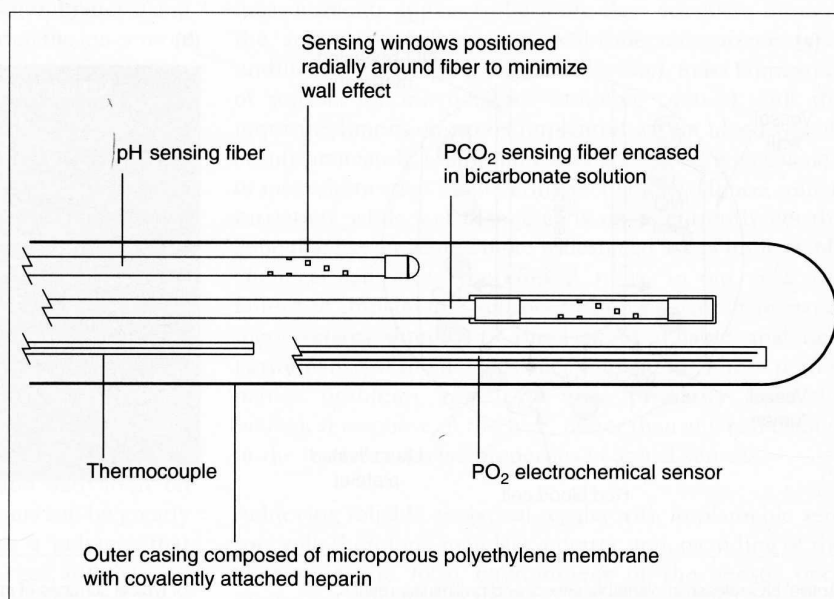
Alternately, Schmidtke and Heller as well as Chen *et al.* have constructed implantable glucose sensors by 'wiring' glucose oxidase to crosslinked poly(4-vinylpyridine) polymer complexed with osmium(II/III)-bis (dipyridine) at the distal end of 250 μm gold wires [26,27]. The use of wire enzymes obviates some of the interference issues from electro-oxidizable species, and also eliminates the need to greatly restrict glucose flux into the enzyme layer, since oxygen is not required as the co-substrate for the enzyme under such conditions.

Obtaining accurate results with the indwelling sensors described above is a significant challenge. Preferably, calibration of the device should be done once, before implantation, necessitating excellent calibration stability following insertion. In theory, use of ratiometric type measurements (two or more wavelengths) with optical sensors helps eliminate dye photobleaching and chemical leaching problems. Nonetheless, even with such approaches, employing a single *in vitro* calibration before continuous *in vivo* measurements has not proven to be possible, owing to the inherent drift in the analytical response of the device and/or changes in calibration due to the biological response of the host (protein adhesion, cell adhesion, encapsulation, etc.). For this reason, most implantable sensors are calibrated after implantation by taking a blood sample and determining the analyte level by a conventional *in vitro* test method. Daily *in situ* recalibrations (one-point) are also suggested to achieve reliable *in vivo* results [28^{**}]. Clearly, if the number of such *in situ* calibrations is excessive, the full benefits of the implantable sensor technology are obviated. Furthermore, using one-point calibrations, even frequently, will not guarantee accuracy, since the background signals in the absence of analyte can change appreciably with time for certain amperometric devices [29]. Hence, the y-intercept of the calibration curve can shift, which cannot be accounted for by the *in situ* calibration of the sensor.

Although implanting miniaturized devices with the appropriate chemistries/membranes, etc. self contained within a single probe (catheter or needle) is the preferred approach for developing *in vivo* chemical sensors, there have also been a series of reports that suggest separating the sampling and analytical detection components of the overall *in vivo* measurement system [30–34]. This can be accomplished by using microdialysis-type probes (including ones prepared with gas-permeable silicone rubber micro-bore), both intravascularly and subcutaneously, and flowing an isotonic recipient solution through the inserted probe continuously. The return line for such a solution can then pass through external analytical flow-through transducers that can detect the analytes. For example, Cooney and Towe [30,31] proposed a microdialysis catheter with an outer diameter of only 0.22 mm, along with external optical detectors, for monitoring pH and PCO_2 . Appropriate pH indicator reagent dyes (e.g. phenol red) merged with the flow lines provide the analytical signal, although no *in vivo* data was provided. Freaney *et al.* [32] suggested a miniaturized chemical analysis system equipped with glucose and lactate enzyme electrodes that can be coupled to a microdialysis sampling to monitor glucose and lactate continuously *in vivo* using a canine model for periods of up to 4 h. The advantage of this general microdialysis approach is that the analytical transducer(s) are external and can thus be calibrated, periodically, by appropriate standard solutions (via bypass valves, etc.). What is more difficult to take into account, however, is changes in the analyte mass transfer through the membrane of the indwelling dialysis probe. Indeed, if mass transfer changes dramatically, owing to cell and protein deposition, then the recovery of analyte in the flowing recipient stream will be altered substantially, and this could lead to large analytical errors. Just such a problem was demonstrated by Lutgers *et al.* [33] who studied the recovery of glucose from microdialysis probes inserted in adipose tissue of human subjects for up to three weeks. Substantial differences in the levels of

Figure 4

Schematic of the Paratrend 7 intravascular PO_2 , PCO_2 and pH sensor constructed from a hybrid design in which oxygen is detected electrochemically while carbon dioxide and pH are detected by fibre optic sensors. Figure adapted from [40] with permission. © Koninklijke Philips Electronics.



glucose measured off-line in the dialysate fluid versus blood glucose were found, and these differences changed considerably with time.

Commercial prototypes and devices

Several prototype and commercially available implantable sensors have been developed and evaluated in human subjects over the past decade. In the early 1990s, the analytical performance of a series of indwelling blood gas sensing catheters developed by various companies were examined with mixed results [35–39]. Often, the performance has been erratic, with good results in certain patients, and poor accuracy in others (when correlated to conventional sampling and *in vitro* determinations on discrete blood samples). Whether the erratic results are due to platelet adhesion/clots, wall effects, or vasoconstriction is not easy to discern when testing is being done in human patients.

Of the various prototypes products developed, most were based on optical pH, PCO_2 and PO_2 sensors. However, the only product that has been commercially available for several years (the others never made it to market, or were taken off the market shortly after product launch) is the Paratrend probe, originally developed by Pfizer/Biomedical Sensors and currently distributed by Diametrics Inc. As shown in Figure 4, this indwelling sensor consists of a novel hybrid design, in which oxygen is sensed electrochemically while pH and PCO_2 are determined by fibre-based optical fluorescence sensors [37]. While performance of the Paratrend has been reported to be acceptable [36,37], frequent *in situ* recalibrations have been suggested [36]. Nonetheless, such an indwelling device does provide, at the very least, valuable trend monitoring type information on the status of the

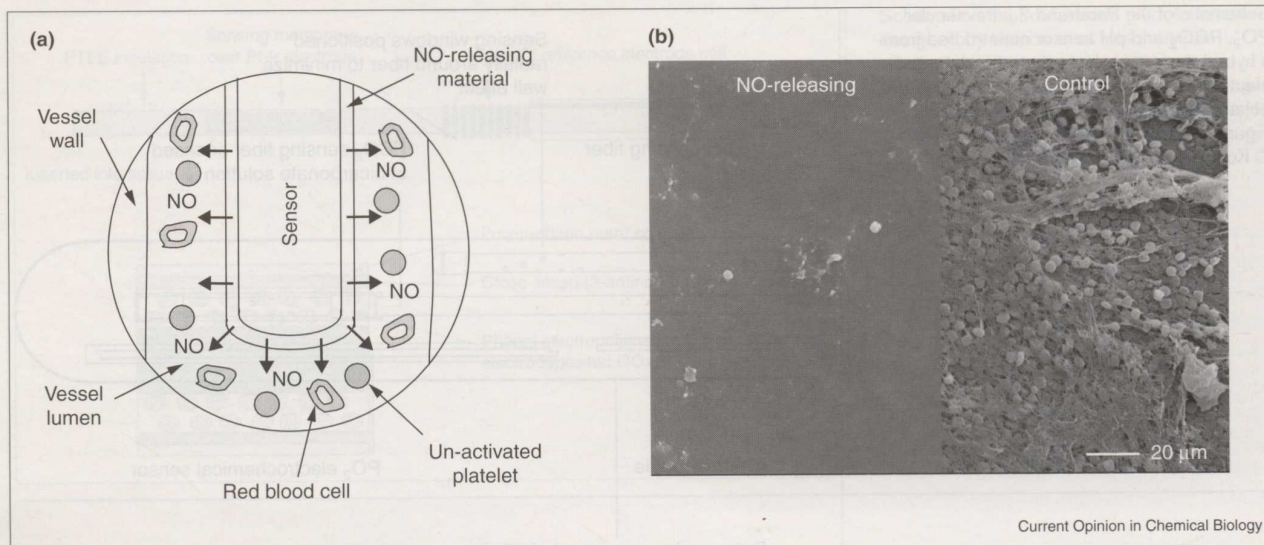
patient, although the analytical results may not always match values obtained by *in vitro* tests on discrete blood samples. A newer version of this product replaces the electrochemical oxygen sensor with an optical fibre design [40].

The only approved glucose sensor that is available for subcutaneous monitoring is made by Minimed Inc. [41]. Although the basic sensing chemistry for this device is quite similar to that of the design shown in Figure 3 (peroxide detection), the sensor is actually fabricated on a very narrow planar substrate, rather than via a cylindrical wire-based system. Relatively frequent calibration of the sensor is required via periodic *in vitro* blood tests, as well as after explantation, and hence the output of the device is not readable by the patient. Only a doctor can view downloaded data. Thus, at present, this implantable sensor does not provide real-time analytical data for dosing insulin levels, but provides the clinician with information of the dynamics of glucose in a particular patient, which can then be used to recommend a more therapeutic insulin dosing regimen. Various other companies are also pursuing the development of commercial electrochemical and optical sensor products for real-time subcutaneous glucose measurements.

New materials for implantable sensors

The requirement for repeated calibration of implantable sensors clearly results from analytical responses *in vivo* that differ significantly from *in vitro* calibration data, and sensitivities that continue to change with time. As discussed above, a substantial component of this *in vivo* 'drift' may well be related to the biological response of the host toward the foreign sensing device (see 'Relevant background'

Figure 5



'Active' NO release implantable sensor and preliminary results.

(a) Concept of an intravascular sensor that releases NO to prevent thrombus formation on the surface and (b) scanning electron micrographs

of typical surfaces of intravascular oxygen sensors that released NO compared with a control that did not release NO after 20 h implantation in porcine carotid arteries with no systemic anticoagulation of the animal.

section). This is sometimes referred to as 'biofouling' and general approaches to reduce such effects, both for fully implantable sensors as well as indwelling microdialysis-type devices, were reviewed recently by Wisniewski and Reichert [42]. In fact, a substantial volume of basic research has focused on fabricating sensors with more biocompatible materials as outer coatings/membranes for the devices. For example, Quinn *et al.* [43] demonstrated that subcutaneous glucose electrodes based on wired glucose oxidase were less encapsulated after three days of implantation within rats when prepared with an outer coating of photocrosslinked terpolymer of 2-hydroxyethyl methacrylate, poly(ethylene glycol) tetra-acrylate and ethylene dimethacrylate. Apparently, the very hydrophilic nature of such polymers greatly reduces protein adsorption, and leads to less of an inflammatory response *in vivo*. Similarly, Kros *et al.* [44,45] examined various sol-gel derived hybrid materials as more biocompatible outer coatings for preparing glucose electrodes. They found that fibroblast cell proliferation was dramatically reduced on sol-gel coatings that contained the polyanions dextran sulfate or polystyrene sulfonate. These same composites were evaluated as outer coatings *in vivo* by the construction of glucose sensors that were implanted subcutaneously. Unfortunately, no significant difference in the *in vivo* tissue response was found among the various composites versus control sensors.

Zhang and co-workers reported on the *in vitro* hemocompatibility of electrochemical oxygen sensing catheters [46] and optical fibre-based pH sensors [47] prepared with a novel copolymer of *n*-butylmethacrylate and 2-methacryloxyethyl phosphorylcholine. A similar polymer with

pendant phosphorylcholine was also employed to prepare a fibre-optic-based oxygen sensor [48] as well as an *in vivo* glucose sensor [49]. The phosphorylcholine mimics the outer lipid layer of many cell membranes, and it is envisioned that such a material in contact with blood should exhibit less biofouling. While *in vitro* data and spectroscopic studies of protein and cell adsorption suggest this may be the case, only the glucose sensor was tested *in vivo*, but only for up to 3h. Clearly much more substantive *in vivo* testing for longer time periods will be necessary to fully support the hypothesis that polymers with phosphorylcholine segments could potentially decrease the biological response to implanted sensors fabricated with these materials.

There has also been considerable effort aimed at enhancing the biocompatibility of the polymer membranes used to construct modern ion-selective electrodes and ion-selective field effect transistors [50] (for K^+ , Na^+ , Ca^{2+} and pH). Such devices can be readily miniaturized for *in vivo* sensing [4]. Improving the blood compatibility of such devices, however, without degrading the analytical response properties is somewhat more difficult than with gas sensors and enzyme-based probes, since the analytical signal (voltage) comes directly from the blood/sensor interface. Hence, altering the chemistry of the polymers could potentially influence ion transfer rates across the interface as well as the thermodynamics of ion extraction into the organic polymeric phase (changing selectivity). Nonetheless, Berrocal *et al.* [51] showed that sodium and potassium electrodes constructed with BioSpan-5, a novel copolymer of polyurethane and poly(dimethylsiloxane), exhibited

reduced platelet adhesion compared with that of conventional poly(vinyl chloride)-based membranes using *in vitro* test methods. Brooks *et al.* [52] reported the ion-sensors constructed with immobilized heparin, a potent anticoagulant, also exhibited improved biocompatibility while maintaining nearly theoretical ion response properties.

One new approach to reduce the biological response to implanted sensors is to prepare sensors that continuously release low levels of an anti-fouling agent, while still allowing the sensor to exhibit a stable analytical response. Espadas-Torre *et al.* [53] initially demonstrated that ion-selective membrane electrodes could be formulated with nitric oxide (NO) release chemistry, and that the polymer materials used in these sensors exhibited greatly reduced platelet adhesion and activation when equilibrated *in vitro* with platelet-rich plasma. Subsequently, Schoenfisch *et al.* [54**] showed that platelet adhesion and activation on intravascular electrochemical oxygen sensors can be greatly reduced when the sensor is coated with a polymer that slowly releases physiological levels of NO gas, and that this can lead to improved analytical performance of such sensors when monitoring oxygen within carotid or femoral arteries of dogs. This concept is illustrated schematically in Figure 5a, for the case of intravascular sensors.

It is well known that NO is a potent naturally occurring anti-platelet agent. Nitric oxide is released continuously at low fluxes (approx. 10^{-10} mol/cm² min) from endothelial cells that line the inner walls of all healthy blood vessels, and it has been suggested that this basal level of NO is critical to prevent platelet adhesion and activation on the walls of the vessels. A newer silicone rubber material possessing the NO release chemistry bound to the polymer backbone [55] has also been tested as a coating *in vivo* using the same oxygen-sensing catheter as a model device [54**]. Figure 5b illustrates the typical difference in the degree of surface fouling by platelets and fibrin clots, when an NO release and control sensor are placed in the carotid arteries of the same pig for a period of 20 h without any systemic anticoagulation. Such reduction in the degree of platelet adhesion and activation has been observed repeatedly in a series of similar *in vivo* experiments, supporting the possibility that NO release chemistry, which mimics a natural physiological process, could provide significant benefit for improving the performance of intravascular sensors. Further, the prospects of using new NO release sol-gels as outer coatings for glucose sensors has also just recently been reported [56], with functional enzyme electrodes obtained in the presence of the NO release chemistry. Whether NO release will also help reduce the biological response toward such sensors when implanted subcutaneously, remains to be seen, although it is well known that NO can enhance blood flow and the wound-healing process [57].

Research needs and future directions

It should be clear from above discussions that the selectivity, sensitivity and response times of the miniaturized

electrochemical and optical sensors developed for *in vivo* measurements appear to be more than adequate to meet the analytical challenges of real-time measurements in undiluted physiological matrices. Further, mass fabrication of sensors (or microdialysis sampling probes) with the requisite dimensions to be implanted within blood vessels or subcutaneously, is now quite easy with modern mechanical or microfabrication engineering technology. Hence, miniaturization, while a challenge early on, is currently not the issue that has prevented more widespread use of implantable chemical sensors in the clinical arena. In our view, the failure of implantable sensors to impact patient management relates directly to the lack of reliable analytical performance of the devices once implanted. These performance problems appear to arise primarily from the biological response of the host, rather than any real change in the innate response properties of actual sensors.

Achieving reliable analytical results with implantable sensors will, therefore, mandate a better understanding of the change in the local environment of the sensor once implanted, whether in blood or subcutaneously. As long as such changes impact analytical calibration of the devices, or yield a local concentration of analyte that is different than the bulk test fluid, then observed performance problems will always exist. Although *in situ* calibration of sensors by periodic *in vitro* measurements (one-point calibrations) can partly overcome such problems, assessing the true sensitivity of implantable probes by required two-point calibrations is not possible by this approach. Further, if the required frequency of *in vitro* calibration is high, there is less value gained from having the sensor implanted at all.

The goal of achieving accurate analytical results with indwelling sensors can only be realized by preparing sensors that elicit less response from the test subject. This can best be accomplished by a combination of using more biocompatible materials to construct the sensors and also by creating devices that release low concentrations of agents that will suppress the biological response process (so-called 'active' sensors). The use of NO release, as summarized above, is only one example of what may be possible in this latter direction. It is well known that the most non-thrombogenic surface is the endothelial cells that line blood vessels. In addition to NO, these cells liberate low levels of prostacyclin, plasminogen activators and other anti-platelet agents. Further the surface of such cells possess anchored heparans, thrombomodulin, ADPase, and other species that are involved in decreasing the thrombotic response. In the case of subcutaneous sensors, slow release of agents that could decrease the inflammatory response and prevent macrophage activation may prove beneficial.

There are two keys to the potential success of this 'active' sensor concept. First, the presence of the released and/or immobilized agent within or on the outer coatings of the sensor cannot interfere with the analytical response of the implanted device. Second, the loading of the reagent

within or on the outer surface of the sensor must be adequate to provide an activity for an extended time period (e.g. at least 3 days for intra-arterial blood gas sensors [2]).

There is already a precedent for the potential success of this research direction. Indeed, considerable recent work has focused on ways to reduce the biological response when stainless steel metal stents are employed to permanently expand coronary arteries during balloon angioplasty procedures. Damage to the underlying smooth muscle cells during placement and expansion of such stents can often result in proliferation of these muscle cells and result in restenosis (i.e. reclosing) of the artery after several months. To combat this problem, researchers are using biocompatible polymeric coatings that also release low levels of anti-cell proliferation agents, locally at the interface of the stent and the vessel wall [58]. The first generation of these so-called drug-eluting stents will soon be commercialized to dramatically decrease the restenosis problem. It seems reasonable, therefore, that a similar strategy could be successful when applied to implantable sensors, provided that the nature of the agents used is carefully chosen so as not to interfere with the normal response properties of the indwelling probes. Hopefully, this more active approach will help resolve the lingering biocompatibility problems and thereby greatly enhance the analytical performance and reliability of the various intravascular and subcutaneous implantable chemical sensors discussed in this review.

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