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Analysis: On the Path to Overcoming Glucose-Sensor-Induced Foreign Body Reactions

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Abstract

It is generally accepted that unreliable *in vivo* performance of implantable glucose sensors originates, in large part, from tissue reactions to the implanted sensor, including foreign body reactions (i.e., inflammation, fibrosis, and vessel regression). Development of glucose sensor coatings with increased biocompatibility would contribute to the development of a reliable long-term glucose sensor. In this issue of *Journal of Diabetes Science and Technology*, Van den Bosch and coauthors report on their initial *in vitro* results on a candidate biocompatibility coating for sensors (silica nanoparticle- polyethylene-glycol-based coating). Although the initial standard testing is encouraging, it is important that sensor-specific testing protocol be utilized to more accurately predict sensor performance *in vivo*. The development and application of sensor-specific testing standards will likely speed the development of biocompatible coatings that will increase sensor accuracy and lifespan in the future.

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Currently, commercial glucose sensors used in continuous glucose monitoring (CGM) are approved by the Food and Drug Administration for only 3–7 days. Increasing the accuracy and lifespan of implantable glucose sensors is essential in advancing CGM management of diabetes, including closed-loop (artificial pancreas) technology. One of the major factors contributing to unreliable short-term and long-term *in vivo* performance of implantable glucose sensors originates from tissue reactions to the implanted sensors (i.e., inflammation, fibrosis, and vessel regression). Frequently, this group of tissue reactions is referred to as a foreign body reaction to the implanted sensor (a foreign body). These tissue reactions decrease glucose availability and retard glucose diffusion within the implantation site, thereby compromising sensor function. Accumulation of vascular cells (e.g., leukocytes or red blood cells) within the implantation site can create "metabolic barriers" to glucose diffusion to the implanted sensor and result in deceptively low and delayed glucose readings by the sensor.[1–3] Sensor-associated fibrosis not only compromises sensor function by slowing glucose diffusion within the implantation site, but even more profoundly, fibrosis impacts sensor function by inducing blood vessel regression at the implantation site. Glucose sensors, unlike many other implantable devices, require a close proximity to blood vessels to determine real-time blood glucose levels. In general terms, one can view the tissue responses to implanted glucose sensors in large part as a cellular response to the sensor, which limits sensor function. Preventing and/or overcoming these cellular responses would be a major step in improving the accuracy

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Abbreviations: (CGM) continuous glucose monitoring

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and reliability of long-term glucose sensors *in vivo*. Development of new glucose sensor coatings with increased biocompatibility as it relates to inflammation, fibrosis, and vessel regression would provide a major contribution to the development of a reliable long-term glucose sensor to clinically manage patients with diabetes.

In this issue of *Journal of Diabetes Science and Technology*, Van den Bosch and coauthors[4] report on the result of their initial *in vitro* study, testing a nonbiofouling coating as a means to enhance compatibility when utilized as a sensor membrane, using standard *in vitro* and limited *in vivo* studies, including cross-hatch cutting, wet paper rub, paper double rub, bending, hydrophilicity, protein adsorption, hemocompatibility, and glucose/oxygen permeability and irritation testing, in an effort to support the potential of VitroStealth® to function as an effective coating for an implantable glucose sensor. These studies indicate that VitroStealth has good physical characteristics, low protein binding, and low cell toxicity *in vitro*. It is tempting to conclude that excellent *in vitro* performance of a nonbiofouling coating showing limited protein adsorption will lead to an equally superior glucose sensor performance *in vivo*. Since acceptable *in vivo* results are not assured, it is then critical to evaluate these coatings in "sensor-specific" *in vitro* and *in vivo* platforms. For example, *in vitro* toxicity assays have value, but more predictive biocompatibility testing needs to go beyond *in vitro* toxicity to address implantable-glucose-sensor-induced tissue reactions (i.e., inflammation, wound healing, and fibrosis) through *in vitro* leukocyte activation assays. Since all implantable devices lead to a foreign body reaction, it is important to determine whether the biomaterial or coating causes activation of leukocytes, such as macrophages. Biomaterials or coatings cause significant *in vitro* activation of primary cultures of macrophages or macrophage cell lines, e.g. THP-1 (human) or RAW (mouse) lines. Markers of leukocyte activation, such as cytokine, growth factor, or cluster of differentiation/cluster of designation expression represent relatively simple, relevant, and quantitative metrics to fully evaluate implantable materials. Although these types of *in vitro* studies with various cell populations are important in initially evaluating biomaterials and devices, it is equally critical to eventually undertake *in vivo* evaluation.

In vivo testing of glucose sensors and coatings requires sensor-specific modeling of the "real world" of sensors, i.e., inflammation, wound healing, and fibrosis, including foreign body reactions. In the past, the National Institutes of Health already emphasized the need for a better understanding of the interactions of proteins and cells with the sensor surface, including the process of fibrosis (e.g., encapsulation of the sensor).[5] Although some progress has been made in these areas, the *in vivo* utility of the sensors is still limited by their brief functional lifespan in clinical use to only several days. In contrast, over the years, the biomaterials community has invested significant efforts in various device coatings in an effort to provide a sensor surface with controlled or limited protein adsorption in the hope that the device will be undetected by the immune system (innate and acquired immunity). Inflammatory cells are important in the host defense against foreign objects, including microorganisms, via metabolically intense activities (i.e., glucose metabolism) such as chemotaxis, phagocytosis, and generation of reactive oxygen species. Sensors, like microorganisms, are foreign objects and also trigger these same intense metabolic activities. In previous studies using a continuous glucose sensor mouse model, our laboratory demonstrated the critical role of inflammatory cells contributing to the sensor performance variation *in vivo*.[1–3] The preeminent role of the mouse is the result of large number of mutant and transgenic mice and related tools (e.g., recombinant proteins, antibodies, and drugs). These tools can provide important insight into tissue reactions and glucose sensor function and have only recently been appreciated. As such, the mouse allows investigators to transcend simple histopathology studies with the identification of the cells, mediators, and mechanisms that can be targeted to overcome the tissue reactions that limit sensor function and lifespan *in vivo*. In fact, using various mouse models of CGM our laboratory demonstrated the critical role of mast cells,[1] macrophages,[6] and cytokines and cytokine inhibitors[6] as well as the importance of glucose metabolism by red blood cells (micro-*hemorrhage*)[2] and by inflammatory cells recruited to the site of sensor implantation. Inflammatory cells at the site of device location are detrimental to its functionality since inflammatory cells are metabolically very active. As such, the migration of inflammatory cells to the site of sensor location creates a metabolic barrier to the diffusion of glucose from the vasculature. These metabolic barriers can prevent detection of hypoglycemic events and, as such, can compromise CGM, which could result in life-threatening consequences, particularly in situations involving closed-loop systems to monitor glucose levels in diabetes patients.

Over the years, the emphasis in the sensor community has been on the development of improved biocompatible sensor coatings. In order to lengthen device durability (e.g., weeks to months and, eventually, years), an equal emphasis on

controlling the tissue reaction around the implanted device is needed if the future shall provide a reliable closed loop outside the clinical setting. It is very unlikely that simple coatings alone will overcome the host responses to the device but composite coatings involving biomatrices, tissue-response modifiers, and long-term drug delivery systems hold great promise for the future. These composite coating/membranes could promote vascularization, and/or inhibiting the inflammatory or fibrotic response will eventual lead to a highly accurate sensor with reliable long-term CGM.

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References:

1. Klueh U, Kaur M, Qiao Y, Kreutzer DL. Critical role of tissue mast cells in controlling long-term glucose sensor function *in vivo*. Biomaterials. 2010;31(16):4540–51.
2. Klueh U, Liu Z, Feldman B, Henning TP, Cho B, Ouyang T, Kreutzer D. Metabolic biofouling of glucose sensors *in vivo*: role of tissue microhemorrhages. J Diabetes Sci Technol. 2011;5(3):583–95.
3. Klueh U, Liu Z, Feldman B, Kreutzer D. Importance of interleukin-1 and interleukin-1 receptor antagonist in short-term glucose sensor function *in vivo*. J Diabetes Sci Technol. 2010;4(5):1073–86.
4. Van den Bosch EEM, de Bont NHM, Qiu J, Gelling OJ. A promising solution to enhance the sensocompatibility of biosensors in continuous glucose monitoring systems. J Diabetes Sci Technol. 2013;7(2):455–64.
5. National Institutes of Health Bioengineering Consortium (BECON). Sensors for biological research and medicine. Natcher Conference Center Bethesda, Maryland, June 24–25, 2002.
6. Klueh U, Frailey J, Antar O, Qiao Y, Kreutzer DL. Critical role of macrophage chemokines in controlling continuous glucose monitoring *in vivo*. J Diabetes Sci Technol. 2012;7(1):A72.