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Optical Biosensing and Bioimaging with Porous Silicon and Silicon Quantum Dots

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(Invited Review)

1. INTRODUCTION

Silicon is the second most abundant element in Earth's crust, and it is considered one of the most important materials for the world. Crystalline silicon has continued to serve as the foundational building block for the microelectronic industry, and new forms of silicon materials have promised an even brighter future with emerging applications from optoelectronic devices, energy and environment technologies and new therapeutics [1–3]. Many of these promises are often associated with reduction of the physical size of the material to the micro/nano scale which yields novel physical properties. For this reason, understanding and learning how to control these features is of high importance, and unsurprisingly, low dimension silicon structures have drawn broad research interests from physicists, chemists, materials engineers and medical scientists.

At the new frontiers of nanostructure silicon research, biomedical applications are very appealing because silicon is highly biocompatible [4]. With the small sized silicon materials suitable for these applications, two distinct structures are porous silicon, and silicon nanocrystals which are also called quantum dots. Porous silicon is a form of crystalline silicon where the surface is embedded with nanometer sized pores [5], while silicon quantum dots are ultrasmall crystals of only a few nanometers in size [6]. They both exhibit unique optical features suitable for sensing and imaging, which can be tuned via comparable surface engineering methods. For this reason, this review combines the two subjects in one article, with the scope of advancing th fields through a comparative approach. Since both porous silicon and silicon quantum dots have been actively researched in the past two decades and multiple excellent reviews have been published [3, 5, 7], this paper will only highlight recent progresses in the past several years.

2. POROUS SILICON

Porous silicon was first discovered by Uhlir in Bell laboratory in 1956 when studying the electro-polishing of silicon in hydrofluoric acid solutions [8]. However, only after 1990 efficient visible photoluminescence of porous silicon at room temperature was discovered [9]. The pioneering work of Lin et al. on porous silicon-based optical biosensors inspired numerous subsequent research in its bio-applications [10]. The advantage of porous silicon in bio-applications lies in its high surface to volume ratio, tunable structure, low cost, facile fabrication and compatibility with existing silicon treatment technology. As several other reviews and book chapters have discussed early research on porous silicon optical biosensors [11–14], only recent studies, particularly emerging sensing strategies based on porous silicon, are present here. In addition, luminescent porous silicon for bioimaging will be briefly covered.

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2.1. Fabrication of Porous Silicon

Anodization of silicon in hydrofluoric acid solution is considered the best technique to form homogeneous porous structures reproducibly, although other methods have also been reported, such as stain etching [15], galvanic etching [16], metal-assisted chemical etching [17, 18], and contactless electrochemical etching [19]. In the anodization process, semiconductor silicon with defect electrons (e^-) or holes (h^+) is dissolved to form pores orthogonal to the Si surface (Figure 1) [20]. The dissolution of silicon only takes place at the pore tips and the already formed porous structure is not affected by subsequent etching due to its self-limited etching process [21]. The substrate resistivity (Si doping level) and anodization conditions (e.g., current density, hydrofluoric acid concentration and temperature) are the main factors controlling porous silicon formation. Among them, the current density is the most important parameter in tuning the pore condition [21].

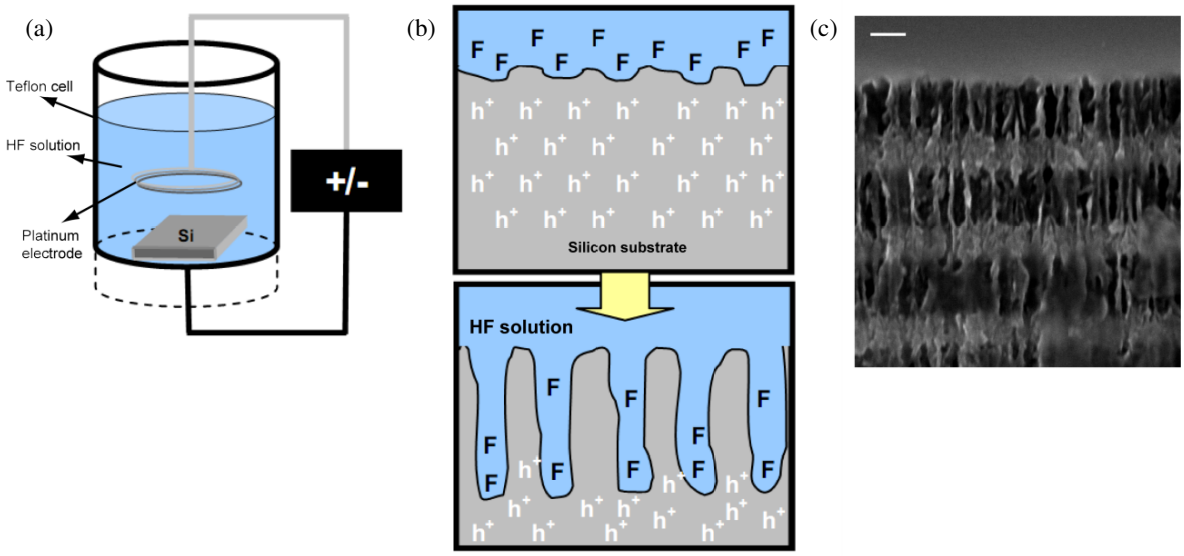

Figure 1. (a) Schematic electrochemical cell of porous silicon formation, (b) depiction of pore growth on p-type silicon during the hydrofluoric acid (HF) etching process, and (c) cross-sectional scanning electron micrograph of resultant pore morphology in "layers" with high (dark grey) and low (light grey) porosity. Scale bar equals 100 nm.

2.2. Optical Properties of Porous Silicon

Photoluminescence is one of the exciting characteristics of porous silicon. The so-called "S-band" or "red-band" luminescence ($\sim$ 560 to 860 nm) has drawn the most attention and has profound technological significance, since it can be efficiently excited with electric fields [22]. The origin of "S-band" luminescence, though controversial, can be explained by a quantum confinement model [9]. It is generally agreed that the existence of nano crystallite Si evokes the quantum confinement, causing the band gap of porous silicon larger than that of bulk silicon to generate visible photoluminescence. While all types of porous silicon can show photoluminescence if porosity is sufficiently high, its intensity is subject to various factors. Interested readers can refer to the early review for detailed information [22].

Besides the luminescent properties, porous silicon is also attractive for its ability to form multilayered structures with unique optical properties [23]. As mentioned, the current density plays an important role in controlling the achieved porosity for a given silicon substrate during the etching process. Porous "layers" with different porosities (e.g., the "layers" shown in Figure 1(c)) can be realized when modulating the current density, owing to the self-limited etching process. The thickness of each "layer" is determined by the etching time. A periodical switching between two distinct current densities will produce porosity multilayers with a varied refractive index profile [24]. These multilayers are able to exhibit a photonic band gap, preventing light propagation at a range of frequencies due

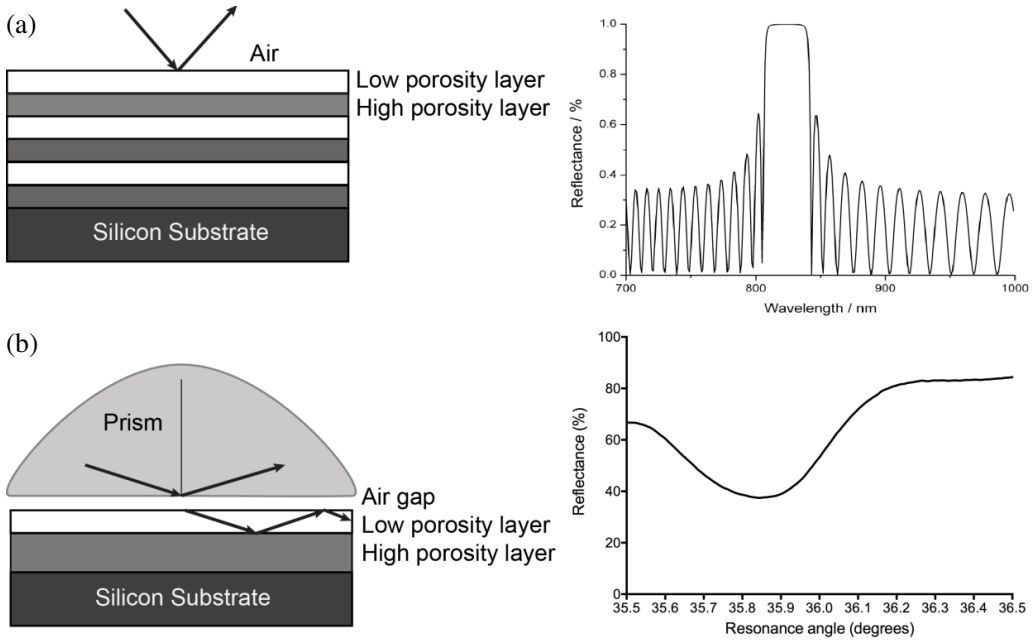

Figure 2. Schematic drawing of the layer structure of porous silicon Bragg reflector (a) and waveguide (b) and their corresponding optical spectra on the right.

to multiple light reflection on interfaces between layers, rendering photonic crystal structures such as Bragg reflectors [25–27], microcavities [28–30] and Rugate filters [31, 32]. Furthermore, they can also be engineered to couple/guide light of specific wavelength and incidence angle into a propagating mode within the layers, forming resonant structures [33] such as waveguides [34–36] and Bloch surface waves [37–39]. The model structures of photonic crystal (Bragg reflector) and resonance porous silicon (waveguide) and their reflectance spectra are displayed in Figure 2. The peak position in Figure 2(a) or the dip position in Figure 2(b) is directly dependent on the refractive index of the porous layers, which is the key to sensing applications.

2.3. Porous Silicon Optical Biosensors

Porous silicon acts as a signal transducer in optical biosensors. With the merit of its diverse optical properties mentioned above, biological signals from the interaction of analytes on the sensing interface can be transduced in several ways. With the luminescent property of porous silicon, sensing relies on the analyte induced fatigue or quench of photoluminescence intensity [40]. For porous silicon photonic crystals, the refractive index of porous layers determined by the porosity is associated with the bandgap position on the optical spectra. Any change occurring in the pores caused by analytes adsorption leads to a direct reflection on the spectral shift. The transduction mechanism for porous silicon resonant structures is similar, as it also relies on the correlation between the changes in the refractive index of porous silicon layers affected by analytes and the angular or spectral shift of the resonance mode. Thus by applying the effective-medium theories of dielectric function [41] to model the spectra of porous silicon, the relationship between the refractive index of porous silicon film and the spectral shift can be quantified. The concentration of the analytes can then be resolved. In the following text, recent progress in porous silicon optical biosensors (including bioimaging) will be discussed according to the transduction mechanism derived from each optical property.

2.3.1. Photoluminescence Based Biosensing and Bioimaging

The interaction of target analytes with luminescent porous silicon can be monitored by the quench of photoluminescence. A comprehensive review by Sailor et al. has covered the sensing work up to

2009 based on photoluminescence quenching through various mechanisms [40]. Most of the subsequent investigations follow the same lead as to relate the decreased intensity with the influence of the analytes [42, 43]. An interesting work demonstrated by Myndrul et al., however, used a gold-coated porous silicon nanocomposite as a photoluminescence sensing platform to detect toxins [44]. With the surface plasmon resonance effect of gold layer on porous silicon to further decrease the photoluminescence intensity, the sensitivity of this biosensor was greatly enhanced. Thus, integrating other optical sensing techniques onto porous silicon might bring opportunities for biosensing research based on photoluminescence.

The luminescent property also makes porous silicon an attractive biomaterial in bioimaging. Luminescent porous silicon nanoparticles with emission on 650–900 nm range, fabricated by lifting off porous silicon film and sonication, are suitable candidates for *in vivo* imaging [45], as like silicon quantum dots, they can be easily degraded in aqueous solutions into non-toxic orthosilicic acid. However, the application of porous silicon luminescent particles in *in vivo* imaging still faces obstacles from tissue autofluorescence and limited penetration of excitation light into tissues. Advanced fluorescence microscopy techniques are being used to bypass this problem. For instance, a time-gated imaging system was developed to distinguish porous silicon luminescence (long emission lifetime, 5–13 μs) from background autofluorescence (short-lived emission, < 10 ns) [46]. Two-photon process was employed to excite the luminescent porous silicon with near-infrared radiation to increase the imaging resolution [47]. Moreover, the luminescent porous silicon nanoparticles can be chemically engineered to present a specific probe to accumulate on the sites of interest for tagging and imaging [47], while the pores can be loaded with drugs for theranostic application [48].

2.3.2. Photonic Crystal Based Biosensing

The exciting performance of porous silicon photonic crystals as label-free optical biosensors was described in previous reviews [11–14], but the applications of these optical sensors in detecting biomolecules of low concentration still face challenges. The sensitivity of these biosensors is usually insufficient in clinical diagnosis or environmental monitoring. Thus in recent years there have been an increasing number of publications on how to amplify the optical signals and overall enhance the performance of porous silicon as label-free biosensors. Here the recent improvement in the biorecognition interfaces, signal processing and sensing strategies in photonic crystal based biosensing will be explained.

Antibodies are popular among other biorecognition elements in porous silicon optical biosensors. Lately, aptamers, short single-strand oligonucleotides that have high and specific binding affinity to proteins, have become new bioreceptors for the detection of various analytes [49–52]. Aptamers have advantages over antibodies in fast, low-cost and controllable synthesis and chemical stability [53–55]. Chhasatia et al. compared the performance of an antibody-modified porous silicon optical biosensor to an aptamer-porous silicon biosensor in the analysis of insulin in islet production [56]. The results suggested a better sensing performance (i.e., shorter response time and lower detection limit) with aptamers as bioreceptors than antibodies. Urmann et al. published a rapid and specific detection method based on aptamer-modified porous silicon with a sandwich assay to enhance the detection sensitivity [57]. Besides aptamers, peptides have also been used in recognizing target analytes in sensing process, especially in bacteria detection. It was reported that a synthetic antimicrobial peptide was used as a sensing probe on nano-porous silicon optical biosensors to detect *Escherichia coli* bacteria [58]. The resulting detection limit was found to be one order of magnitude lower than that of the same porous silicon optical biosensor with antibody as a sensing probe. Additionally, the adjustment on the orientation of sensing probes to increase the bioreceptor density was also reported to improve the performance of porous silicon biosensors [59].

Barillaro and his group worked on a novel signal-processing technique for the optical readout of porous silicon interferometric biosensors to improve biomolecule detection by 1000 fold [60, 61]. The technique, called interferogram average over wavelength (IAW) reflectance spectroscopy [62], processes the reflection spectra of porous silicon after the absorption of biomolecules inside the pores by subtracting a reference reflection spectrum acquired in buffer solution. The reference spectrum is used to obtain a background of instrumental noise. As a result, the IAW strategy in data process is able to achieve higher reproducibility and lower detection limit, as compared to conventional analysis based on optical shifts. Furthermore, in the paper a new porous silicon preparation method with two steps of

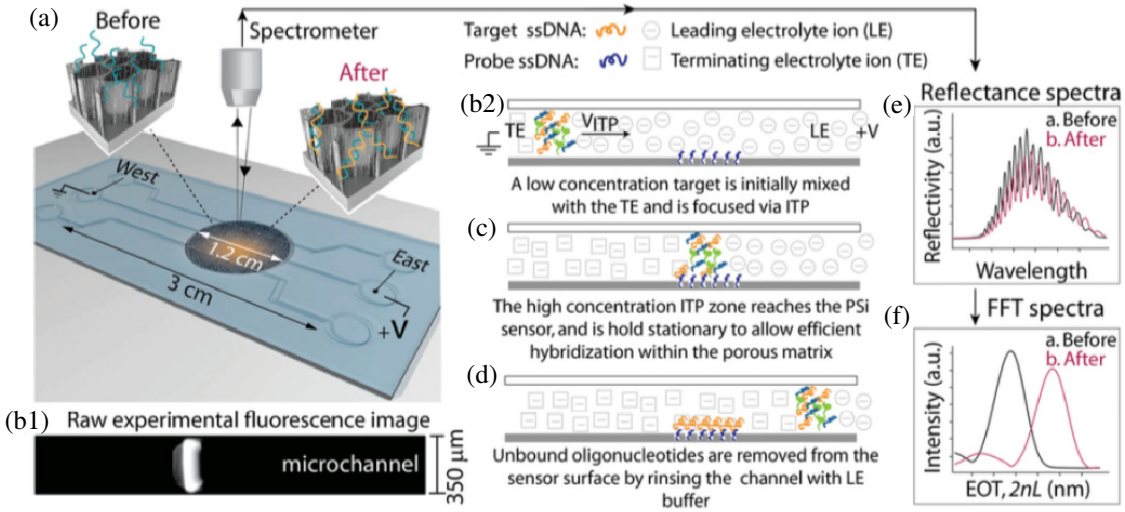

Figure 3. (a) Schematic illustration of the porous silicon interferometric biosensor coupled with ITP on a microfluidic device. (b)–(d) The concentration process of target DNA under ITP and the binding to the immobilized probe DNA. (e), (f) The spectral shift due to binding of the target ssDNA and the resulting increase of effective optical thickness (EOT) of the porous silicon biosensor [63] Copyright 2015 Wiley-VCH.

etching (sacrificial layer and porous layer) was introduced to further improve the sensitivity by a factor of 10. It was found that the current density used for sacrificial layer could change the pore size of the next etched porous layer while the porosity of the next etched layer is constant. Hence, this new porous silicon fabrication method allows bigger pore size for more effective diffusion and binding of analytes into pores, without changing the porosity, i.e., the refractive index of porous silicon to sacrificing the sensitivity.

A different signal enhancement technique based on pre-concentration of analytes was reported by Segal and coworkers on porous silicon optical biosensors as shown in Figure 3. Isotachophoresis (ITP), a form of electrophoresis to separate charged analytes based on ionic mobility under external electric fields, was incorporated into sensing process to pre-concentrate analytes (nucleic acid and proteins) for improving the sensor sensitivity by 1000 fold [63, 64]. The experimental design was shown in the following schematic illustration. Note that the porous silicon structure need to be oxidized prior to sensing application in order to isolate from the high voltage required for electric field. Despite the increased sensitivity, this signal enhancement technique is only applicable to analytes with charges.

Another limitation imposed on the applications of porous silicon optical biosensors is the inefficient analyte transport, i.e., slow diffusion rate of analyte into pores [65, 66]. A simple way to deal with it is to change the setup of porous silicon biosensors by flowing analytes through rather than over the sensing device [67]. The idea is to fabricate open-ended porous silicon membranes as sensing platforms for easy infiltration and efficient interaction of analytes (especially large biomolecules) with the sensing interface (Figure 8). According to the paper by Weiss and her group [68], a 6-fold improvement in sensor response can be realized with the flow-through porous silicon membrance versus the flow-over porous silicon film when detecting streptavidin in solution with a typical biotin modification on the surfaces (Figure 4).

In contrast to Weiss's strategy to ease the in-diffusion of analytes to the pores, Gooding and his group have made effort to increase the out-diffusion of the byproducts of enzymatic reactions from the pores to improve the protease detection with a porous silicon Rugate filter biosensor [69, 70]. The sensor was fabricated by pre-filling the pores with a synthetic polymeric enzyme substrate containing peptide sequences. Upon the cleavage of the peptide sequence by target protease, polymeric segments were removed from the pores, resulting in a significant drop in the refractive index and further a larger optical shift compared to conventional shifts induced by analyte absorption/desorption. A lower detection limit and higher sensitivity can be achieved accordingly.

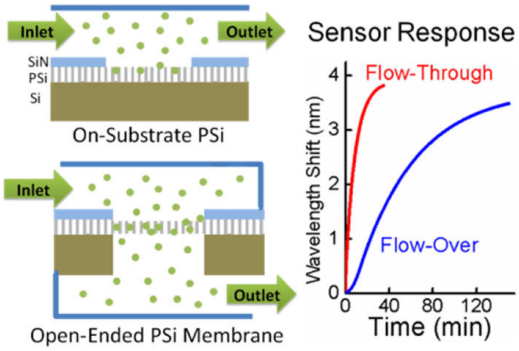

Figure 4. The schematic illustration of a flow-over porous silicon biosensor versus a flow-through porous silicon membrane-based biosensor and their difference in terms of sensing response [68]. Copyright 2016 American Chemical Society.

2.3.3. Resonance Based Biosensing

Porous silicon resonant structure (e.g., surface waves and waveguides) based sensors are receiving substantial interests in its applications on biosensing [71–75], as the resonant layer of porous silicon is able to increase the intensity of light to interact with biomolecules, thus improving the detection sensitivity. Some of the most recent examples can be found here.

Qiao et al. fabricated a protease biosensor using prism-coupled porous silicon Bloch surface wave structure that consists of a truncated defect layer on top of a Bragg reflector (Figure 5) [76]. The "open" surface wave layer renders easy diffusion for biomolecules and the Bragg reflector underneath serves an internal reference to realize sensitive protease detection. Rather than using a prism to couple the evanescent waves on the sensing interface, a series of patterned porous silicon resonance biosensors were developed by Weiss and her group [77]. Rodriguez et al. developed a grating-coupled Bloch surface wave/ waveguide biosensor for selectively detecting small and large molecules, suggesting the Bloch surface wave sensor superior for detecting large molecules due to the easy access to the active sensing

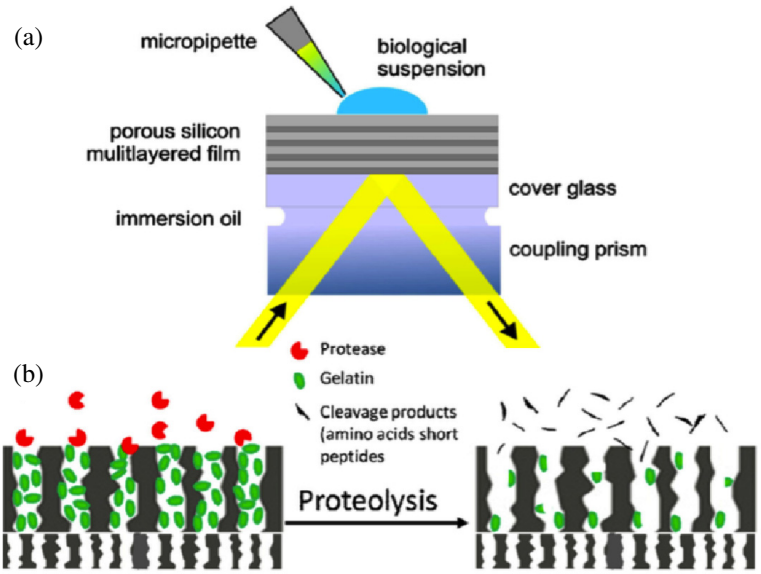

Figure 5. (a) Diagram of the porous silicon Bloch surface wave biosensor with a prism for light coupling. (b) Illustration of the open sensing space of the surface wave layer enable easy access for protease and fast out-diffusion of gelatin cleavage products [76] Copyright 2015 Elsevier.

layer [78, 79]. The grating was realized with polymer (photoresist) by electron beam lithography on porous silicon surface. Similar to the grating-coupled sensor, a ring-patterned porous silicon waveguide was reported to detect nucleic acid molecules [80]. The resonance shift on transmission spectrum of the waveguide was used to quantify the attachment of nucleic acids on the modified surfaces with a high quality factor (up to 10,000 indicating high sensitivity).

2.3.4. Other Advances in Biosensing

Directly utilizing the optical properties of porous silicon to engineer high sensitive optical biosensors is the current status in porous silicon-based optical biosensing. In addition to the efforts that have been summarized above, many researchers have been exploring the possibilities of porous silicon hybrid structure (such as porous silicon with polymers, quantum dots, metal nanoparticles, etc.) in optical biosensing. For instance, Voelcker and his group have established a few high-sensitive biosensors based on fluorescence enhancement of porous silicon microcavity [81–84]. Fluorescent molecules were incorporated into porous silicon structure as signal transducers. With the cavity mode to confine and enhance fluorescence, the detection limit of porous silicon microcavity biosensors has been greatly improved [85]. In addition, the fluorescent enhancement was also observed on porous silicon Bragg reflector, which in combined with quantum dots was also utilized in the detection of parasitic disease [86]. In spite of the fact that porous silicon functions as a medium for signal enhancement rather than a signal transducer in these cases, the hybrid concept might boost future bio-applications of porous silicon.

3. SILICON QUANTUM DOTS

Reducing the size of silicon in three dimensions yields nanocrystals which are also called quantum dots and they are endowed with unique photophysical properties. Examples of these properties include increased inter band transition probabilities, size-tunable emissions and high stability against photo bleaching. These properties have allowed developments of new applications with quantum dots, including next generation optoelectronic devices, solar cells and fluorescent bioimaging/sensing agents [87, 88]. For bio-application of quantum dots, material safety has been a major concern, because conventional quantum dots are typically made from heavy metal materials which are known to be toxic and for this reason a biologically benign alternative is in urgent need. The earth abundant material, silicon, is nontoxic as crystalline silicon naturally oxidizes in air and degrades to orthosilicic acid when water is present, and therefore an ideal candidate to meet this need. This section covers the optical biosensing and imaging applications using silicon quantum dots. This part begins by first describing the quantum confinement model in the case of silicon; then proceeds to the preparation strategies where the importance of surface chemistry will be highlighted; this section completes by covering the use of silicon quantum dots for biosensing and imaging applications. Note that since there have been several excellent full reviews on this topic in the past several years [3, 6, 7, 89–91], we aim at providing a tutorial typed guide to the frontier of the area, instead of a thorough history of the entire field.

3.1. Quantum Confinement in the Case of Silicon

With the physical dimensions of materials transition from the classic to the quantum regime, the quantum confinement effect occurs [87]. In the case of semiconductor nanocrystals, unique changes are seen in their band structures, and this is reflected in their distinct optical signatures. The first and foremost is that the energy levels of quantum dots become discrete instead of continuum. The inter band transition of electrons across these discrete energy levels is a radiative process, which yields photon emission of equivalent energy. Therefore, the degree of quantum confinement, which is related to the recombination rates, lifetime, quantum yield and emission wavelength becomes directly determined by the size of the particle.

$$\Delta E = E_g + \Sigma_{i=x,y,z} \frac{h^2 \pi^2 n_i^2}{2 d_i^2} \left(\frac{1}{m_e} + \frac{1}{m_h} \right) \tag{1}$$

To illustrate, in an idealized scenario, such as a cubic dot with infinite potential barriers, the separation of energy states of the exciton can be described as the above equation. Here E_g is the sum

of the band gap energy of the host material; d_i is the size of the quantum dots in the ith dimension; n is the quantum number; m_e and m_h are the effective mass of the conduction band electrons and valence band holes. For bulk materials (i.e., d_i is large), the confinement energies are small; however when the material dimension shrinks such that $1/d$ is no longer negligible, the confinement energy term becomes significant [87]. This is important as this relationship suggests that fine tuning the size of quantum dots can lead to significant change in their photophysical properties. This is very encouraging because the tuning of sizes can be achieved fairly easily in practice, which is usually done through colloidal chemistry or other fabrication techniques [92]. Another feature was the strong impact of doping on the optoelectronic properties of silicon nanocrystals, as evidenced from reported cases of both traditional dopants (B and P) and metals (Mn, Ni, Co, Cu) dopants [93, 94].

A feature that is distinct for silicon quantum dots is that bulk silicon is an indirect band-gap semiconductor [97]. What this means is that in the Brillouin zone of silicon, there exists a momentum difference within the crystal lattice (i.e., k-vector) between the minimum energy state of the conduction band and the maximum energy state of the valence band. This band mismatch leads to several consequences. The first is that the probability of inter band transition is much lower for bulk silicon. Usually the emission quantum yield of nanocrystals from confinement state is much lower compared with their direct bandgap counterparts [87, 98], while some exceptions have been observed [99, 100]. The second is that a lattice matched barrier layer used in heavy metal dots, is absent for most preparation methods. The missing of lattice matched shell structures indicate the interfacial chemistry becomes highly impactful to the electron transfer processes at the surface for silicon quantum dots and hence optical properties [101]. For instance, Figure 6 shows a recent study by the Veinot group showing simply changing the surface capping groups of silicon quantum dots of the same size leads to severe alternation to the emission of nanoparticles [95, 96], an observation uncommon for conventional quantum dots where emission wavelength is usually tuned by particle size.

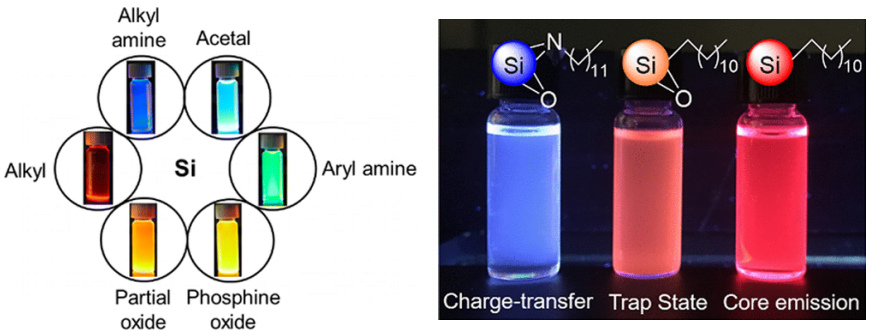

Figure 6. Impacts of different surface groups on the optical properties of silicon quantum dots. Reprints with permission from Veinot et al. [95, 96]. Copyrights 2014 & 2017 American Chemical Society.

3.2. Preparation and the Impacts of Surface Chemistry

We next discuss the preparation and surface modification methods for silicon quantum dots. In general there are three classes of methods of fabricating silicon quantum dots: the top-down approach, the bottom-up approach and combination of the both. As the name suggests, the top down based methods break down large pieces of bulk silicon into small, nano-sized crystals. The most common way of doing so is through acid based etching. In reality this is often done by hydrofluoric acid [102–104], sometimes with aid of applied electrochemical potentials [102, 105, 106]. One advantage of performing a top down approach is that these methods offer good tunability of color via control of particle size. Usually with etching based methods, color tuning can be achieved to cover the entire visible to near infrared range [102]. The near infrared emission is particularly attractive as it allows penetration of biological tissues where scattering signals and absorption level is at minimal, a feature that is unachievable with visible emissions [107]. An emerging recent method of the top down method of preparing silicon quantum dots is fast modifications [108–110]. This comes as a need from two factors: the first being that silicon naturally oxidize and unmodified particles lose fluorescence over time, and the second most of these

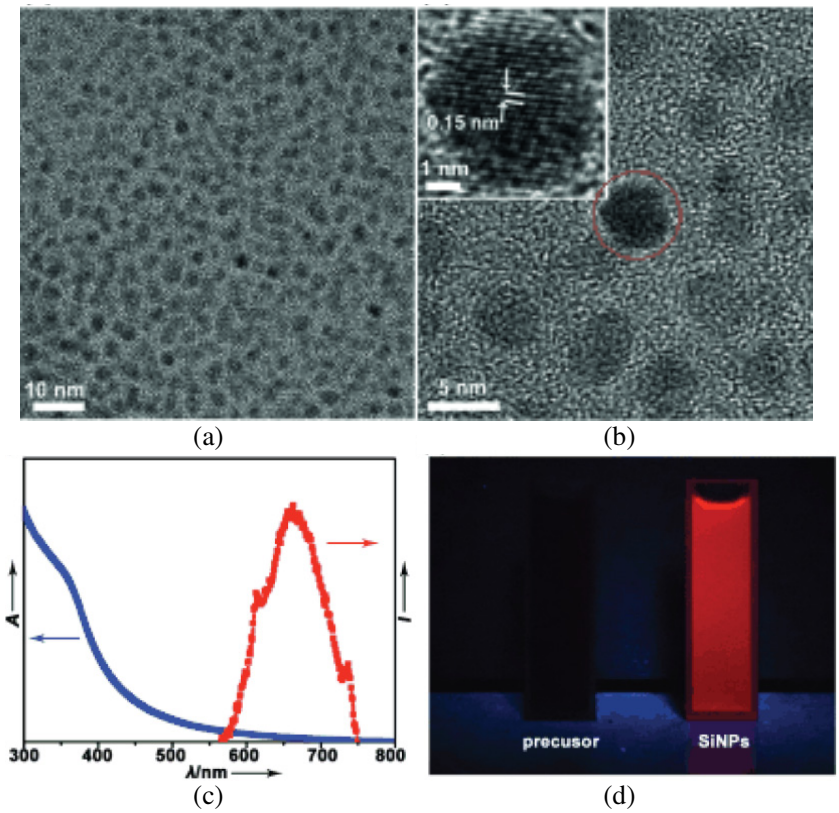

Figure 7. Microwave assisted synthesis of red emitting silicon nanoparticles showing high monodispersity. Figure adapted from Zhong et al. [108] Copyright Wiley 2012.

surface treatment methods are slow, represented by the hydrosilylation reaction approach [111]. As can be see in Figure 7, silicon nanoparticles were prepared by first using etching to obtain silicon nanowires, then microwave is applied to facilitates attachment of hydrophilic proteins, which kept the red fluorescence from particles, and the procedures only took very short time to complete, which is highly desirable for imaging probes [108, 112]. Another example was shown by the Veinot group which uses xenon difluoride which results in instantaneous attachment of surface molecules.

The bottom up approach works entirely the opposite by using self-assembly principles to build nanocrystals up from small chemical building blocks. Early examples belonging to this approach is the use of precipitation reaction using molecular precursors [113, 114]. Early examples were represented by the use of Zintl salts which yield blue fluorescent silicon nanocrystals [114–116]. Unsurprisingly, the use of surfactants seems to improve the monodispersity of the particles when a high quality micelle is formed, as demonstrated by the reduction of halogenated silane aided by phase transfer agents [101, 113, 117–120]. One of the key advantage of using a bottom up approach is that these methods are highly comparable to typical fabrication methods of conventional quantum dots. The solution based approach also allows wet chemistry and surface modifications to be done with ease, and this is important when introducing functionalities.

There is increasing evidence to suggest that the blue emission is governed by electron transfer from the surface states [96, 121]. Remarkably, recent observations suggest that certain electron donating, nitrogen containing species can drastically improve the blue emitting quantum yield of silicon quantum dots significantly. i.e., Li et al. showed that performing only surface modification with electron donors containing nitrogen drastically improves the emission quantum yield of SiQDs to 50–75% [122]. (Figure 8) This surprising finding was proven by the Sun group [123], where ultrafast microscopy was used to indicate that this high quantum yield emission is indeed related to the surface trap states. Using comparable concepts, the Jin group has successfully synthesized highly bright silicon nanoparticles, with

Figure 8. Highly luminescent silicon quantum dots by attaching electron rich, nitrogen containing ring species with quantum yield exceeding 75%. Reprints with permission from Li et al. [112] Copyrights 2013 American Chemical Society.

quantum yield exceeds 90% [124], a current record number for silicon quantum dots. This significant progress has opened many new opportunities of using silicon quantum dots for practical applications where brightness of emission is among one of the most important parameters in the performance of the final application.

Among the third approach which is combination of the previous two methods non-thermal plasma assisted approach has distincted itself as being a prominent method for fabrication [99, 125–129]. In principle, the plasma environment initiates the breaking down of silane precursors in the reaction chamber. Through controlling interactions of the charged clusters within the plasma, silicon nanocrystals with a broad range can be fabricated. This approach has been shown to be particularly attractive in performing doping experiments with silicon nanocrystals [94, 130]. For instance, using non-thermal plasma synthesis, Kortshagen and coworkers have recently fabricated heavily doped boron and phosphorous doped silicon nanocrystals showing surface plasmons in the region of mid infrared [94, 130]. This is particularly useful when the target applications are electronic and photonic devices, although it would be interesting to see whether the localized plasmon wavelength can be decreased to the near infrared region where bio-applications usually relies on.

3.3. Bio-Applications

The developments of materials preparation and surface modifications strategies of silicon quantum dots are exciting. What is even more appealing is that these studies have paved way for their practical application in nanomedicine. This section covers the rapidly expanding field of using silicon quantum dots for biosensing and imaging applications [90], highlighting recent progresses.

A biosensor is an analytical device which performs biochemical measurements. For fluorescent quantum dots, the most common approach to develop a new sensor is by controlling the emission properties of the particles via controlling the interfacial properties. For example, this can be done by controlling the energy transfer processes, such as Förster resonance energy transfer (FRET) by changing the surface ligands to respond specifically to the target analyte. FRET is a dipole-dipole coupling process seen between spatially adjacent fluorophores with high spectral overlaps and aligned dipoles. When FRET effect occurs between two or more fluorophores, what is observed is the decrease of the donor fluorescence intensity, and increase of the acceptor fluorescence. Since this effect is highly distance dependent, it has lead to the development of various highly accurate biochemical assays [131]. FRET can be used to probe the interactions between proteins, protein domains, nucleic acids and it has become a standard method in probing such interactions within intracellular contexts [132–134]. In the case of silicon quantum dots, detection of small molecule analytes have been achieved, such as TNT [135], dopamine [136], and certain metal ions [137], while measurement of macromolecular activities was seen only recently [138]. Cheng et al. showed that attaching a short dye labelled peptide to silicon quantum dots, with thiol-ene 'click' chemistry allowed the generation of FRET between silicon quantum dots and the dye whose intensity is enzyme responsive [101, 139, 140]. By this way they were able to for the first time control the surface to allow measurement of protease activity using colloidal silicon quantum dots. This advance allowed new opportunities of using silicon nanocrystals in biosensing, in particular sensing of macromolecular activities, which has been so predominant in the quantum dots sensing community [141, 142].

Silicon quantum dots have shown multiple applications in conventional fluorescent imaging and labelling settings [3, 6, 110]. Another interesting new frontier was using the particles as theranostic platforms [144, 145]. In the example of Ji et al., for the first time that silicon quantum dots were shown to be used as drug carrier (Figure 9) [146]. This work is important as it opened a new research area of combining the imaging and delivery modality into one nontoxic platform of silicon nanocrystals [146]. Another trend for the field is the incorporation of advanced fluorescence imaging to enhance the capability of particles in fluorescent imaging [120, 147]. Cheng et al. used fluorescence lifetime imaging, which is commonly used by the biophysical community, into the study of silicon quantum dots [120]. It was shown that although the predominant reduction methods yield particles with blue fluorescence which is nearly unresolvable by conventional fluorescence techniques due to their biological backgrounds, lifetime based imaging techniques can easily de-convolute the signal in both one photon and two photon channels and the technique is applicable to FRET systems. This indicates suggest that the when material preparation strategy reaches its limit, other well developed imaging method can assist in the development of new probes and together this will make one step further in developing the probes in practical applications.

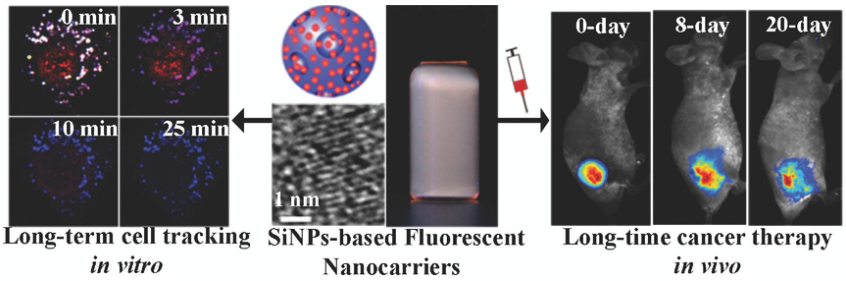

Figure 9. Silicon quantum dots as theranostic platforms. Shown above as using particles for both fluorescent imaging and drug carrier in in vivo environments. Reprints with permission from Ji et al. [143] Copyrights Wiley 2015.

4. CONCLUSIONS

The past two decades have seen remarkable progress in using crystalline silicon micro and nanostructures in biosensing and bioimaging applications. This review article tends to provide a tutorial typed guide to the frontiers using two representative structures of porous silicon and silicon quantum dots. In light of the remarkable optical properties, porous silicon offers a good prospect for bio-applications. The recent advances in establishing better biological recognition interfaces, enhancing signal processing, developing new sensing strategies and adopting novel optical structures (such as Bloch surface wave) have shown promises in achieving high-sensitivity sensing performance on porous silicon. Especially, the integration of other materials or sensing techniques into porous silicon to form hybrid structure with dual or multiple properties has expanded the potential with porous silicon. In the case of silicon quantum dots, the charge confinement effect leads to significant increase of emission quantum yield. Due to the chemical nature of silicon, the interfacial property of particle surface chemistry is particularly influential over their optical properties. There has been much focus on these surface effects, and by using comparable silicon surface modification techniques, the emission properties have been tuned via mechanisms such as FRET. We foresee that the new frontiers brought by the two types of nanostructures will open new opportunities in using these materials for more application in nanomedicine.

REFERENCES

1. Priolo, F., T. Gregorkiewicz, M. Galli, and T. F. Krauss, "Silicon nanostructures for photonics and photovoltaics," *Nature Nanotechnology*, Vol. 9, 19, 2014.

2. Chan, C. K., H. Peng, G. Liu, K. McIlwrath, X. F. Zhang, R. A. Huggins, and Y. Cui, "High-performance lithium battery anodes using silicon nanowires," *Nature Nanotechnology*, Vol. 3, 31, 2007.
3. Peng, F., Y. Su, Y. Zhong, C. Fan, S.-T. Lee, and Y. He, "Silicon nanomaterials platform for bioimaging, biosensing, and cancer therapy," *Accounts Chem. Res.*, Vol. 47, No. 2, 612–623, 2014.
4. Liu, J., F. Erogbogbo, K.-T. Yong, L. Ye, J. Liu, R. Hu, H. Chen, Y. Hu, Y. Yang, J. Yang, I. Roy, N. A. Karker, M. T. Swihart, and P. N. Prasad, "Assessing clinical prospects of silicon quantum dots: Studies in mice and monkeys," *ACS Nano*, 2013.
5. Anglin, E. J., L. Y. Cheng, W. R. Freeman, and M. J. Sailor, "Porous silicon in drug delivery devices and materials," *Adv. Drug Deliver Rev.*, Vol. 60, No. 11, 1266–1277, 2008.
6. Cheng, X., S. B. Lowe, P. J. Reece, and J. J. Gooding, "Colloidal silicon quantum dots: From preparation to the modification of self-assembled monolayers (SAMs) for bio-applications," *Chem. Soc. Rev.*, Vol. 43, No. 8, 2680–700, 2014.
7. McVey, B. F. and R. D. Tilley, "Solution synthesis, optical properties, and bioimaging applications of silicon nanocrystals," *Acc Chem. Res.*, Vol. 47, No. 10, 3045-51, 2014.
8. Uhlir, A., "Electrolytic Shaping of Germanium and silicon," *Bell Syst. Tech. J.*, Vol. 35, 333–347, 1956.
9. Canham, L. T., "Silicon quantum wire array fabrication by electrochemical and chemical dissolution of wafers," *Appl. Phys. Lett.*, Vol. 57, No. 10, 1046–1048, 1990.
10. Lin, V. S.-Y., K. Motesharei, K.-P. S. Dancil, M. J. Sailor, and M. R. Ghadiri, "A porous silicon-based optical interferometric biosensor," *Science*, Vol. 278, No. 5339, 840–843, 1997.
11. Jane, A., R. Dronov, A. Hodges, and N. H. Voelcker, "Porous silicon biosensors on the advance," *Trends Biotechnol.*, Vol. 27, 230–239, 2009.
12. Dhanekar, S. and S. Jain, "Porous silicon biosensor: Current status," *Biosensors and Bioelectronics*, Vol. 41, 54–64, 2013.
13. Harraz, F. A., "Porous silicon chemical sensors and biosensors: A review," *Sensors and Actuators B: Chemical*, Vol. 202, 897–912, 2014.
14. Shtenberg, G. and E. Segal, "Porous silicon optical biosensors," *Handbook of Porous Silicon*, L. Canham (ed.), 857–868, Springer, Switzerland, 2014.
15. Fathauer, R. W., T. George, A. Ksendzov, and R. P. Vasquez, "Visible luminescence from silicon wafers subjected to stain etches," *Applied Physics Letters*, Vol. 60, No. 8, 995–997, 1992.
16. Pyatilova, O. V., S. A. Gavrilov, Y. I. Shilyaeva, A. A. Pavlov, Y. P. Shaman, and A. A. Dudin, "Influence of the doping type and level on the morphology of porous Si formed by galvanic etching," *Semiconductors*, Vol. 51, No. 2, 173–177, 2017.
17. Li, X. and P. W. Bohn, "Metal-assisted chemical etching in HF/H_2O_2 produces porous silicon," *Applied Physics Letters*, Vol. 77, No. 16, 2572–2574, 2000.
18. Balderas-Valadez, R. F., V. Agarwal, and C. Pacholski, "Fabrication of porous silicon-based optical sensors using metal-assisted chemical etching," *RSC Advances*, Vol. 6, No. 26, 21430–21434, 2016.
19. Zhao, M., R. Balachandran, J. Allred, and M. Keswani, "Synthesis of porous silicon through interfacial reactions and measurement of its electrochemical response using cyclic voltammetry," *RSC Advances*, Vol. 5, No. 96, 79157–79163, 2015.
20. Rauscher, M. and H. Spohn, "Porous silicon formation and electropolishing," *Physical Review E*, Vol. 64, No. 3, 031604, 2001.
21. Canham, L., "Porous silicon formation by anodisation," *Properties of Porous Silicon*, Halimaoui, A., Ed., IEE, London; L. Canham Ed., Vol. 1, 12, 1997.
22. Cullis, A., L. Canham, and P. Calcott, "The structural and luminescence properties of porous silicon," *J. Appl. Phys.*, Vol. 82, 909, 1997.
23. Theiss, W., "Optical properties of porous silicon," *Surf. Sci. Rep.*, Vol. 29, 91–192, 1997.
24. Vincent, G., "Optical properties of porous silicon superlattices," *Appl. Phys. Lett.*, Vol. 64, 2367, 1994.

25. Mazzoleni, C. and L. Pavesi, "Application to optical components of dielectric porous silicon multilayers," *Appl. Phys. Lett.*, Vol. 67, No. 20, 2983–2985, 1995.
26. Berger, M. G., C. Dieker, M. Thoenissen, L. Vescan, H. Lueth, H. Muender, W. Theiss, M. Wernke, and P. Grosse, "Porosity superlattices: a new class of Si heterostructures," *J. Phys. D*, Vol. 27, No. 6, 1333, 1994.
27. Berger, M. G., M. Thoenissen, R. Arens-Fischer, H. Münder, H. Lüth, M. Arntzen, and W. Thei, "Investigation and design of optical properties of porosity superlattices," *Thin Solid Films*, Vol. 255, Nos. 1–2, 313–316, 1995.
28. Frohnhoff, S. and M. G. Berger, "Porous silicon superlattices," *Adv. Mater.*, Vol. 6, No. 12, 963–965, 1994.
29. Pellegrini, V., A. Tredicucci, C. Mazzoleni, and L. Pavesi, "Enhanced optical properties in porous silicon microcavities," *Physical Review B*, Vol. 52, No. 20, R14328, 1995.
30. Pavesi, L., C. Mazzoleni, A. Tredicucci, and V. Pellegrini, "Controlled photon emission in porous silicon microcavities," *Appl. Phys. Lett.*, Vol. 67, No. 22, 3280–3282, 1995.
31. Lorenzo, E., C. J. Oton, N. E. Capuj, M. Ghulinyan, D. Navarro-Urrios, Z. Gaburro, and L. Pavesi, "Porous silicon-based rugate filters," *Appl. Opt.*, Vol. 44, No. 26, 5415–5421, 2005.
32. Berger, M. G., R. Arens-Fischer, M. Thönissen, M. Krüger, S. Billat, H. Lüth, S. Hilbrich, W. Theiß, and P. Grosse, "Dielectric filters made of PS: advanced performance by oxidation and new layer structures," *Thin Solid Films*, Vol. 297, Nos. 1–2, 237–240, 1997.
33. Arrand, H. F., T. M. Benson, P. Sewell, A. Loni, R. J. Bozeat, R. Arens-Fischer, M. Kruger, M. Thonissen, and H. Luth, "The application of porous silicon to optical waveguiding technology," *IEEE Journal of Selected Topics in Quantum Electronics*, Vol. 4, No. 6, 975–982, 1998.
34. Loni, A., L. T. Canham, M. G. Berger, R. Arens-Fischer, H. Munder, H. Luth, H. F. Arrand, and T. M. Benson, "Porous silicon multilayer optical waveguides," *Thin Solid Films*, Vol. 276, Nos. 1–2, 143–146, 1996.
35. Nagata, S., C. Domoto, T. Nishimura, and K. Iwameji, "Single-mode optical waveguide fabricated by oxidization of selectively doped titanium porous silicon," *Appl. Phys. Lett.*, Vol. 72, No. 23, 2945–2947, 1998.
36. Ferrand, P., R. Romestain, and J. C. Vial, "Photonic band-gap properties of a porous silicon periodic planar waveguide," *Physical Review B*, Vol. 63, No. 11, 115106, 2001.
37. Sapienza, R., P. Costantino, D. Wiersma, M. Ghulinyan, C. J. Oton, and L. Pavesi, "Optical analogue of electronic bloch oscillations," *Phys. Rev. Lett.*, Vol. 91, No. 26, 263902, 2003.
38. Guillermain, E., V. Lysenko, R. Orobtchouk, T. Benyattou, S. Roux, A. Pillonnet, and P. Perriat, "Bragg surface wave device based on porous silicon and its application for sensing," *Appl. Phys. Lett.*, Vol. 90, No. 24, 241116-3, 2007.
39. Dal Negro, L., C. J. Oton, Z. Gaburro, L. Pavesi, P. Johnson, A. Lagendijk, R. Righini, M. Colocci, and D. S. Wiersma, "Light transport through the band-edge states of fibonacci quasicrystals," *Phys. Rev. Lett.*, Vol. 90, No. 5, 055501, 2003.
40. Sailor, M. J. and E. C. Wu, "Photoluminescence-based sensing with porous silicon films, microparticles, and nanoparticles," *Advanced Functional Materials*, Vol. 19, No. 20, 3195–3208, 2009.
41. Spanier, J. E. and I. P. Herman, "Use of hybrid phenomenological and statistical effective-medium theories of dielectric functions to model the infrared reflectance of porous SiC films," *Physical Review B*, Vol. 61, No. 15, 10437, 2000.
42. Syshchyk, O., V. A. Skryshevsky, O. O. Soldatkin, and A. P. Soldatkin, "Enzyme biosensor systems based on porous silicon photoluminescence for detection of glucose, urea and heavy metals," *Biosensors and Bioelectronics*, Vol. 66, 89–94, 2015.
43. Melnyk, Y., K. Pavlova, V. Myndrul, R. Viter, V. Smyntyna, and I. Iatsunskyi, "Porous silicon photoluminescence biosensor for rapid and sensitive detection of toxins," *SPIE Organic Photonics+ Electronics*, 6, SPIE, 2017.

44. Myndrul, V., R. Viter, M. Savchuk, M. Koval, N. Starodub, V. Silamikelis, V. Smyntyna, A. Ramanavicius, and I. Iatsunskyi, "Gold coated porous silicon nanocomposite as a substrate for photoluminescence-based immunosensor suitable for the determination of Aflatoxin B1," *Talanta*, Vol. 175, 297–304, 2017.
45. Park, J.-H., L. Gu, G. von Maltzahn, E. Ruoslahti, S. N. Bhatia, and M. J. Sailor, "Biodegradable luminescent porous silicon nanoparticles for in vivo applications," *Nat. Mater.*, Vol. 8, No. 4, 331–336, 2009.
46. Gu, L., D. J. Hall, Z. Qin, E. Anglin, J. Joo, D. J. Mooney, S. B. Howell, and M. J. Sailor, "In vivo time-gated fluorescence imaging with biodegradable luminescent porous silicon nanoparticles," *Nature Communication*, Vol. 4, 2326, 2013.
47. Kim, D., J. Kang, T. Wang, H. G. Ryu, J. M. Zuidema, J. Joo, M. Kim, Y. Huh, J. Jung, K. H. Ahn, K. H. Kim, and M. J. Sailor, "Two-photon in vivo imaging with porous silicon nanoparticles," *Adv. Mater.*, Vol. 29, No. 39, 1703309, 2017.
48. Chen, X., F. Wo, Y. Jin, J. Tan, Y. Lai, and J. Wu, "Drug-porous silicon dual luminescent system for monitoring and inhibition of wound infection," *ACS Nano*, Vol. 11, No. 8, 7938–7949, 2017.
49. Song, S., L. Wang, J. Li, C. Fan, and J. Zhao, "Aptamer-based biosensors," *TrAC Trends in Analytical Chemistry*, Vol. 27, No. 2, 108–117, 2008.
50. Urmann, K., J.-G. Walter, T. Scheper, and E. Segal, "Label-free optical biosensors based on aptamer-functionalized porous silicon scaffolds," *Analytical Chemistry*, Vol. 87, No. 3, 1999–2006, 2015.
51. Hamula, C. L. A., H. Zhang, F. Li, Z. Wang, X. Chris Le, and X.-F. Li, "Selection and analytical applications of aptamers binding microbial pathogens," *TrAC Trends in Analytical Chemistry*, Vol. 30, No. 10, 1587–1597, 2011.
52. Urmann, K., S. Arshavsky-Graham, J. G. Walter, T. Scheper, and E. Segal, "Whole-cell detection of live lactobacillus acidophilus on aptamer-decorated porous silicon biosensors," *Analyst*, Vol. 141, No. 18, 5432–5440, 2016.
53. Tombelli, S., M. Minunni, and M. Mascini, "Analytical applications of aptamers," *Biosensors and Bioelectronics*, Vol. 20, No. 12, 2424–2434, 2005.
54. Mairal, T., V. Cengiz Ozalp, P. Lozano Sánchez, M. Mir, I. Katakis, and C. K. O'Sullivan, "Aptamers: molecular tools for analytical applications," *Analytical and Bioanalytical Chemistry*, Vol. 390, No. 4, 989–1007, 2008.
55. Kirsch, J., C. Siltanen, Q. Zhou, A. Revzin, and A. Simonian, "Biosensor technology: recent advances in threat agent detection and medicine," *Chemical Society Reviews*, Vol. 42, No. 22, 8733–8768, 2013.
56. Chhasatia, R., M. J. Sweetman, F. J. Harding, M. Waibel, T. Kay, H. Thomas, T. Loudovaris, and N. H. Voelcker, "Non-invasive, in vitro analysis of islet insulin production enabled by an optical porous silicon biosensor," *Biosensors and Bioelectronics*, Vol. 91, 515–522, 2017.
57. Urmann, K., P. Reich, J.-G. Walter, D. Beckmann, E. Segal, and T. Scheper, "Rapid and label-free detection of protein a by aptamer-tethered porous silicon nanostructures," *Journal of Biotechnology*, Vol. 257, 171–177, 2017.
58. Tenenbaum, E. and E. Segal, "Optical biosensors for bacteria detection by a peptidomimetic antimicrobial compound," *Analyst*, Vol. 140, No. 22, 7726–7733, 2015.
59. Naveas, N., J. Hernandez-Montelongo, R. Pulido, V. Torres-Costa, R. Villanueva-Guerrero, J. P. García Ruiz, and M. Manso-Silván, "Fabrication and characterization of a chemically oxidized-nanostructured porous silicon based biosensor implementing orienting protein A," *Colloids and Surfaces B: Biointerfaces*, Vol. 115, 310–316, 2014.
60. Mariani, S., L. M. Strambini, and G. Barillaro, "Femtomole detection of proteins using a label-free nanostructured porous silicon interferometer for perspective ultrasensitive biosensing," *Analytical Chemistry*, Vol. 88, No. 17, 8502–8509, 2016.
61. Mariani, S., L. Pino, L. M. Strambini, L. Tedeschi, and G. Barillaro, "10 000-fold improvement in protein detection using nanostructured porous silicon interferometric aptasensors," *ACS Sensors*, Vol. 1, No. 12, 1471–1479, 2016.

62. Mariani, S., L. M. Strambini, L. Tedeschi, and G. Barillaro, "Interferogram average over wavelength spectroscopy: An ultrasensitive technique for biosensing with porous silicon interferometers," *ECS Transactions*, Vol. 77, No. 11, 1815–1823, 2017.
63. Vilensky, R., M. Bercovici, and E. Segal, "Oxidized porous silicon nanostructures enabling electrokinetic transport for enhanced DNA detection," *Adv. Funct. Mater.*, Vol. 25, No. 43, 6725–6732, 2015.
64. Arshavsky-Graham, S., R. Vilenski, F. Faratore, M. Bercovici, and E. Segal, "1,000-fold sensitivity enhancement of porous Si-based optical biosensors for nucleic acid and proteins detection," *Optics in the Life Sciences Congress, Optical Society of America*, OmM4D.6, 2017.
65. Nair, P. R. and M. A. Alam, "Performance limits of nanobiosensors," *Applied Physics Letters*, Vol. 88, No. 23, 233120, 2006.
66. Sheehan, P. E. and L. J. Whitman, "Detection limits for nanoscale biosensors," *Nano Letters*, Vol. 5, No. 4, 803–807, 2005.
67. Kumar, N., E. Froner, R. Guider, M. Scarpa, and P. Bettotti, "Investigation of non-specific signals in nanoporous flow-through and flow-over based sensors," *Analyst*, Vol. 139, No. 6, 1345–1349, 2014.
68. Zhao, Y., G. Gaur, S. T. Retterer, P. E. Laibinis, and S. M. Weiss, "Flow-through porous silicon membranes for real-time label-free biosensing," *Analytical Chemistry*, Vol. 88, No. 22, 10940–10948, 2016.
69. Gupta, B., K. Mai, S. B. Lowe, D. Wakefield, N. Di Girolamo, K. Gaus, P. J. Reece, and J. J. Gooding, "Ultrasensitive and specific measurement of protease activity using functionalized photonic crystals," *Analytical Chemistry*, Vol. 87, No. 19, 9946–9953, 2015.
70. Soeriyadi, A. H., B. Gupta, P. J. Reece, and J. J. Gooding, "Optimising the enzyme response of a porous silicon photonic crystal via the modular design of enzyme sensitive polymers," *Polymer Chemistry*, Vol. 5, No. 7, 2333–2341, 2014.
71. Rong, G., J. D. Ryckman, R. L. Mernaugh, and S. M. Weiss, "Label-free porous silicon membrane waveguide for DNA sensing," *Applied Physics Letters*, Vol. 93, No. 16, 161109, 2008.
72. Rong, G., A. Najmaie, J. E. Sipe, and S. M. Weiss, "Nanoscale porous silicon waveguide for label-free DNA sensing," *Biosensors and Bioelectronics*, Vol. 23, No. 10, 1572–1576, 2008.
73. Wei, X., C. Kang, M. Liscidini, G. Rong, S. T. Retterer, M. Patrini, J. E. Sipe, and S. M. Weiss, "Grating couplers on porous silicon planar waveguides for sensing applications," *J. Appl. Phys.*, Vol. 104, No. 12, 123113, 2008.
74. Wei, X. and S. M. Weiss, "Guided mode biosensor based on grating coupled porous silicon waveguide," *Opt. Express*, Vol. 19, 2011.
75. Wei, X., J. W. Mares, Y. D. Gao, D. Li, and S. M. Weiss, "Biomolecule kinetics measurements in flow cell integrated porous silicon waveguides," *Biomed. Opt. Express*, Vol. 3, 2012.
76. Qiao, H., A. H. Soeriyadi, B. Guan, P. J. Reece, and J. J. "Gooding, The analytical performance of a porous silicon Bloch surface wave biosensors as protease biosensor," *Sensors and Actuators B: Chemical*, Vol. 211, No. Supplement C, 469–475, 2015.
77. Zhao, Y., G. A. Rodriguez, Y. M. Graham, T. Cao, G. Gaur, and S. M. Weiss, "Resonant photonic structures in porous silicon for biosensing," *SPIE BiOS*, 10, 2017.
78. Rodriguez, G. A., J. D. Ryckman, Y. Jiao, and S. M. Weiss, "A size selective porous silicon grating-coupled Bloch surface and sub-surface wave biosensor," *Biosensors and Bioelectronics*, Vol. 53, 486–493, 2014.
79. Rodriguez, G. A., J. D. Lonai, R. L. Mernaugh, and S. M. Weiss, "Porous silicon Bloch surface and sub-surface wave structure for simultaneous detection of small and large molecules," *Nanoscale Research Letters*, Vol. 9, No. 1, 383, 2014.
80. Rodriguez, G. A., S. Hu, and S. M. Weiss, "Porous silicon ring resonator for compact, high sensitivity biosensing applications," *Opt. Express*, Vol. 23, No. 6, 7111–7119, 2015.

81. Krismastuti, F. S. H., S. Pace, and N. H. Voelcker, "Porous silicon resonant microcavity biosensor for matrix metalloproteinase detection," *Advanced Functional Materials*, Vol. 24, No. 23, 3639–3650, 2014.
82. Jenie, S. N. A., Z. Du, S. J. P. McInnes, P. Ung, B. Graham, S. E. Plush, and N. H. Voelcker, "Biomolecule detection in porous silicon based microcavities via europium luminescence enhancement," *Journal of Materials Chemistry B*, Vol. 2, No. 44, 7694–7703, 2014.
83. Jenie, S. N. A., B. Prieto-Simon, and N. H. Voelcker, "Development of l-lactate dehydrogenase biosensor based on porous silicon resonant microcavities as fluorescence enhancers," *Biosensors and Bioelectronics*, Vol. 74, 637–643, 2015.
84. Krismastuti, F. S. H., A. Cavallaro, B. Prieto-Simon, and N. H. Voelcker, "Toward multiplexing detection of wound healing biomarkers on porous silicon resonant microcavities," *Advanced Science*, Vol. 3, No. 6, 1500383, 2016.
85. Jenie, S. N. A., S. E. Plush, and N. H. Voelcker, "Recent advances on luminescent enhancement-based porous silicon biosensors," *Pharmaceutical Research*, Vol. 33, No. 10, 2314–2336, 2016.
86. Li, Y., Z. Jia, G. Lv, H. Wen, P. Li, H. Zhang, and J. Wang, "Detection of Echinococcus granulosus antigen by a quantum dot/porous silicon optical biosensor," *Biomed. Opt. Express*, Vol. 8, No. 7, 3458–3469, 2017.
87. Alivisatos, A. P., "Perspectives on the physical chemistry of semiconductor nanocrystals," *J. Phys. Chem.-Us*, Vol. 100, No. 31, 13226–13239, 1996.
88. Bruchez, M., M. Moronne, P. Gin, S. Weiss, and A. P. Alivisatos, "Semiconductor nanocrystals as fluorescent biological labels," *Science*, Vol. 281, No. 5385, 2013–2016, 1998.
89. Dasog, M., J. Kehrle, B. Rieger, and J. G. C. Veinot, "Silicon nanocrystals and silicon-polymer hybrids: Synthesis, surface engineering, and applications," *Angewandte Chemie International Edition*, Vol. 55, No. 7, 2322–2339, 2016.
90. Gonzalez, C. M. and J. G. C. Veinot, "Silicon nanocrystals for the development of sensing platforms," *J. Mater. Chem. C*, Vol. 4, No. 22, 4836–4846, 2016.
91. Su, Y., X. Ji, and Y. He, "Water-dispersible fluorescent silicon nanoparticles and their optical applications," *Adv. Mater.*, Vol. 28, No. 47, 10567–10574, 2016.
92. Peng, X., L. Manna, W. Yang, J. Wickham, E. Scher, A. Kadavanich, and A. P. Alivisatos, "Shape control of CdSe nanocrystals," *Nature*, Vol. 404, No. 6773, 59–61, 2000.
93. McVey, B. F. P., J. Butkus, J. E. Halpert, J. M. Hodgkiss, and R. D. Tilley, "Solution synthesis and optical properties of transition-metal-doped silicon nanocrystals," *The Journal of Physical Chemistry Letters*, Vol. 6, No. 9, 1573–1576, 2015.
94. Kramer, N. J., K. S. Schramke, and U. R. Kortshagen, "Plasmonic properties of silicon nanocrystals doped with boron and phosphorus," *Nano Letters*, Vol. 15, No. 8, 5597–5603, 2015.
95. Dasog, M., G. B. De los Reyes, L. V. Titova, F. A. Hegmann, and J. G. Veinot, "Size vs surface: tuning the photoluminescence of freestanding silicon nanocrystals across the visible spectrum via surface groups," *ACS Nano*, Vol. 8, No. 9, 9636-48, 2014.
96. Sinelnikov, R., M. Dasog, J. Beamish, A. Meldrum, and J. G. C. Veinot, "Revisiting an ongoing debate: What role do surface groups play in silicon nanocrystal photoluminescence?" *ACS Photonics*, Vol. 4, No. 8, 1920–1929, 2017.
97. Herman, F., "The electronic energy band structure of silicon and germanium," *Proceedings of the IRE*, Vol. 43, No. 12, 1703–1732, 1955.
98. Heath, J. R., "A liquid-solution-phase synthesis of crystalline silicon," *Science*, Vol. 258, No. 5085, 1131–1133, 1992.
99. Jurbergs, D., E. Rogojina, L. Mangolini, and U. Kortshagen, "Silicon nanocrystals with ensemble quantum yields exceeding 60%," *Appl. Phys. Lett.*, Vol. 88, No. 23, 2006.
100. Kelly, J. A., A. M. Shukaliak, M. D. Fleischauer, and J. G. Veinot, "Size-dependent reactivity in hydrosilylation of silicon nanocrystals," *J. Am Chem. Soc.*, Vol. 133, No. 24, 9564-71, 2011.

101. Cheng, X., R. Gondosiswanto, S. Ciampi, P. J. Reece, and J. J. Gooding, "One-pot synthesis of colloidal silicon quantum dots and surface functionalization via thiol-ene click chemistry," *Chem. Commun. (Camb)*, Vol. 48, No. 97, 11874-6, 2012.
102. Hessel, C. M., D. Reid, M. G. Panthani, M. R. Rasch, B. W. Goodfellow, J. Wei, H. Fujii, V. Akhavan, and B. A. Korgel, "Synthesis of ligand-stabilized silicon nanocrystals with size-dependent photoluminescence spanning visible to near-infrared wavelengths," *Chem. Mater.*, Vol. 24, No. 2, 393–401, 2012.
103. Locritani, M., Y. Yu, G. Bergamini, M. Baroncini, J. K. Molloy, B. A. Korgel, and P. Ceroni, "Silicon nanocrystals functionalized with pyrene units: efficient light-harvesting antennae with bright near-infrared emission," *J. Phys. Chem. Lett.*, Vol. 5, No. 19, 3325-9, 2014.
104. Yu, Y., C. M. Hessel, T. D. Bogart, M. G. Panthani, M. R. Rasch, and B. A. Korgel, "Room temperature hydrosilylation of silicon nanocrystals with bifunctional terminal alkenes," *Langmuir*, Vol. 29, No. 5, 1533–1540, 2013.
105. Kang, Z., C. H. A. Tsang, Z. Zhang, M. Zhang, N.-B. Wong, J. A. Zapien, Y. Shan, and S.-T. Lee, "A polyoxometalate-assisted electrochemical method for silicon nanostructures preparation: From quantum dots to nanowires," *Journal of the American Chemical Society*, Vol. 129, No. 17, 5326–5327, 2007.
106. Kang, Z. H., C. H. A. Tsang, N. B. Wong, Z. D. Zhang, and S. T. Lee, "Silicon quantum dots: A general photocatalyst for reduction, decomposition, and selective oxidation reactions," *Journal of the American Chemical Society*, Vol. 129, No. 40, 12090-+, 2007.
107. Hong, G., A. L. Antaris, and H. Dai, "Near-infrared fluorophores for biomedical imaging," *Nature Biomedical Engineering*, Vol. 1, 0010. 2017.
108. Zhong, Y., F. Peng, X. Wei, Y. Zhou, J. Wang, X. Jiang, Y. Su, S. Su, S.-T. Lee, and Y. He, "Microwave-assisted synthesis of biofunctional and fluorescent silicon nanoparticles using proteins as hydrophilic ligands," *Angewandte Chemie International Edition*, Vol. 51, No. 34, 8485–8489, 2012.
109. Zhong, Y., F. Peng, F. Bao, S. Wang, X. Ji, L. Yang, Y. Su, S.-T. Lee, and Y. He, "Large-scale aqueous synthesis of fluorescent and biocompatible silicon nanoparticles and their use as highly photostable biological probes," *Journal of the American Chemical Society*, Vol. 135, No. 22, 8350–8356, 2013.
110. He, Y., Y. L. Zhong, F. Peng, X. P. Wei, Y. Y. Su, Y. M. Lu, S. Su, W. Gu, L. S. Liao, and S. T. Lee, "One-pot microwave synthesis of water-dispersible, ultraphoto- and pH-stable, and highly fluorescent silicon quantum dots," *Journal of the American Chemical Society*, Vol. 133, No. 36, 14192–14195, 2011.
111. Hua, F., M. T. Swihart, and E. Ruckenstein, "Efficient surface grafting of luminescent silicon quantum dots by photoinitiated hydrosilylation," *Langmuir*, Vol. 21, No. 13, 6054–6062, 2005.
112. Zhong, Y., F. Peng, F. Bao, S. Wang, X. Ji, L. Yang, Y. Su, S.-T. Lee, and Y. He, "Large-scale aqueous synthesis of fluorescent and biocompatible silicon nanoparticles and their use as highly photostable biological probes," *Journal of the American Chemical Society*, 2013.
113. Tilley, R. D. and K. Yamamoto, "The microemulsion synthesis of hydrophobic and hydrophilic silicon nanocrystals," *Adv. Mater.*, Vol. 18, No. 15, 2053–2056, 2006.
114. Yang, C.-S., R. A. Bley, S. M. Kauzlarich, H. W. H. Lee, and G. R. Delgado, "Synthesis of alkyl-terminated silicon nanoclusters by a solution route," *Journal of the American Chemical Society*, Vol. 121, No. 22, 5191–5195, 1999.
115. Bley, R. A. and S. M. Kauzlarich, "A low-temperature solution phase route for the synthesis of silicon nanoclusters," *Journal of the American Chemical Society*, Vol. 118, No. 49, 12461–12462, 1996.
116. Mayeri, D., B. L. Phillips, M. P. Augustine, and S. M. Kauzlarich, "NMR study of the synthesis of alkyl-terminated silicon nanoparticles from the reaction of $SiCl_4$ with the Zintl salt, NaSi," *Chem. Mater.*, Vol. 13, No. 3, 765–770, 2001.

117. Prabakar, S., A. Shiohara, S. Hanada, K. Fujioka, K. Yamamoto, and R. D. Tilley, "Size controlled synthesis of germanium nanocrystals by hydride reducing agents and their biological applications," *Chem. Mater.*, Vol. 22, No. 2, 482–486, 2010.
118. Shiohara, A., S. Prabakar, A. Faramus, C. Y. Hsu, P. S. Lai, P. T. Northcote, and R. D. Tilley, "Sized controlled synthesis, purification, and cell studies with silicon quantum dots," *Nanoscale*, Vol. 3, No. 8, 3364-70, 2011.
119. Cheng, X., S. B. Lowe, S. Ciampi, A. Magenau, K. Gaus, P. J. Reece, and J. J. Gooding, "Versatile 'click chemistry' approach to functionalizing silicon quantum dots: applications toward fluorescent cellular imaging," *Langmuir*, Vol. 30, No. 18, 5209-16, 2014.
120. Cheng, X., E. Hinde, D. M. Owen, S. B. Lowe, P. J. Reece, K. Gaus, and J. J. Gooding, "Enhancing quantum dots for bioimaging using advanced surface chemistry and advanced optical microscopy: application to silicon quantum dots (SiQDs)," *Adv. Mater.*, Vol. 27, No. 40, 6144-50, 2015.
121. Dasog, M., Z. Yang, S. Regli, T. M. Atkins, A. Faramus, M. P. Singh, E. Muthuswamy, S. M. Kauzlarich, R. D. Tilley, and J. G. Veinot, "Chemical insight into the origin of red and blue photoluminescence arising from freestanding silicon nanocrystals," *ACS Nano*, Vol. 7, No. 3, 2676-85, 2013.
122. Li, Q., Y. He, J. Chang, L. Wang, H. Chen, Y. W. Tan, H. Wang, and Z. Shao, "Surface-modified silicon nanoparticles with ultrabright photoluminescence and single-exponential decay for nanoscale fluorescence lifetime imaging of temperature," *J. Am. Chem. Soc.*, Vol. 135, No. 40, 14924-7, 2013.
123. Wang, L., Q. Li, H.-Y. Wang, J.-C. Huang, R. Zhang, Q.-D. Chen, H.-L. Xu, W. Han, Z.-Z. Shao, and H.-B. Sun, "Ultrafast optical spectroscopy of surface-modified silicon quantum dots: unraveling the underlying mechanism of the ultrabright and color-tunable photoluminescence," *Light: Science & Applications*, Vol. 4, No. 1, e245, 2015.
124. Li, Q., T.-Y. Luo, M. Zhou, H. Abroshan, J. Huang, H. J. Kim, N. L. Rosi, Z. Shao, and R. Jin, "Silicon nanoparticles with surface nitrogen: 90% quantum yield with narrow luminescence bandwidth and the ligand structure based energy law," *ACS Nano*, Vol. 10, No. 9, 8385–8393, 2016.
125. Mangolini, L., E. Thimsen, and U. Kortshagen, "High-yield synthesis of luminescent silicon quantum dots in a continuous flow non-thermal plasma reactor," *Amorphous and Nanocrystalline Silicon Science and Technology-2005*, R. W. Collins, P. C. Taylor, M. Kondo, R. Carius, R. Biswas, Eds., Vol. 862, 307–312, 2005.
126. Mangolini, L. and U. Kortshagen, "Plasma-assisted synthesis of silicon nanocrystal inks," *Adv. Mater.*, Vol. 19, No. 18, 2513-+, 2007.
127. Zhou, S., Z. Ni, Y. Ding, M. Sugaya, X. Pi, and T. Nozaki, "Ligand-free, colloidal, and plasmonic silicon nanocrystals heavily doped with boron," *ACS Photonics*, Vol. 3, No. 3, 415–422, 2016.
128. Ni, Z., L. Ma, S. Du, Y. Xu, M. Yuan, H. Fang, Z. Wang, M. Xu, D. Li, J. Yang, W. Hu, X. Pi, and D. Yang, "Plasmonic silicon quantum dots enabled high-sensitivity ultrabroadband photodetection of graphene-based hybrid phototransistors," *ACS Nano*, Vol. 11, No. 10, 9854–9862, 2017.
129. Yu, T., F. Wang, Y. Xu, L. Ma, X. Pi, and D. Yang, "Graphene coupled with silicon quantum dots for high-performance bulk-silicon-based schottky-junction photodetectors," *Advanced Materials*, Vol. 28, No. 24, 4912–4919, 2016.
130. Rowe, D. J., J. S. Jeong, K. A. Mkhoyan, and U. R. Kortshagen, "Phosphorus-doped silicon nanocrystals exhibiting mid-infrared localized surface plasmon resonance," *Nano Letters*, Vol. 13, No. 3, 1317–1322, 2013.
131. Howes, P. D., R. Chandrawati, and M. M. Stevens, "Colloidal nanoparticles as advanced biological sensors," *Science*, Vol. 346, No. 6205, 2014.
132. Ma, Y., Y. Yamamoto, P. R. Nicovich, J. Goyette, J. Rossy, J. J. Gooding, and K. Gaus, "A FRET sensor enables quantitative measurements of membrane charges in live cells," *Nat. Biotech.*, Vol. 35, No. 4, 363–370, 2017.
133. Zhang, C. Y., H. C. Yeh, M. T. Kuroki, and T. H. Wang, "Single-quantum-dot-based DNA nanosensor," *Nat. Mater.*, Vol. 4, No. 11, 826-31, 2005.

134. Medintz, I. L., A. R. Clapp, H. Mattoussi, E. R. Goldman, B. Fisher, and J. M. Mauro, "Self-assembled nanoscale biosensors based on quantum dot FRET donors," *Nat. Mater.*, Vol. 2, No. 9, 630-8, 2003.
135. Ban, R., F. Zheng, and J. Zhang, "A highly sensitive fluorescence assay for 2,4,6-trinitrotoluene using amine-capped silicon quantum dots as a probe," *Analytical Methods*, Vol. 7, No. 5, 1732–1737, 2015.
136. Zhang, X., X. Chen, S. Kai, H.-Y. Wang, J. Yang, F.-G. Wu, and Z. Chen, "Highly sensitive and selective detection of dopamine using one-pot synthesized highly photoluminescent silicon nanoparticles," *Analytical Chemistry*, Vol. 87, No. 6, 3360–3365, 2015.
137. Zhang, J. and S.-H. Yu, "Highly photoluminescent silicon nanocrystals for rapid, label-free and recyclable detection of mercuric ions," *Nanoscale*, Vol. 6, No. 8, 4096–4101, 2014.
138. Cheng, X., B. F. P. McVey, A. B. Robinson, G. Longatte, P. B. O'Mara, V. T. G. Tan, P. Thordarson, R. D. Tilley, K. Gaus, and J. Justin Gooding, "Protease sensing using nontoxic silicon quantum dots," *BIOMEDO*, Vol. 22, No. 8, 2017.
139. Ruizendaal, L., S. P. Pujari, V. Gevaerts, J. M. Paulusse, and H. Zuilhof, "Biofunctional silicon nanoparticles by means of thiol-ene click chemistry," *Chem. Asian J.*, Vol. 6, No. 10, 2776-86, 2011.
140. Cheng, X., B. F. P. McVey, A. B. Robinson, L. Guillaume, P. B. O'Mara, V. T. G. Tan, T. Pall, R. D. Tilley, G. Katharina, and G. John Justin, "Protease sensing using nontoxic silicon quantum dots," 22(-)–22-7, 2017.
141. Medintz, I. L., H. T. Uyeda, E. R. Goldman, and H. Mattoussi, "Quantum dot bioconjugates for imaging, labelling and sensing," *Nat. Mater.*, Vol. 4, No. 6, 435–446, 2005.
142. Diaz, S. A., A. P. Malonoski, K. Susumu, R. V. Hofele, E. Oh, and I. L. Medintz, "Probing the kinetics of quantum dot-based proteolytic sensors," *Anal. Bioanal. Chem.*, Vol. 407, No. 24, 7307-18, 2015.
143. Ji, X., F. Peng, Y. Zhong, Y. Su, X. Jiang, C. Song, L. Yang, B. Chu, S.-T. Lee, and Y. He, "Highly fluorescent, photostable, and ultrasmall silicon drug nanocarriers for long-term tumor cell tracking and in-vivo cancer therapy," *Advanced Materials*, Vol. 27, No. 6, 1029–1034, 2015.
144. Song, C., Y. Zhong, X. Jiang, F. Peng, Y. Lu, X. Ji, Y. Su, and Y. He, "Peptide-conjugated fluorescent silicon nanoparticles enabling simultaneous tracking and specific destruction of cancer cells," *Anal. Chem.*, Vol. 87, No. 13, 6718-23, 2015.
145. Jiang, A., B. Song, X. Ji, F. Peng, H. Wang, Y. Su, and Y. He, "Doxorubicin-loaded silicon nanoparticles impregnated into red blood cells featuring bright fluorescence, strong photostability, and lengthened blood residency," *Nano Res.*, 2017.
146. Ji, X., F. Peng, Y. Zhong, Y. Su, X. Jiang, C. Song, L. Yang, B. Chu, S. T. Lee, and Y. He, "Highly fluorescent, photostable, and ultrasmall silicon drug nanocarriers for long-term tumor cell tracking and in-vivo cancer therapy," *Adv. Mater.*, Vol. 27, No. 6, 1029-34, 2015.
147. Hinde, E., K. Thammasiraphop, H. T. T. Duong, J. Yeow, B. Karagoz, C. Boyer, J. J. Gooding, and K. Gaus, "Pair correlation microscopy reveals the role of nanoparticle shape in intracellular transport and site of drug release," *Nature Nanotechnology*, Vol. 12, 81, 2016.