REVIEW

Contact Lens Sensors in Ocular Diagnostics

Nicholas M. Farandos, Ali K. Yetisen, Michael J. Monteiro, Christopher R. Lowe, and Seok Hyun Yun*

Contact lenses as a minimally invasive platform for diagnostics and drug delivery have emerged in recent years. Contact lens sensors have been developed for analyzing the glucose composition of tears as a surrogate for blood glucose monitoring and for the diagnosis of glaucoma by measuring intraocular pressure. However, the eye offers a wider diagnostic potential as a sensing site and therefore contact lens sensors have the potential to improve the diagnosis and treatment of many diseases and conditions. With advances in polymer synthesis, electronics and micro/nanofabrication, contact lens sensors can be produced to quantify the concentrations of many biomolecules in ocular fluids. Non- or minimally invasive contact lens sensors can be used directly in a clinical or point-of-care setting to monitor a disease state continuously. This article reviews the state-of-the-art in contact lens sensor fabrication, their detection, wireless powering, and readout mechanisms, and integration with mobile devices and smartphones. High-volume manufacturing considerations of contact lenses are also covered and a case study of an intraocular pressure contact lens sensor is provided as an example of a successful product. This Review further analyzes the contact lens market and the FDA regulatory requirements for commercialization of contact lens sensors.

1. Introduction

The number of contact lens users worldwide has been increasing and now stands at over 71 million,[1] half of which are in the US.[2] Over the last decade, there have been significant developments in the materials used in contact lenses, which has increased their appeal and accessibility to consumers.[2,3] Today, contact lenses are worn for vision correction, therapeutic and cosmetic purposes.[4] For the detection and treatment of many diseases, it is advantageous for a diagnostic device to monitor diagnostic parameters continuously.[5] For example, the potential to continuously monitor intraocular pressure for glaucoma using a non-invasive technique would have direct benefit at point-of-care (POC). However, many of the current POC diagnostic devices that rely on invasive methods to monitor a disease state have limitations.[6–8] This has resulted, as in the case of diabetes, in a 67% reduction in patient compliance in routinely monitoring capillary blood glucose concentrations.[9] Therefore, a practical POC device that can both continuously monitor the health of a patient in a minimally invasive way is of particular relevance.

Recent advances in electronics, microfabrication techniques, materials and sensing technologies have led to the development of contact lenses with diagnostic capabilities. Such "contact lens sensors" could meet stringent clinical requirements and result in improved patient prognosis. Research into these devices began over a decade ago, and prototype devices have been developed to detect and monitor diseases in real time. So far, these devices have mainly focused on monitoring glucose levels for diabetes management, and intraocular pressure (IOP) for glaucoma diagnosis, but their diagnostic potential is substantially greater. The prospect of a contact lens sensor that is powered externally and permits wireless readout using a auxiliary device has generated significant interest from Google, Novartis, and Microsoft (**Figure 1**).[10–12]

The purpose of this article is to i) overview ocular diagnosis, ii) provide a brief guide for contact lens design, iii) evaluate contact lenses as a platform technology, covering both technologies that monitor tear composition and intraocular pressure, iv) outline the major limitations of this technology, and v) highlight potential future directions. The scope of this Review encompasses clinically relevant biomarkers, sensing mechanisms, and the fabrication, powering, and readout of these sensors. We further evaluate their performance in ex vivo and in vivo clinical trials. The Review concludes with potential strategies to overcome limitations and highlights research gaps in the field. Contact lens-facilitated drug delivery applications are not covered by this Review.

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1.1. The Eye as a Sensing Site

1.1.1. Physical Characteristics of the Eye

Current diagnostic tests which utilize the physical characteristics of the eye typically analyze five principal parameters: i) The shape and color of the optic nerve (ophthalmoscopy), ii) the angle of the eye, where the iris meets the cornea (gonioscopy), iii) intraocular pressure (tonometry), iv) the thickness of the cornea (pachymetry), and v) the thickness of the retinal nerve fiber layer.[14] In addition to these parameters, a high blink-rate can be related to ocular dopamine activity, which may indicate the onset of healthy ageing to dementia.[15] Neurodegenerative disease may also be diagnosed by monitoring the eye's muscle activities, such as in rapid eye movement sleep behavior disorder.[16] To date, only IOP has been used as a diagnostic parameter by a contact lens sensor, which has been achieved indirectly by measuring the curvature (radius) of the cornea.[17] The IOP of healthy human eyes lies between 10–21 mmHg[18,19] ($\mu = 16 \pm 2.5$);[20] in disease, this range can be 5–40 mmHg.[21] At present, IOP is the primary indicator in the diagnosis and management of glaucoma.[21,22] Current clinical methods to measure the IOP of the eye involve numbing the eye and applying pressure using a device or a short puff of warm air.[23] This method is limited since it is a single, one-off measurement and is therefore unlikely to establish peak IOP, which is required for an effective treatment. This was verified by a 24 h continuous IOP monitoring study of glaucoma patients, which showed that the peak IOP was on average 4.9 mmHg higher than that established by a single measurement ($p < 0.0001$), and in 13.8% of patients in the study was 12 mmHg higher than the single measurement value. This information led to a change in the clinical management of 79% of participants, and 56.5% of the total patients in the study were subsequently offered trabeculectomy.[24] Consequently, continuous monitoring of IOP using contact lens sensors is an attractive alternative to current clinical methods.

1.1.2. Tear Fluid Composition

The tear film comprises three layers and serves to clean and lubricate the eye (basal tears) (**Table 1**). Tear fluid production can also occur as a result of laughing or crying (psychic tears) or through eye irritation (reflex tears). The blood and tear fluid is separated by the "blood-tear barrier," which consists of the acinar and ductal cells within the lacrimal gland, the conjunctival epithelium, and the corneal epithelium. This barrier creates a compositional difference between tear fluid and blood.[25] However, there is a close relationship for certain metabolites between both fluids due to plasma leakage, where the components in the blood supply to the brain pass through the blood-tear barrier into the tear fluid.[26–28] **Table 2** shows the difference in composition of the major metabolites between tear fluid and blood. Therefore, where a correlation between the two fluids exists, it is possible to use tear fluid as a proxy for the analysis of blood composition.[29] For example, by using the relationship between blood and tear glucose, Na^+, K^+, and Cl^- ion concentrations, contact lens sensors can act as continuous and minimally invasive diagnostic devices.[28,30]

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The protein composition of human tear fluid has been used to diagnose a variety of diseases and conditions (**Table 3**).[52,53] The major proteins in tears include enzymes (lysozyme), neuropeptides, antibodies and lactoferrin. Based on electrophoretic techniques, high-performance liquid chromatography (HPLC) and mass spectrometry,[54] 97 unique proteins have been

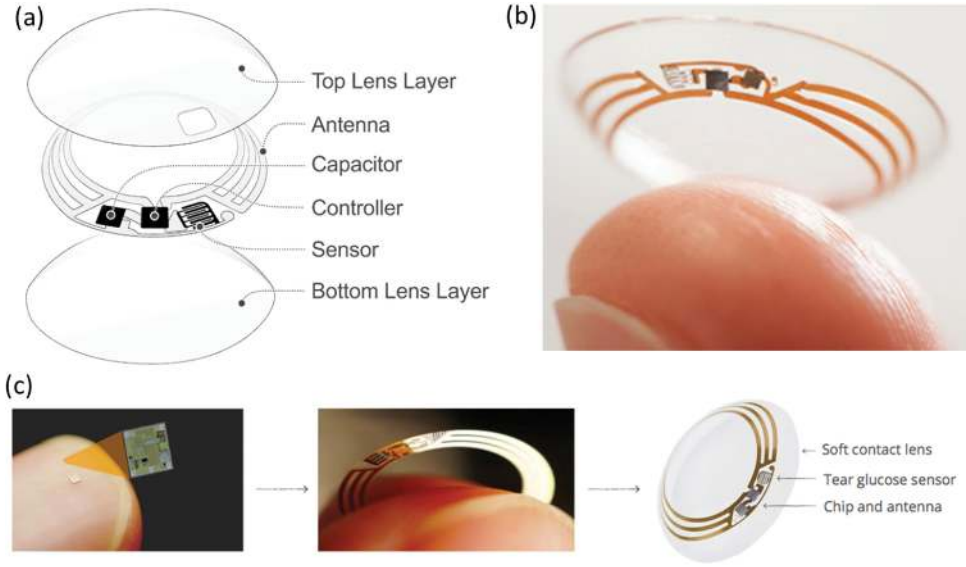

Figure 1. The contact lens sensor under co-development by Google and Novartis. It measures glucose concentration in tears using a miniaturized electrochemical sensor embedded into a hydrogel matrix. a) A schematic of the contact lens sensor, showing the electrical circuitry of the sensing system. b) The contact lens sensor prototype. c) The wireless chip, which is mounted, with the sensor, onto an electronic ring, and then embedded into the contact lens. Reproduced with permission.[13] Copyright 2014, Google X.

identified in tear fluid.[55] However, the number of proteins reported varies significantly with sampling method, ranging from 54 to 1543.[31]

Although the presence of antibodies in tear fluid can indicate particular disease conditions, quantitative measurements can provide information about the severity of the disease.[80] Unlike many of the other analytes in tears, antibodies are directly secreted into the tears as opposed to leaching in from blood vessels through the blood-tear barrier, and so they are potentially more reliable biomarkers.[54] In addition to basal tears, other human lachrymal liquids include reflex tears and psychic tears. Reflex tears, secreted through transient receptor channels in the ophthalmic nerve, serve as a defense mechanism to remove irritants that may have contacted the eye. This type of tear is secreted when i) the eye is irritated by foreign stimuli such as particles or vapor, ii) is exposed to an intense light source, iii) hot or peppery stimuli contacts the tongue, and iv) the person vomited, coughed, or yawned.[81] Psychic effects such as pleasure, anger, or physical pain can also induce lacrimation. In comparison to basal or reflex tears, psychic tears contain higher concentrations of hormones such as prolactin, adrenocorticotropic hormone, and leucine encephalin.[82] Hence, the contact lens sensors should not cause irritation, which can induce the secretion of reflex and psychic tears. Hence, tear fluid composition is subject to change based on the method of sample collection. Erroneous sample collection methods have caused significant disagreement in the literature. For example, the concentration range of glucose in tear fluid has been reported as 0.1–3.6 mM,[51] 0.6 mM,[49] 128–166 μM,[83] and recently 0.013 mM.[34] The glucose concentration of tear fluid samples collected through filter paper can be higher than capillary since filter paper mechanically stimulates the corneal and conjunctival epithelium.[84,85] The tear fluid should be collected with minimum tear stimulation and eye irritation. In addition, erroneous concentration ranges might also be a result of sampling methods that required high sample volume. Hence, caution should be taken in the consideration of concentration ranges, which might be affected by the sampling method.

Table 1. The composition and function of the tear film.

Tear layer	Composition and source	Function	Thickness [μm]	Ref.
Outer lipid	Both a polar and non-polar lipid layer with intercalated proteins. It is secreted by the meibomian glands.	It maintains tear film stability and also prevents the aqueous layer from evaporating. It may provide a barrier against pathogens.	≈0.1	[28,31,32]
Aqueous	Many electrolytes, proteins and small molecule metabolites and is secreted by the main and accessory lacrimal glands.	Protects the eye from infection via specific and non-specific defense mechanisms.	≈270	[31,33]
Inner mucin	>20 glycoproteins containing O-linked carbohydrates and a protein core. Its major source is goblet cells in the conjunctiva, but corneal conjunctival epithelial cells and lacrimal glands may also be contributors	The hydrophillic interfacial layer between the ocular surface and the aqueous layer	≈30	[31]

Table 2. Principal analytes in the basal tear film with their typical concentrations, and their corresponding concentrations in blood.

Analyte	Tear fluid concentration [mM]	Blood concentration [mM]	Diagnostic application	Ref.
Glucose	0.013–0.051	3.3–6.5	Diabetes management	[34]
Lactate	2.0–5.0	0.36–0.75	Ischemia, sepsis, liver disease and cancer	[35]
Na^+	120–165	130–145	Hyper/hyponatremia	[36]
K^+	20–42	3.5–5.0	Hyper/hypokalemia and an indicator of ocular disease	[26]
Ca^{2+}	0.4–1.1	2.0–2.6	Hyper/hypocalcemia	[37]
Mg^{2+}	0.5–0.9	0.7–1.1	Hyper/hypomagnesemia	[38]
Cl^-	118–135	95–125	Hyper/hypochloremia	[39]
HCO_3^-	20–26	24–30	Respiratory quotient indicator	[40]
Urea	3.0–6.0	3.3–6.5	Renal function	[41]
Pyruvate	0.05–0.35	0.1–0.2	Genetic disorders of mitochondrial energy metabolism	[42]
Ascorbate	0.22–1.31	0.04–0.06	Diabetes	[43,44]
Total Protein	≈7 g/L	≈70 g/L	Dry eye conditions, ocular insult and inflammation	[45,46]
Dopamine	0.37	475×10^{-9}	Glaucoma	[47,48]

Analyte concentrations are taken from the literature.[49–51] The diagnostic applications were taken from the sources indicated in the "Ref." column.

1.1.3. Physiological Issues

The relationship between the concentration of several analytes in tear fluid and blood has been difficult to establish. Complications include flow-dependent concentration effects and low sample volumes, which in conjunction with low concentrations (μM) requires high sensitivity for analysis.[86] Wearing contact lenses has also been shown to alter the protein profile of tear fluid in a complex, but consistent way.[87,88] In addition, the contact lens solution used for sterilization may modify the

Table 3. Diseases and conditions that can be diagnosed by analyzing the composition of tear fluid.

Disease/Condition	Changes in Composition	Ref.
Meibimian gland disease	Proteomic analysis	[56]
AIDS	IgA	[57]
Ocular chlamydia trachomatis	IgA, antichlamydial IgG	[33,58]
Autoimmune thyroid eye disease	Proteomic analysis	[59]
Pterygium	α-defensins, and S100 A8 and A9	[60]
Keratoconus	Total protein, lactoferrin, and secretory IgA	[61]
Ocular rosacea	Matrix metalloproteinase-8 (MMP-8), oligosaccharides	[62,63]
Blepharitis	Proteomic and lipidomic analysis, serum albumin precursor, α-1 antitrypsin, lacritin precursor, lysozyme, Ig-κ chain VIII, prolactin inducible protein (PIP/GCDFP-15), cystatin-SA III, pyruvate kinase, and phosphoethanolamine & sphingomyelin for chronic blepharitis	[63,64]
Allergic conjunctivitis	Total protein, serum albumin precursor, Ig gamma-2, leukocyte elastase inhibitor, sPLA2-IIa	[65]
Diabetes	Proteomic analysis (glycosylation of the tear proteins, e.g., albumin), glucose	[66,67]
Dry eye (xerophthalmia)	Proteomic analysis, peptide/protein markers (proline-rich protein 4, Mammaglobin B, lipophilin A, calgranulin S100A8)	[68]
Sjörgen's syndrome	Proteomic analysis, lysozyme, epidermal growth factor, AQP5, IL-1α and β, IL-6, IL-8, TGF- β1, IL-1Ra and TNF-α, RNA transcripts for MUC5AC, protein levels of MUC5AC, MUC5AC-positive conjunctival cells, glycosylation of mucins, (GalNAc transferase, GalNAc-T2 and -T6 isoenzymes), O-glycan residues, MMP-9	[45,69–71]
Glaucoma	Autoantibody reactivities (HSP10, HSP27, MBP and Protein S100)	[72]
Conjunctivochalasis	S100 family (A8, A9, A4), guanosine triphosphate-binding protein 2, L-lactate dehydrogenase A-like 6B, fatty acid-binding protein, keratin type I cytoskeletal 10, glutathione S-transferase P, peroxiredoxin-1, peroxiredoxin-5, and cullin-4B+ glyceraldehyde 3-phosphate dehydrogenase, Pro-MMP-9	[73,74]
Diabetic retinopathy	Proteomic analysis	[66,67,75]
Lacrimal gland dysfunctions	Proteomic and lipidomic analysis	[76]
Cancer	Lacryglobin, actin, albumin	[63,77,78]
Herpes Simplex Virus	HSV-specific sIgA, HSV-specific IgG antibodies	[33,79]

tear protein composition during wear. A further complication in establishing the relationship between analytes in tear fluid for healthy and unhealthy people is that the protein composition of tear fluid may be influenced by certain medications the patient may be taking. For example, a patient taking glaucoma medication can have an altered protein concentration profile in tear fluid.[89] Despite the complications between the concentration of analytes in blood and tear fluid, the analysis of tear fluid has been used successfully in the diagnosis and monitoring of a variety of diseases.

1.2. Origins of Contact Lenses

Leonardo da Vinci is generally credited with the concept of the contact lens (circa 1508), in which he envisioned a clear shell that could be filled with water and placed against the eye for vision correction. The first practical devices (in 1887) were made from blown glass, and subsequently poly(methyl methacrylate) (PMMA) in the 1940s that fitted onto the eye, covering the sclera. Further developments led to the cornea-covering contact lenses, which is the form that we use today.[90–92] Although PMMA increased comfort, it behaves as a glass at room temperature due to its high glass transition temperature, and consequently it suffers from low oxygen permeability. These advances permitted consumers to wear the contact lenses for more than a few hours at a time without causing corneal misting or halation due to the increased pressure imposed on the eye by the lens. However, these "rigid" materials had limitations due to their bio-incompatibility: They were i) restrictive for an equivalent water content to the human eye, ii) non-biologically inert causing eye irritation, and iii) impermeable to oxygen required by the corneal epithelial cells, resulting in discomfort. However, modern "soft" contact lenses have overcome these limitations. Poly(2-hydroxyethyl methacrylate) (pHEMA) was the first such material, which led to the introduction of soft contact lenses to consumer products in the 1970s when it gained FDA approval.[4,93,94] Since then, soft materials for contact lenses have been developed to increase their permeability to oxygen and improve their hydration properties. In 1999, silicon-based materials such as polydimethylsiloxane (PDMS) were copolymerized with other monomers to produce contact lenses with high oxygen permeability and user comfort. However, the silicon content rendered the material hydrophobic, which required rewetting agents to reduce hydrophobicity. In addition to soft lenses, there are also hybrid rigid/soft contact lenses, which are composed of a small rigid lens mounted onto a soft lens that provides enhanced optical and comfort properties, particularly for visual defects such as irregular astigmatism.[94,95] Soft materials have formed the foundation into which sensing technologies have been ultimately integrated to form contact lens sensors.

2. Contact Lenses as a Platform Technology

Contact lenses as minimally invasive sensors have been used to measure IOP and concentrations of glucose, lactate, and K^+ ions in tear fluid. This Section provides an overview of the contact lens sensor materials, fabrication methodologies, sensing mechanisms, powering techniques, and readout. Contact lens sensors should have a number of desirable features. First, they should not cause eye irritation, which alters the concentrations of metabolites and proteins within the tear fluid. For example, higher concentrations of glucose are detected when the test causes ocular irritation compared with minimally invasive sampling methods.[28] This is due to the release of glucose from damaged cells and subsequent diffusion into the tear fluid.[96] The alteration effect is opposite with urea, a result consistent with leakage of urea through the damaged epithelial barrier into the tissues surrounding the anterior chamber fluid.[84] The concentration of lactate and pyruvate does not change upon irritation, suggesting that the epithelium is not a barrier for lactate or pyruvate. Although contact lenses currently on the market cause little or no irritation after an adjustment period, irritation is a potential consequence to avoid when integrating sensors into contact lenses.[97] Secondly, contact lens sensors should provide a stable, fully reversible, sensitive, and specific response to the target analyte. The sensitivity of the contact lens sensors may deteriorate over time if a sensing mechanism exhibits hysteresis due to the degradation of sensor materials. This can occur through denaturation of the protein sensing units, photobleaching of organic dyes in fluorescence sensing, or leaching of the materials of the sensor into the tear fluid.[96] A common technique employed to avoid leaching is to encapsulate the sensing material before embedding it into the polymer matrix. However, this technique introduces additional variables such as a reduction in the rate of diffusion.[98] Thirdly, the time between detection and readout should be rapid. For example, this is particularly important for glucose concentration to ensure the user receives prompt warning of the onset of hypo/hyperglycemia. The delay between changes in glucose concentrations in blood and the corresponding change in tear fluid is ≈5 min;[99] however, delay times between detection and readout of up to 20 min have been recorded, indicating that minimizing readout time is critical.[100] Response time is mainly determined by the sensing materials and the surrounding polymer matrix. To minimize response time, a calibration curve can be obtained for each patient to compensate partially for the delay in the case of glucose measurements.[101] The key to a rapid (and reversible) response lies in the optimization of the binding kinetics under normal physiological conditions in the eye to allow rapid complexation and release of analyte/product molecules. The sensing materials should release the analyte/product molecules after binding to avoid any delays resulting from a local concentration imbalance on the accuracy of the readout.[96] Additional factors such as the shelf life, temperature stability, and sterility of the sensing material should be balanced with the aforementioned requirements to achieve a reliable and clinically useful readout for a contact lens sensor.

2.1. Contact Lens Design

2.1.1. Physical Parameters

In designing the dimensions and functionality of a contact lens, four physical parameters need to be considered. The base curve

radius (BCR), ranging from 8–10 mm, is selected to provide a comfortable fit between the cornea and the contact lens to facilitate tear exchange and allow sufficient oxygen transfer between the corneal epithelial cells and the surrounding air.[21,102] The BCR is equal to the curvature of the back surface of the contact lens, usually equivalent to the curvature of the anterior surface of the cornea.[103] The contact lens diameter, selected to provide a fit onto the eye, can be determined from an eye examination, typically carried out by an optician. The center thickness (CT), ≈0.1 mm, together with the oxygen permeability of the contact lens material, determines oxygen flux to the cornea.[103] CT is the thickness between the inner and outer surfaces through the central axis of the contact lens. Another parameter, the optical power (OP) of the contact lens is paramount in vision correction and is the refractive power of the lens, measured in diopters. OP is optimized by testing visual acuity (the clarity or sharpness of vision).[104] Hence, all physical parameters should be established prior to selecting the substrate and fabricating the lens.

2.1.2. Materials

The mechanical, chemical, and optical properties of the substrate should combine certain characteristics in order to construct contact lenses. Stresses in the lens imposed by repeated use and eye movement can cause irreversible deformation and fracture, which reduces optical performance and comfort. Mechanical properties that should therefore be optimized are tensile strength, elongation-at-break and Young's modulus.[105] Water content, free-to-bound water ratio and biological inertness are also important chemical properties that determine in-eye performance.[106,107] The important optical properties of the substrate are transparency and a sufficiently high refractive index. There are also production constraints on the materials involving cost, compatibility with manufacturing processes, and availability.[108]

Rigid and soft contact lenses are the two major lens types. Rigid lenses are more durable and resistant to deposit buildup than soft lenses. They tend to have a lower cost over their lifetime than soft contact lenses, and they typically provide clearer vision. Additionally, more time is needed for the user to become accustomed to wearing rigid lenses than soft lenses; the adjustment periods are of the order weeks for rigid lenses compared to days for soft lenses.[109] While PMMA-based rigid contact lenses are presently obsolete, soft lens materials include hydrogels such as pHEMA, polyvinyl alcohols (PVA), polyacrylamides (pAAm), 2,3-dihydroxypropyl methacrylate, polydimethylsiloxane (PDMS), poly(ethylene terephthalate) (PET), silicones and mixtures of these materials.[108] Hence, a wide array of materials can be combined with the physical parameters to construct a contact lens into which a sensor can be incorporated.

2.2. Intraocular Pressure Contact Lens Sensors

An elevated intraocular pressure (IOP) is the main indication of glaucoma. A change in the forces that act upon a contact lens can be correlated with IOP, and hence contact lens sensors have utility in this application. Five different forces act on a corneal contact lens: Atmospheric pressure (P_0) acts over its free surface, the hydrostatic pressure of the post-lens tear film (P_1) acts over the surface of the lens in contact with the cornea, the force of gravity (p) (lens weight), the force exerted on the contact lens by the eyelid during a blink (F_{lid}), and the surface tension of the prelens tear film ($F\sigma$). The effect of these forces determines the position of the contact lens on the cornea (**Figure 2**). There are four categories of contact lens sensors that monitor IOP: i) Capacitative sensors, ii) piezo-resistive sensors, iii) strain gauge sensors, and iv) microinductor sensors.

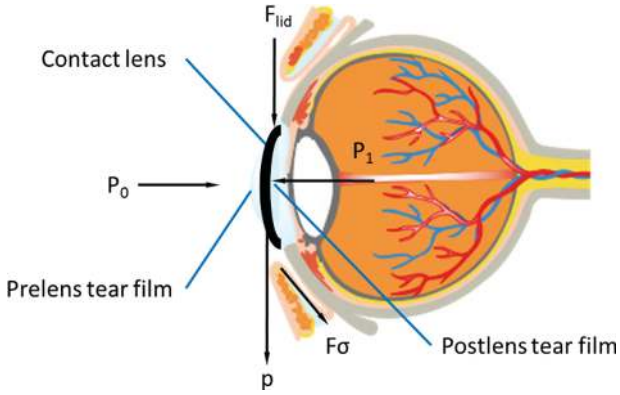

Figure 2. The five major forces acting on a contact lens: P_0, atmospheric pressure; P_1, hydrostatic pressure of the postlens tear film; p, lens weight; F_{lid} the lid force during blinking; and $F\sigma$, the surface tension of the prelens tear film.

2.2.1. Capacitative Sensors

Capacitative contact lens sensors have been developed to measure IOP from the curvature of the cornea, and are suited to low-force applications. These sensors consist of two independent layers: an outer reference layer and an inner sensing layer. The sensing layer can detect changes in the curvature of the lens (relative to the reference layer), which is related to changes in IOP. As the curvature of the lens changes, the resonance frequency of the inductor-capacitor circuit also changes, which can be measured and correlated to IOP.

A capacitative contact lens sensor was fabricated from medical-grade silicone using transfer molding, and the electrodes and inductive coil were etched from copper foil (**Figure 3**).[21] To ensure biocompatibility, the electrodes of the capacitator and inductive coil were coated with parylene-C. The circuitry was separately cast inside the silicone layer and subsequently cured. The sensing and reference layers were wire-bonded together using a silicone adhesive, forming the sensing material. This sensor was tested on silicone eyes and enucleated porcine eyes. With the silicone eyes the sensor had a linearity of $R^2 = 0.99$, and with enucleated porcine eyes the sensor was able to quantify IOP with a sensitivity of 240 ppm/mmHg between 7.8 and 42.8 mmHg at constant pressure. Upon cycling IOP in the enucleated porcine eyes from 6 to 28 mmHg in <100 s,

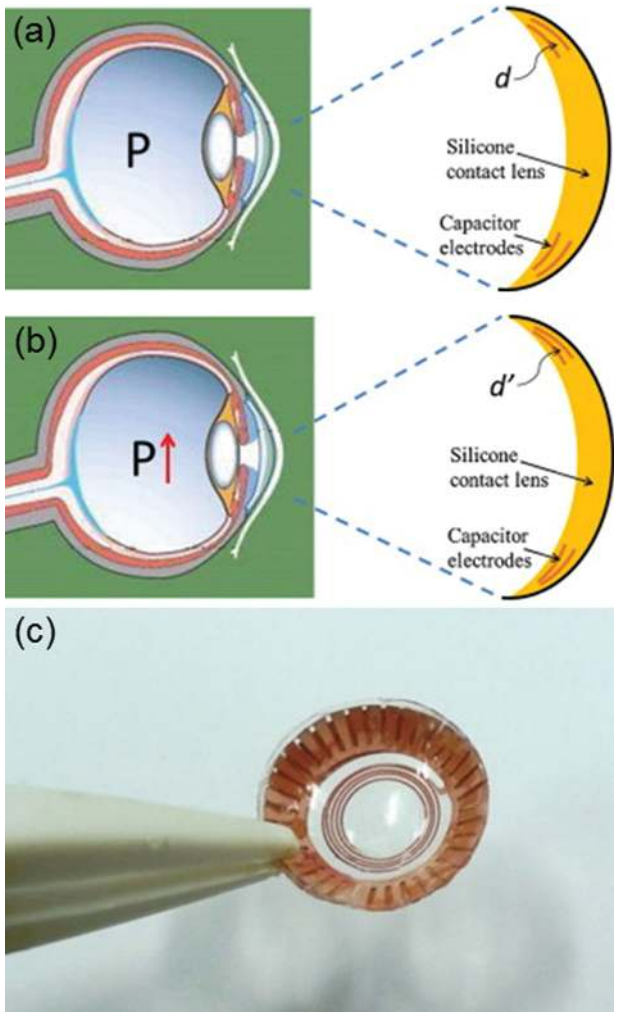

Figure 3. The sensing mechanism of the capacitative contact lens sensor. a) Contact lens sensor configuration on eye with normal IOP, and b) the contact lens sensor configuration with elevated IOP, which reduces the distance between the capacitative electrodes. c) The capacitative contact lens sensor that tracked IOP, made from medical grade silicon. Reproduced with permission.[21] Copyright 2013, Elsevier.

there was negligible delay between change in pressure and detection. This was due to the time dependent shape response of the viscoelastic cornea to IOP, and subsequent transmission of this force, via surface tension, to the contact lens sensor. Readout was achieved wirelessly up to 2.5 cm distant using a conventional telemetry system. The receiver was taped to the eye socket, which was impractical for real-life use. However, if the readout circuit can be incorporated into more user-friendly devices such as glass frames or facial patches, this contact lens sensor may become a practical diagnostic device.[21] The efficacy of the lenses in vivo should also be tested, as living systems have more complex parameters that must be accounted for such as eye blinking.[110] Overall, capacitative sensors are a simple platform to measure IOP profiles for the diagnosis of glaucoma as well as offering a long sensor lifetime.

2.2.2. Piezo-Resistive Sensors

The discovery of flexible, conducting, all-organic bilayer films with piezo-resistive properties allows IOP to be measured using a contact lens sensor.[111,112] The advantages of this technology include transparent sensing materials, and a highly deformation-sensitive bilayer. The bilayers consist of a polycarbonate film covered on one side by a layer of nanostructure crystals of an organic molecular conductor. The resulting piezo resistance is caused by the softness of the nanocrystals of the conducting salt that are deformed under a small strain, which alters their conducting properties.[112,113]

A device that uses this sensing technique has been tested with enucleated pig eyes.[114] The device employed a polycarbonate layer of poly(bisphenol A carbonate) and a conducting layer of bis(ethylenedithio)tetrathiafulvalene (BEDT-TTF) (**Figure 4**). The sensing materials were incorporated into a hard contact lens that was gas permeable. Biocompatibility of the contact lens sensor was confirmed by in vivo guinea pig eyes. Its physiological stability was demonstrated using artificial tear fluid, which had no influence on readout. The device was subsequently tested ex vivo on 15 porcine eyes over a pressure range of 20–50 mmHg, and the results showed linearity, reproducibility with cyclic pressure loading, and a sensitivity of 1.5 Ω/mmHg. The device was powered through cable connection and readout was achieved using a laptop with Bluetooth connectivity. Although the sensor tracked IOP over time, at 10 mmHg the sensor had a slight resistance drift, which became apparent after 5 min, of ≈2 Ω after 20 min. The resistance drift is independent of absolute IOP, but is dependent on the duration of the measurement at constant IOP. This was, however, smaller than changes in resistance experienced by the conducing layer when there was a real change in IOP (>2 mmHg). Piezo-resistive sensors offer a long sensor lifetime and a high sensitivity. However, the sensing material can suffer from temperature degradation and for practicality a wireless powering system is desirable.

2.2.3. Mechanical Strain Gauge Sensors

These sensors use strain gauges to convert changes in the curvature of the cornea into an electrical signal to measure IOP. The advantages of mechanical strain gauge sensors are that a wide range of materials is available for construction, and temperature variability can be mitigated, unlike piezo-resistive sensors. A contact lens sensor that incorporates mechanical strain gauges has been developed and tested on enucleated porcine

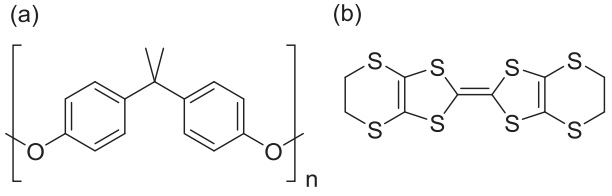

Figure 4. a) Poly (bisphenol A carbonate), and b) bis(ethylenedithio)-tetrathiafulvalene (BEDT-TTF), that form the organic bilayer of the piezo-resistive sensor.

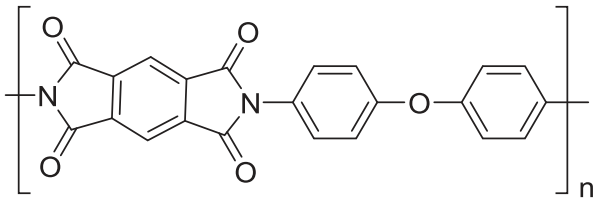

Figure 5. The polyimide PI2611 (DuPont) sandwiching the strain gauge sensors, forming an insulating and biocompatible layer.

eyes.[115] The sensor has four strain gauges embedded into a silicone-based contact lens: two sensing resistive strain gauges to measure the curvature of the cornea, and two compensation-resistive strain gauges for thermal compensation. Two compensation-resistive strain gauges doubles the sensitivity to corneal curvature. A silicon-based material was selected due to its insensitivity to hydration, therefore all changes in shape result from changes in IOP. The strain gauges were fabricated from Pt–Ti, sandwiched between two layers of polyimide (PI2611, DuPont), which was electrically insulating and biocompatible (**Figure 5**). Readout was achieved by embedding a telemetry microprocessor and a gold antenna into the contact lens. The microprocessor was attached to the contact lens using a flip-chip technique, and interconnected with the sensing strain gauges by conducting epoxy glue. The gold antenna was incorporated into the contact lens using electro-deposition. The telemetry microprocessor permits both wireless powering from a frame worn in close proximity to the eye, and data transfer to a portable readout device.[115] Ex vivo trials were carried out using enucleated porcine eyes. The first trial simulated ocular pulsation by dynamically changing IOP between 11 and 14 mmHg, demonstrating that the sensor can track changes in IOP. The second trial varied IOP of an enucleated pig eye from 20–30 mmHg in static steps of 1 mmHg, and measured a reproducibility, linearity and sensitivity of ±0.2 mmHg ($p < 0.05$), $R^2 = 0.99$, and 113 ± 19 μV/mmHg respectively.[115] Mechanical strain gauge sensors offer the capability to measure IOP with thermal compensation.

2.2.4. Microinductor Sensors

These sensors measure IOP by correlating changes in the inductance value of the sensing material to changes of corneal curvature resulting from IOP variations. The inductance value is quantified by an inductor-capacitor (LC) circuit. A device incorporating a microinductor sensor was fabricated by spin coating a silicon substrate with a photoresistor (AZ4621) or evaporated film aluminum to form a sacrificial layer.[116] Parylene-C was subsequently spin-coated and used as the substructure for the sensing material. After photolithography, a Ti and Au biocompatible composite material was deposited, and the remaining photoresist lifted off to define the metal alignment. Anisotropic dry etching of the Parylene-C was performed to define the film shape and open a contact window of the pad to provide a connection with the integrated electrical circuit. The sensing material was subsequently integrated onto a pHEMA-based contact lens. The resultant swelling due to hydration of pHEMA matrix makes it difficult to embed the sensor into the contact lens, but provides a greater user comfort. Preliminary trials showed that the microinductor radius variation and oscillation frequency were linearly related, with a sensitivity of 167 ppm/mmHg. For practicality, it is desirable to incorporate a wireless readout/powering system. The outlined fabrication technique allows the incorporation of microinductor sensors into soft contact lens materials to achieve user-comfort.

2.3. Sensing Analytes in Tear Fluid

Contact lens sensors that monitor analytes in tear fluid convert compositional information into signals that can be read optically by an observer or by an instrument. They may be classified according to their sensing principle: i) fluorescence, ii) holographic, iii) colloidal crystal array, and iv) electrochemical sensing. These sensors have been used to detect concentrations of glucose and lactate in tear fluid. This Section provides an overview of devices that employ these sensing principles.

2.3.1. Fluorescence Sensing

An excitable synthetic molecule (a fluorophore) absorbs irradiant radiation of a specific wavelength and subsequently emits a photon of lower energy (longer wavelength). The emitted and background radiation can be distinguished using filtering techniques, rendering this measurement highly sensitive for single molecule detection.[117] Fluorescence occurs within microseconds and accompanies transitions from singlet excited states to ground states. Excitation and emission photon wavelengths are highly dependent on the chemical composition and attributes of the target fluorophore, and therefore fluorescence sensing is highly molecule-specific. Once a fluorophore is excited, intermolecular interactions can occur with the surrounding chemical environment. Therefore, the surrounding environment has an influence on the properties of the emitted photon.[96,118,119] Förster resonance energy transfer (FRET) is the distance-dependent transfer of energy from a donor fluorophore to an acceptor fluorophore.[120] In FRET, a donor fluorophore is excited by the incident light, and if an acceptor is in close proximity, the excited state energy from the donor is transferred. As a result, the fluorescence intensity and excited state lifetime of the donor decreases while the emission intensity of the acceptor increases. The rate of this energy transfer can be related to the nanometer scale distances and changes in distances, both in vitro and in vivo between donor and acceptor molecules. This provides versatile measurements since the fluorescence decay times can be measured in addition to the intensity. FRET also allows in vivo measurements without light scattering from tissue. FRET is independent of the fluorophore concentration, which may produce photobleaching through diffusion or degradation. FRET does not cause any damage to the biological host system due to its non-invasive nature: Exciting photons typically occur in the low-energy near-infrared spectrum. Therefore, the molecules can be excited and the corresponding emitted photons analyzed optically.[121]

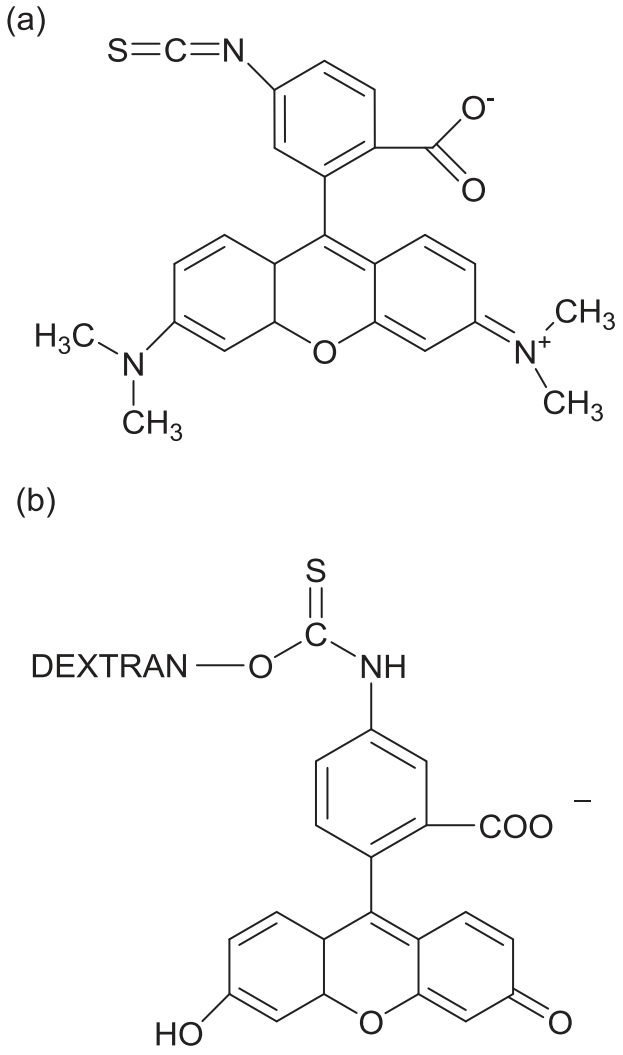

Figure 6. The sensing materials: a) TRITC and b) FITC-dextran. The site of attachment of FITC to the dextran is assumed to be randomly associated with any free hydroxyl group.

Fluorescence sensing has received attention in contact lens sensor design due to its versatility, sensitivity, and specificity. One contact lens sensor using this principle that detects glucose in tear fluid employed hydrogel-encapsulated nanospheres containing tetramethylrhodamine isothiocyanate concanavalin A (TRITC-Con A) and fluorescein isothiocyanate dextran (FITC-dextran).[122] **Figure 6** shows the structures of tetramethylrhodamine isothiocyanate and fluorescein isothiocyanate dextran (FITC-dextran).

The nanospheres were embedded within a PVA-based (Nelfilcon A) matrix by first adding the nanospheres (≈1000) to the monomer solution and then inducing polymerization, within a contact lens-shaped mold, using the UV initiator 2-hydroxy-2-methylpropiophenone over 6 h. Polymerization immobilized the nanospheres within the polymer matrix. As the concentration of glucose increases, FITC-dextran molecules are displaced from their binding positions to TRITC-Con A molecules, by glucose molecules that diffuse into the nanospheres, which decreases FRET, resulting in an increase in the intensity of fluorescence; and vice versa. This contact lens sensor tracked the concentration of blood glucose of five diabetic and one non-diabetic human patients over 3 h. However, there was a delay between the actual tear glucose concentration and that measured by the contact lens sensor.[122] This was due to both a delay in glucose entering the tear film through the blood-tear barrier and also its diffusion to the sensing material through the polymer matrix ($t \approx 500$ μm). This delay can be mitigated by deriving a polynomial (so called "delay algorithm") for each individual patient by establishing the quantitative relationship between the concentrations of blood and tear glucose.[101] In vitro experiments have demonstrated that the delay detection and readout is under 1.5 min,[122] and is comparable to the delay encountered in forearm glucose sampling.[101] Readout was achieved using a hand-held photofluorometer, which rested on the orbital rim of the eye for ≈1 s in order to block out ambient light. Blue light set to 2.6 MHz and signal locked from a circular array of light emitting diodes (LEDs) in the photofluorometer was focused onto the eye. The resultant green fluorescence then passed through a collator lens and barrier filter, which excluded the blue light, to a diode detector. The signal was averaged and passed through integrated circuit amplifiers to give a digital readout on the photofluorometer. Additionally, a telemetry transmitter was attached to the photofluorometer that could transmit the readout to an insulin pump fitted with a telemetry receiver.[122,123]

Other FRET-based contact lens sensors have been proposed, where the main difference is the method employed to immobilize the glucose-sensing unit to prevent leaching. One such device employs silica nanoparticles, which encapsulates an organic dye into the contact lens material which can be pHEMA or PDMS.[124] These sensing materials have been embedded, forming mono or multilayers, into the polymer matrix using photopolymerization and have detected glucose at a sensitivity of 0.5–5.0 mM with a 90% lag time from changes in the concentration of blood glucose of 5 min. Encapsulating the organic dye in silica-based nanoparticles has advantages over encapsulating within a hydrogel polymer. These include polymer materials that are prone to structural changes with variation in temperature and pH, which may cause leakage and erroneous results, whereas silica nanoparticles maintain structural integrity. However, there are some structurally rigid polymer-based nanoparticles which may be used for encapsulation.[125] In addition, unlike hydrogels, many different organic dyes can be encapsulated within a single silica nanoparticle so a single contact lens can potentially be multiplexed to detect various metabolites and metal ions. Hence, fluorescence sensing offers the potential to produce sensitive glucose-responsive contact lenses, which can be read by handheld readers.

2.3.2. Holographic Sensors

Photonic structures consisting of holographic gratings have been developed to quantify the concentration of glucose in tear fluid at physiological conditions (pH and ionic strength).[126,127] This method allows the formation of a multilayered periodic structure within a polymer matrix, which can be functionalized

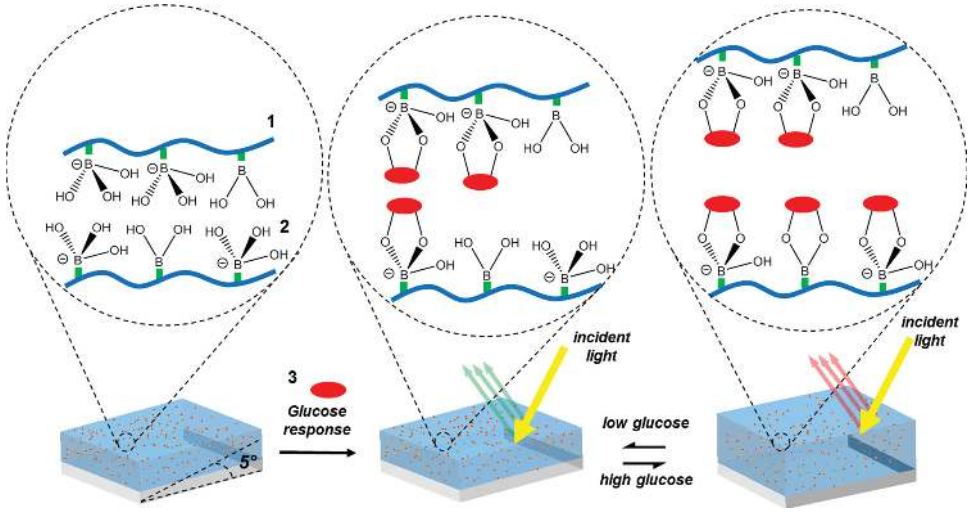

Figure 7. Optical characterization and principle of operation of the holographic sensor. The sensor composed of pAAm-based hydrogel (1) functionalized with 3-(acrylamido)phenylboronic acid (3-APB) (2) for sensing the target glucose (3). Reversible swelling of the photonic nanosensor by glucose modulates both the nanoparticle distribution spacing and the refractive-index contrast, and systematically shifts the diffracted light from shorter to longer wavelengths as the hydrogel expands in the direction normal to the underlying substrate. Reproduced with permission.[131] Copyright 2014, American Chemical Society.

with boronic acid derivatives that are able to bind to carbohydrates covalently and reversibly (**Figure 7**).[30,128] Unlike fluorescence-based sensors, holographic sensors do not require dyes. Hence, they are not subject to photobleaching, which allows them to be used within contact lens sensors for a longer time. Furthermore, holographic sensors can operate at near-infrared range, which offers the possibility of readout by existing consumer technologies (i.e., smartphone cameras).[129,130]

Prototype contact lenses incorporating holographic sensors have been fabricated by photochemistry and photopolymerization. A hydrogel matrix was prepared by co-polymerizing acrylamide (AAm), 3-(acrylamido)phenylboronic acid (3-APB) and *N*, *N*′-methylenebisacrylamide in the presence of a photoinitiator (2,2-Dimethoxy-2-phenylacetophenone (DMPA)) (**Figure 8**) over a silanized glass slide under UV light to form a 10 µm thick film. The polymer matrix was impregnated with silver nitrate solution and submerged into lithium bromide solution containing photosensitizing dye to form photosensitive silver bromide crystals. In "Denisyuk" reflection mode, this film was subsequently exposed to a pulse of a laser beam to record a holographic image of a mirror. The standing wave formed as a result of the incident and the reflected waves creates alternating bright and dark fringe regions. In bright fringe regions, the laser light is absorbed by the photosensitizing dye and silver bromide; hence, this renders bright fringe regions susceptible to a developing agent. These regions were subsequently developed, followed by a rinse in a stop bath. To incorporate the holographic film into a contact lens, it was removed from its substrate by an alkaline solution. To ensure that the film maintained a constant thickness, it was treated with PVA contact lens formulation prior to film removal. After removal, a small circular portion of the holographic film was cut out and incorporated into a PVA contact lens formulation, cured and autoclaved.[50,127] When the holographic diffraction gratings are illuminated with a white light source, they diffract narrow-band (monochromatic) light as governed by Bragg's law.

When glucose binds to the *cis*-diol groups of 3-APB, the polymer matrix swells due to an increase in Donnan osmotic pressure, altering the lattice spacing of silver nanoparticles in the polymer matrix.[50,128] However, boronic acids can bind with a range of *cis*-diols including a variety of carbohydrates and hydroxy acids that are found in tear fluid, such as lactate that can interfere with the glucose measurements.[131] Due to the comparable concentrations of lactate and glucose in tear fluid, lactate is a significant interferent, which can lead to false positives. However, besides glucose, other sugars are not found in high concentrations in tear fluid. Some are present as glycoprotein macromolecules, but these do not interfere with the measurements as they are unable to diffuse into the hologram and bind to the *cis*-diol groups.[96] An in vivo trial to test the efficacy of a holographic sensor in detecting glucose in tear fluid was carried out by anaesthetizing a rabbit and subcutaneously implanting the hologram below the subject's eye. A 25 nm red Bragg shift was measured in ≈3 min and converted

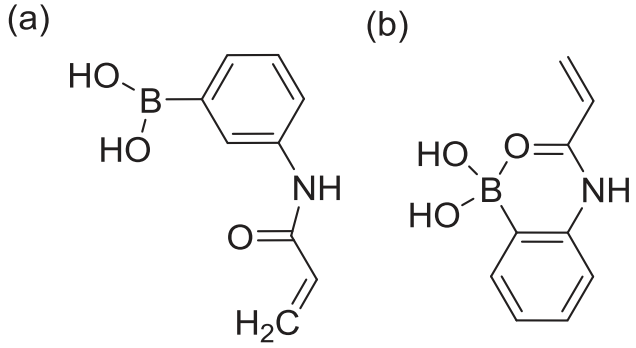

Figure 8. Boronic acid derivatives used with holographic contact lens sensors for monitoring glucose concentrations in tear fluid. a) 3-(Acrylamido)phenylboronic acid (3-APB) and b) 2-acrylamidophenylboronic acid (2-APB).

to a concentration of glucose in tear fluid, which was correlated with the glucose concentration in blood.[132]

In order to reduce lactate interference, the mole fraction of 3-APB was optimized at ≈20 mol%, which increased the sensitivity to glucose compared to lactate. Increasing the fraction of 3-APB up to 20 mol% produced an improved Bragg shift as a higher concentration of boronate anions will be formed in the polymer matrix upon binding with glucose. However, increasing the mole fraction of 3-APB over 20 mol% increases the hydrophobicity of the hydrogel, resulting in less swelling in aqueous solutions and a weak Bragg shift.[50] In addition, using 2-acrylamidophenylboronic acid (2-APB) (Figure 8) co-polymerized with co-monomers such as (3-acrylamidopropyl)trimethylammonium chloride instead of 3-APB leads to a lower dependency on pH over the physiological range, and a negligible interference from lactate.[133] Other analytes such as metal ions and pH can be monitored using holographic sensors.[134–136] Holographic sensors offer a generic technique for the fabrication of diffraction gratings in contact lenses. They may also be printed onto polymer matrices.[137] Since laser light is used to record the diffraction gratings, there is flexibility in producing these sensors across a wide range of materials quickly, while also permitting control over the angle of diffraction, the diffracted light wavelength, and the characteristics of the image.

2.3.3. Colloidal Crystal Arrays

Self-assembly of polystyrene particles has been utilized to construct photonic crystal arrays that are sensitive to variations in glucose concentrations in tear fluid. Photonic crystal array-based sensors consist of a colloidal system embedded within a polymer matrix that diffracts light in the visible spectrum. As with holographic sensors, this platform technology provides a scalable solution for the fabrication of optical sensors for quantitative glucose measurements without the use of an organic dye.

Fabrication of colloidal crystal arrays involves the self-assembly of highly charged monodispersed polystyrene nanospheres within a crystalline colloidal suspension, which form the photonic crystal templates.[138] Asher et al. pioneered boronic acid recognition chemistry with colloidal crystal arrays. These arrays were embedded within a polyacrylamide-based contact lens matrix, which was functionalized with boronic acid derivatives such as 4-amino-3-fluorophenylboronic acid and 4-carboxy-3-fluorophenylboronic acid (**Figure 9**). These boronic acid derivatives allowed sensing glucose at physiological pH values. At 1 mol/L glucose, the 4-acetamido-3-fluorophenylboronic acid derivative and 3-fluoro-4-*N*-methylcarboxamidephenylboronic acid derivative have apparent pK_a values of 5.8 and 5.15, respectively. The boronic acid groups bind to glucose, forming a "sandwich-like" complex, which results in additional crosslinkages within the contact lens matrix. The measurement mechanism utilizes this effect: as the cross-linkages form, the acrylamide matrix shrinks, which alters the lattice spacing of the colloidal array, and hence the diffraction of light from the colloidal dispersion changes. This blue Bragg peak shift is proportional to the concentration of glucose in synthetic tear fluid. The sensitivity and response times of the device were optimized,

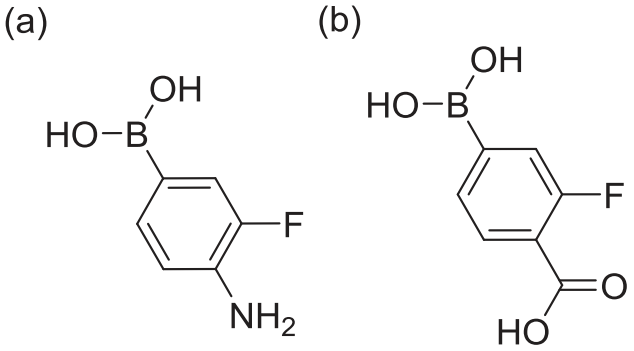

Figure 9. Boronic acid derivatives used with colloidal crystal array contact lens sensors for monitoring glucose concentrations in tear fluid. a) 4-amino-3-fluorophenylboronic acid, and b) 4-carboxy-3-fluorophenylboronic acid.

which resulted in changes of concentrations of glucose at rates comparable to those expected in blood, with a delay of 5 min upon addition of glucose solution (0.15 mM) at pH 7.4 and 37 °C (≈11 nm diffraction blue shift).[139] The detection limit of the device was ≈1 μM in synthetic tear fluid at pH 7.4,[138] which was formulated according to the Geigy formula.[140] For clinical use, a readout system that involves the user looking into a mirror and comparing the color of the diffracted light with a calibration chart was envisioned. Self-assembly of photonic colloidal crystal arrays offers flexibility across many design parameters such as base materials and functional groups, as well as having potential manufacture scalability.

2.3.4. Electrochemical Sensors

Fabrication techniques that are well established in the semiconductor industry have been used to construct microsystem-based sensors that can be incorporated into contact lenses. These systems can exhibit a high selectivity by using enzymes that can facilitate the reactions that are involved in electrochemical sensing. These sensors employ standard 3-electrode systems to quantify the concentration of glucose in tear fluid. Using glucose oxidase (GOx), an electrical charge is created due to the formation of an enzyme-substrate complex, and subsequent product formation and release, via reactions (1) and (2). The magnitude of the charge created is proportional to the concentration of glucose oxidized.

$$\text{D-glucose} + O_2 \xrightarrow{\text{GOx}} H_2O_2 + \text{D-gluconolactone} \quad (1)$$

$$H_2O_2 \rightarrow 2H^+ + O_2 + 2e^- \quad (2)$$

An electrochemical contact lens sensor comprising an amperometric unit monitored the concentration of glucose in artificial tear fluid.[141] The circuitry was fabricated using photoresist and thin metal deposition onto a poly(ethylene terephthalate) (PET) film, and subsequently heat molded into a contact lens shape (**Figure 10**). The sensing material was GOx immobilized onto a titania sol/gel membrane covered by a layer of

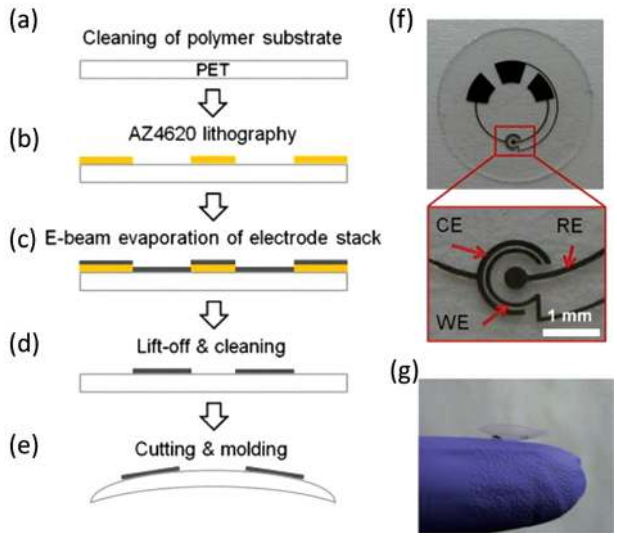

Figure 10. The fabrication of an electrochemical contact lens sensor. a) Cutting a PET film into 100 mm wafers using a CO_2 laser, and then cleaning the wafers with acetone, IPA, and DI water soaks which where subsequently spin dried, followed by an O_2 plasma treatment. b) The wafers were lithographically patterned using AZ 4620 positive resist by spin coating at 4500 rpm for 30 s. c) The metal electrodes were formed, by e-beam evaporation of a 10 nm Ti/20 nm Pd/100 nm Pt stack under high vacuum conditions. d) Lift-off, in acetone, and subsequent cleaning in acetone, IPA, and DI water. e) Release by laser cutting of the PET circles and heat molded into a contact lens. f) The L-lactate contact lens sensor's (below) flat substrate with sensing structure, interconnects, and electrode pads (counter electrode (CE), working electrode (WE), and reference electrode (RE)) for connection to the external potentiostat. g) The completed L-lactate contact lens sensor. Reproduced with permission.[35] Copyright 2012, Elsevier.

Nafion. A computer chip was developed for wireless (2.4 GHz) readout, and was powered with far-field electromagnetic radiation using a wireless device that supplied energy (3 μW) to the sensor at 15 cm. The prospect of powering the sensor using near-field inductive coupling was also investigated.[142] This approach required a large inductance/transformer, which was difficult to incorporate into a contact lens as all components are constrained by size. The readout delay was 20 s and the method had repeatability over three trials and a sensitivity of 10 μM over 0.1–0.6 mM glucose concentrations was measured. The sensor had low interference from urea, ascorbic acid and lactate; however, at low glucose concentrations (≤0.3 mM), the current produced by interferents had comparable magnitude (≈38 nA) to that produced by glucose.

Photovoltaic (PV) cells have also been used to power electrochemical contact lens sensors.[143,144] The PV cells were fabricated using highly doped silicon-on-insulator wafers and photolithography, and were bound to biocompatible PET contact lenses. A maximum power conversion efficiency of 1.24% and a mean short circuit current of 11.5 μA was produced by the PV cells, which was sufficient to power the contact lens sensors. These contact lens sensors were tested in vivo on live rabbits and no adverse effects were observed.

Another electrochemical contact lens sensor for monitoring tear fluid has been tested in vivo with rabbits.[145] The sensor incorporated a flexible microelectrode system, bound onto a PDMS layer. GOx was employed as the sensing material and immobilized onto the electrode by a coating of the co-polymer mixture 2-methacryloyloxyethyl phosphorylcholine and 2-ethylhexyl methacrylate to prevent leakage (**Figure 11,12**).[145] The sensor tracked blood glucose concentration with a 10 min delay. The device also underwent trials with synthetic tear fluid, and could detect glucose over the range 0.03–5.00 mM. An improvement on this device used 3D architectures with gold electrodes, microfabricated onto a PDMS film. The 3D pillar-like geometries provided an enhanced electrode surface area up to 300% greater than the 2D analogues. This sensor had a sensitivity of 0.04 mM for in vitro glucose measurements.[146]

In addition to glucose detection, lactate sensors have also been developed using the same principles with lactate oxidase (LOx) via reactions (3) and (2). With a sufficient potential difference, the hydrogen peroxide produced can be oxidized at a platinum electrode.

$$\text{L-lactate} + O_2 \xrightarrow{\text{LOx}} \text{pyruvate} + H_2O_2 \quad (3)$$

An amperometric L-lactate sensor was incorporated into a PET contact lens. The contact lens was lithographically patterned using a positive resist (AZ 4620) to incorporate circuitry.[35] The LOx was immobilized throughout the sensing area by covalent bonds between the enzyme and bovine serum albumin (BSA) using glutaraldehyde. The sensors were connected to external circuitry with wires, and tested using artificial tear fluid; however, the sensor can potentially be powered wirelessly. The sensor had a response time of 35 s with a resolution of 53 μA/mM cm^2 to lactate concentrations in tear fluid at physiological conditions.[35] There was no interference from glucose or urea; however, ascorbic acid, which is present in tear fluid, interfered with measurements at 50 μM. Ascorbic acid interference was prevented by designing a dual sensor. The dual sensor was fabricated from Ti/Pd/Pt stack as electrode material, and it was functionalized with PBS with LOx, BSA and glutaraldehyde, for recognition of both L-lactate and ascorbic acid. As a control, the second sensor was treated with PBS containing BSA and glutaraldehyde for exclusive detection of ascorbic acid, hence it allowed compensatation for the interference signal. Sensor stability is important and this may be compromised by the temperature-dependent activity of LOx. However, over the range of physiological and storage temperatures, LOx maintained its activity over 24 h. Electrochemical contact lens sensors are amenable to high-volume manufacture. In conjunction with high selectivity and rapid readout, the electrochemical sensing system is well suited to potential contact lens sensor diagnostics.

3. High-Volume Manufacturing

Prior to designing a contact lens sensor it is advantageous to consider the existing manufacturing techniques to achieve a degree of compatibility. Hence, understanding the manufacturing techniques optimizes the design requirements and reduces time to market. The early contact lenses were manufactured from blown glass, and later by grinding and polishing glass into the desired configuration.[147] After the development

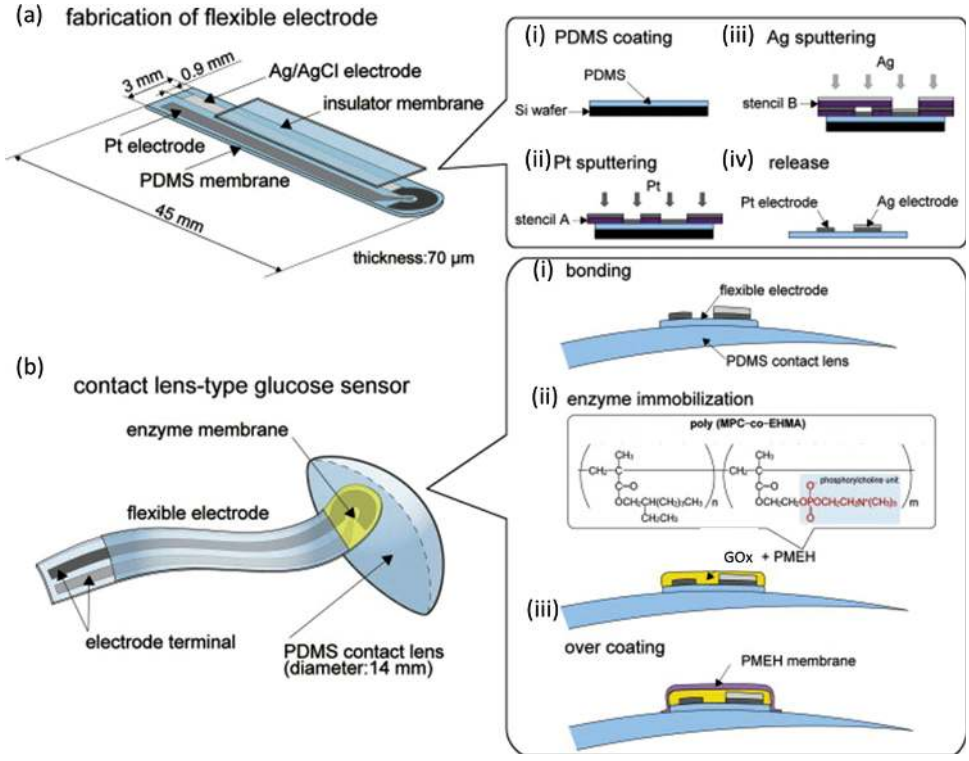

Figure 11. Fabrication method of the glucose contact lens sensor. a) The electrodes (a 200 nm thick Pt working electrode and a 300 nm thick Ag/AgCl counter/reference electrode) were formed onto a 70 μm thick PDMS membrane. b) The flexible electrodes were bonded onto the surface of the contact lens using PDMS and then GOx was immobilized with PMEH. Reproduced with permission.[145] Copyright 2011, Elsevier.

of modern contact lens materials, three main manufacturing techniques have emerged: i) lathing, ii) spin coating, and iii) cast molding.[148]

Lathing involves cutting out and polishing the contact lens from a cylindrical disk of dehydrated polymerized soft contact lens material. The cylindrical polymer disk is spun at up to 10 000 RPM and the desired amount of material is removed to form the contact lens.[149] The product is then polished with fine abrasive paste, oil, and a small cotton ball rotating at high speed. The method can also be used to manufacture rigid lenses by using the appropriate non-hydrated polymerized cylindrical disk. This method is applied when a wide variety of optical lens powers is required with different shapes, and a small batch size, that is, toric lens manufacture.[148]

Spin casting employs a single concave mold piece to facilitate the formation of the front lens surface and allows the contact lenses to be manufactured, hydrated and packaged in a single automated process. There are three main stages: i) mold wetting, ii) lens spinning and curing, and iii) lens hydration and release. In the first stage, the mold is dosed with a monomer mixture, wetting the entire surface to prevent voids. The monomer mixture is spun about the mold's central axis, under an inert atmosphere, at a controlled rate of >600 rpm during the second phase.[108] The centripetal forces acting on the monomer mixture generate the back surface of the contact lens. The monomer mixture is then cured by UV or heat to form the contact lens. For lens hydration, the product is immersed in an aqueous solution, which causes it to expand and release from the mold.[108] Due to the generation of the open surface by the centripetal force, it is parabolic in shape, which is a compromised profile relative to the spherical cornea. This method optimizes the balance of manufacturing cost with high product quality, for both user comfort and function, and is appropriate when a large batch size is required.

Cast molding uses both concave and convex mold pieces to form the front and back surfaces of the contact lens. The first step is to dose the concave plastic mold with a monomer solution, forming the front surface of the lens. The second convex mold is then placed on top of the first, forming the back surface of the lens. The monomers are cured by UV or heat to form the contact lens. This method is suitable for the largest batches, and when precise profiling of the contact lens is required for both front and back surfaces.[108,148]

Spin casting and cast molding have similar complications due to the use of a monomer mixture. Any O_2 present manifests itself as an induction period during which polymerization does not occur due to the preferential reaction of the initiator with O_2. The O_2 can be removed by sparging with

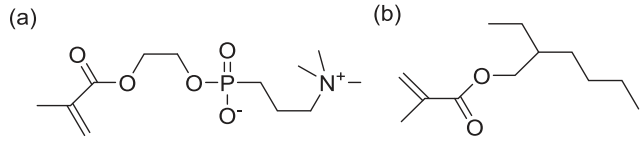

Figure 12. The polymer matrices used in the immobilization of GOx in electrochemical sensing. The copolymers a) 2-methacryloyloxyethyl phosphorylcholine and b) 2-ethylhexyl methacrylate prevented GOx leakage.

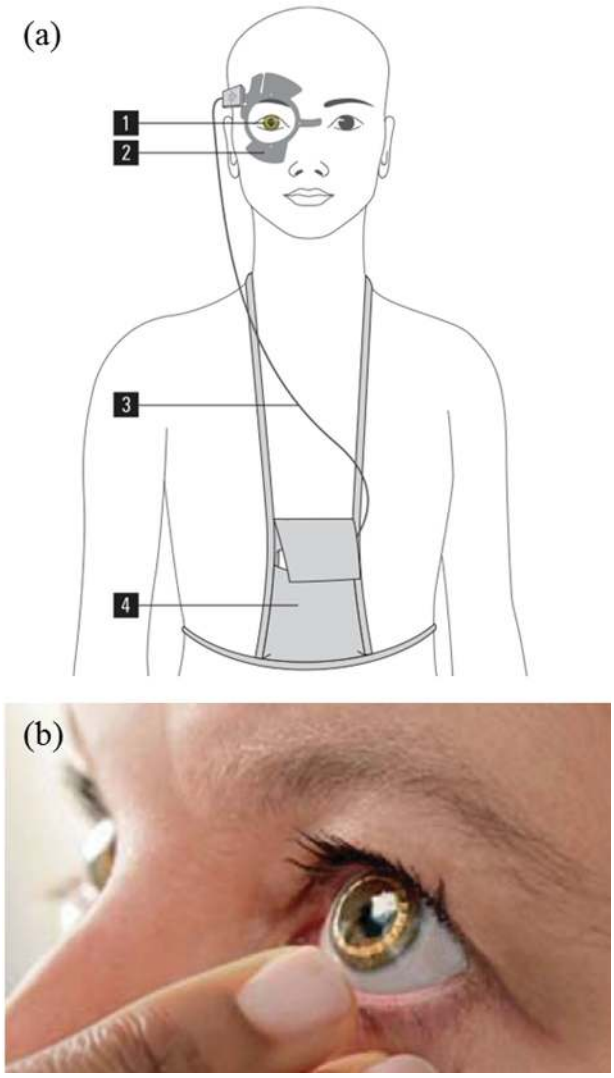

Figure 13. The SENSIMED Triggerfish holds Class IIa CE approval. a) The contact lens sensor transmits IOP measurements wirelessly to the antenna unit, which transmits the data to the data recording unit device. Data from the recording device is sent to a practitioner's computer for analysis. 1) The sensor; 2) wireless telemetry system; 3) cable that transmits data from the wireless telemetry system to the recorder unit; and 4) the data recording unit. b) The contact lens sensor worn in a human eye. Reproduced with permission.[156,157] Copyright 2013, Sensimed AG.

N_2, applying a vacuum, or adding an excess of initiator species to scavenge the O_2. However, small quantities of O_2 can diffuse out of the molds and into the monomer mixture, which can affect the lens surface characteristics such as its stickiness.[108] This issue makes the contact lens prone to contamination and results in poor handling. Due to using two mold surfaces as opposed to one, cast molding is more susceptible to this manufacturing defect than spin casting. Other potential defects are the formation of voids due to incomplete wetting of mold surfaces, which results in reduced yields. Shrinkage can occur during polymerization, which effects the fit and optical focus of the contact lens,[108] and is typically 10–20% of the lens volume.[148]

4. A Case Study in Intraocular Pressure Sensing

Sensimed AG (Switzerland) produces the SENSIMED Triggerfish, which is a single-use, soft contact lens that measures ocular dimensional changes that are related to IOP and currently holds Class IIa CE approval (**Figure 13**). This device is used in select centers for clinical management of glaucoma patients, but is not on sale in the US.[150] The sensing material consists of two Pt–Ti sensing-resistive strain gauges capable of recording dimensional changes in the area of the corneoscleral junction, which are correlated with IOP. A microprocessor in the contact lens sends a signal proportional to changes in the strain gauges to a wireless readout device attached to the user's waist, which also wirelessly powers the contact lens. The sensor records IOP-related dimensional changes for up to 24 h, with a maximum of 288 measurements. The sensor remains active when the patient is sleeping, which is an advantage as peak IOP may occur during sleep. However, a complication is that during sleep corneal swelling occurs due to decreased O_2 flux to the cornea because the eyelids are closed. The contribution of wearing soft contact lenses to corneal swelling, which is the ratio of the measured to baseline corneal thickness, is well established. This swelling with ordinary lenses is measured up to 3.2% greater during lens wear than without.[151] Wearing the SENSIMED Triggerfish overnight does not cause statistically significant differences in corneal swelling than when not wearing the device.[151] However, the SENSIMED Triggerfish® causes greater corneal curvature irregularities that are due to a center thickness of 585 μm, which is greater than most soft silicon-based contact lenses (<200 μm) resulting in a lower oxygen transmissibility to the cornea.[152,153] The effect of these corneal curvature irregularities may modify the IOP signal recorded.[153] Readout is given as an arbitrary unit (not in mmHg), so clinical interpretation of the result is difficult and it cannot be worn in both eyes simultaneously.[154] The contact lens sensor is safe and comfortable.[155] However, further investigations into the other variables that may affect the readings of the SENSIMED Triggerfish, including corneal and scleral rigidity, should be undertaken before general sale.

5. The Market and Regulations

An effective sensing technology should be combined with an understanding of physiology and visual performance of the eye, clinical development and regulations, as well as commercialization of contact lenses to develop a successful product. The contact lens market has already established norms and standards, and therefore this expertise can be leveraged and applied to contact lens sensors. **Table 4** shows the major players in the contact lens market and **Figure 14** shows their market share. Recently, Novartis announced that its eye care division Alcon has partnered with Google Inc. to in-license its electrochemical glucose sensor technology for ocular diagnostics.[12] Google's expertise in the miniaturization of electronics complements Novartis' pharmaceuticals and medical technologies to develop a glucose contact lens sensor and accommodative vision correction device for presbyopia. Hence, it is expected that such

Table 4. The major players in the contact lens market, their acquisitions and year of entry.

Contact lens manufacturer (year of entry)	Acquisitions
Bausch & Lomb (1971)	Polymer Technology
	Custom Tint Labs
	Unilens
Novartis (CIBA Vision) (1980)	Technolas Perfect Vision
	Alcon (merger completed in 2011) [158]
	Wesley Jesson
	Barnes Hind/Hydrocurve
	Syntex
	Precision Cosmet
	American optical lenses and solutions
	Burton Parsons
	LenSx
Johnson & Johnson (Vistakon) (1981)	Frontier
	Danalens
X-Cel (1994) [159]	Flexlens
CooperVision (1980)	Union
	Coast vision
	Biocompatibles
	Ocular Sciences

*Acquisitions pre 2005 taken from Barr 2005[160] unless given otherwise
** Year of entry taken from Schifrin and William 1984[161] unless given otherwise

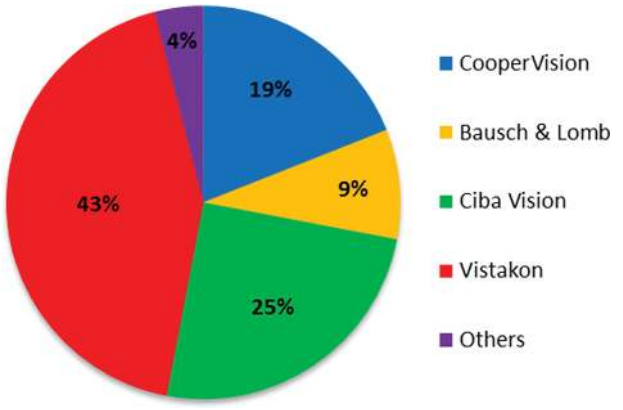

Figure 14. The market shares of the major players in the soft contact lens market (2014). Data taken from the literature.[162]

collaborations will continue across the market to accelerate the entry of contact lens sensors to consumer products.

To gain marketing approval for contact lens sensors, it is important to understand the governing regulations. Incorporation of regulatory requirements into the product design can accelerate the commercialization of contact lens sensors. Device regulations are generally in place to guarantee that there are no negative health effects for the patients, and to limit the frequency of Type I/II errors.[163] The marketing approval also specifies the manufacturer's claims that effectively define the potential patient population. There are three regulatory classes for medical devices, Class I being the lowest risk and least regulated, to Class III that are the highest risk and are subject to the most stringent regulations.[164,165] **Figure 15** illustrates the process of gaining an FDA approval, which can take up to a year after clinical trials. Device classification depends on the performance of the sensor and its materials. Notably, the FDA has classified diagnostic contact lens sensors made of PMMA as Class II.

6. Future Prospects

Having a sensor performance that meets clinical requirements is not always sufficient to guarantee consumer acceptance. A vital factor for consumer acceptance is usability, which in this context encompasses aesthetics, ease and convenience of powering/readout, shelf life, wear schedule, and cleaning. It is widely accepted that contact lenses are aesthetically pleasing in comparison to alternative wearable technologies due to their minimalistic nature. Wireless powering/readout is paramount in ease and convenience of use. Powering the contact lens sensors wirelessly has so far only been achieved using electromagnetic radiation with a high frequency, which may have negative health effects. Therefore, the prospect of using near-field inductive coupling at lower frequencies to power the sensor appears attractive. However, miniaturization of the components is necessary before they can be integrated into a contact lens.[7,8,142,166,167] Biological fuel cells are an alternative power source for electronic contact lenses that are convenient as they power the sensor in situ, negating the need for an external powering device.[168] A direct electron transfer and cofactor-free, non-toxic enzymatic fuel cell operating with basal tears was fabricated. At the anode, glucose was oxidized to gluconolactone by homogeneous *Corynascus thermophilus* cellobiose dehydrogenase, with the transfer of 4 electrons to the anode. At the cathode the bioelectroreduction of oxygen to water occurred, transferring 4 electrons to *Myrothecium verrucaria* bilirubin oxidase. This cell produced 0.57 V and ≈1 μW/cm^2 maximum power density, which is sufficient to power low-power electronic devices by utilizing the glucose and oxygen in tear fluid.[168] Alternative powering mechanisms for contact lens sensors can be found elsewhere.[169]

Bluetooth connectivity, which is a well-established technology, is a convenient method through which to transmit readouts wirelessly because many common devices including smartphones, tablet computers and laptops, are equipped with this capability.[170] However, security and health concerns are potential drawbacks of this technology. The security vulnerabilities of Bluetooth systems have been examined, and the manipulation of the readout to the consumer could prove harmful.[171] The health effects have been investigated, and although Bluetooth has a weak transmitting power, there remains open controversy in both science and the public on the implications of Bluetooth on human health.[172] LEDs have been incorporated into contact lenses, and may serve as an alternative readout method.[143] The light is directed away from the user's eye, and is powered

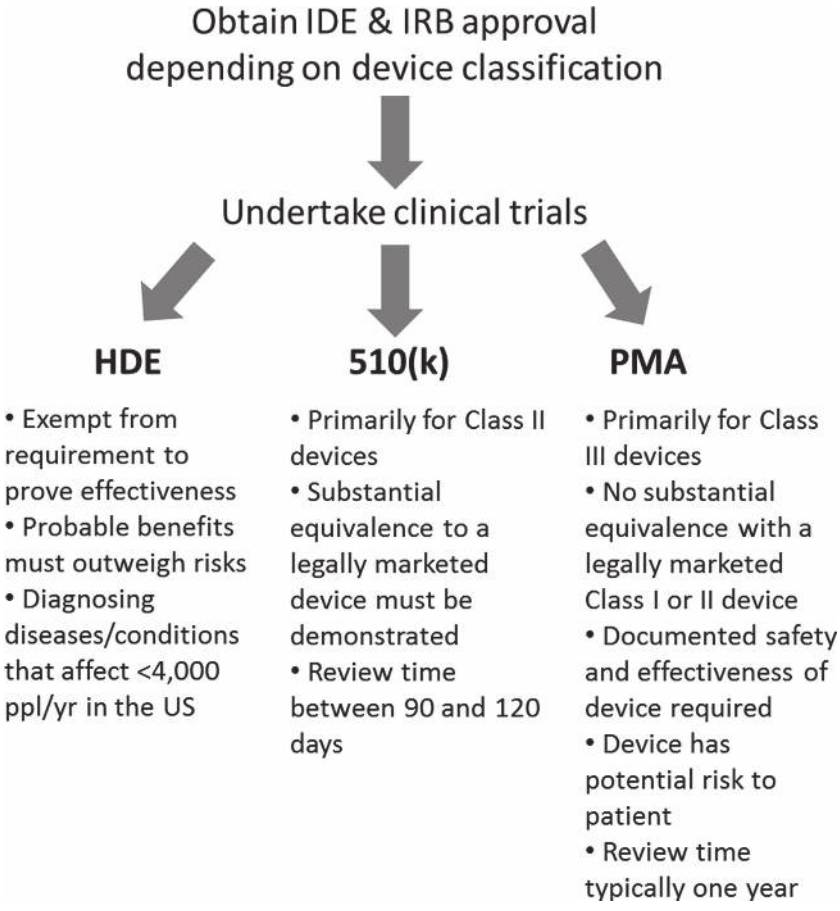

Figure 15. The FDA process to gain a marketing approval. The Investigations Device Exemption (IDE) is required to conduct clinical trials. If the device poses a significant risk, approval from the Institutional Review Board (IRB) is also required prior to conducting clinical trials. The Humanitarian Device Exemption (HDE) is irrelevant for the diseases and conditions listed in Table 3. PMA is the premarket approval.

in situ. Due to the optical readout, a secondary computational device is unnecessary, as using a mirror or third party can alert the user, which increases convenience.

The shelf-life of the contact lens sensor is limited by the sensing material. When enzymes are used, degradation of enzyme activity can occur due to light exposure and temperature effects. This instability has partly been overcome by encapsulating the enzymes. Due to the infancy of this field, existing contact lens sensors have only been tested over 24 h, which necessitates a daily wear schedule that may not suit everybody's needs. In addition, emerging technologies may increase the probability of user acceptance by providing additional benefits. For example, materials such as graphene and other nanostructures can provide miniaturized and thinner circuitry, opening up the possibility of multiplexed sensors with reduced power consumption.[173,174]

7. Conclusions

Contact lenses have seen widespread use in vision correction, therapeutics, and cosmetics. Advances over the last decade have opened up the new field of contact lens sensors, which are one of the most promising emerging sensor platforms as they are minimally invasive and can monitor biomarkers continuously. This has led to applications in diabetes management by measuring the glucose concentrations in tear fluid, and glaucoma diagnosis by monitoring intraocular pressure, but the scope of use of contact lens sensors is wider due to the high number of biomarkers in tear fluid and the eye. Capacitative, piezo-resistive, mechanical strain gauge, and microinductor intraocular pressure sensors have been used to diagnose glaucoma. For example, the SENSIMED Triggerfish, which uses mechanical strain gauges, has gained regulatory approval and entered the market. Fluorescent, holographic, colloidal crystal array, and electrochemical sensors have been used to measure the glucose concentration in tear fluid in vivo with high reproducibility.

In addition to a sufficient selectivity, sensitivity, and reproducibility, contact lens sensors must also be practical devices to ensure patient compliance. Synthetic sensing materials, such as boronic acid derivatives, require improvements in selectivity, and biological sensing materials, such as enzymes, require modifications to enhance reproducibility due to degradation and drift. Hence, there is a need for the development of sensing materials with high selectivity and sensitivity as well as reversibility with no hysteresis. In addition to improvements on existing sensors, we expect the use of novel materials such as graphene, quantum dots, plasmonic nanoparticles, liquid crystals and bioinspired photonic structures. In addition to advances in current fabrication techniques, we expect the investigation of alternatives such as layer-by-layer stacking, inverse opals, block copolymers and nanocomposites, and incorporation of 3D printing for prototyping. Readout time should be minimized in cases where a delay could be harmful such as in diabetes. Attempts to reduce this delay have so far included developing materials to mitigate time dependent processes such as diffusion, and deriving compensating mathematical algorithms. The development of powering and readout mechanisms is essential for a practical contact lens sensor. They should be wireless and utilize existing consumer devices such as smartphones, tablet computers and laptops. Recent efforts have included using far-field electromagnetic radiation, biological fuel cells, photovoltaic cells, contact lens-integrated LEDs, and Bluetooth connectivity. Many of these techniques are limited by the size of the circuitry required, and therefore the potential to miniaturize the electronic components, for example, through graphene-based circuitry, may be explored.

For contact lens sensors to fulfill their potential, the relationship between ocular biomarkers and disease, particularly the relationship between tear and blood concentration,

should be established. Additional complications which should be accounted for are the movement of the contact lens across the eye, particularly for intraocular pressure measurement, blinking, and interpersonal eye features.

Contact lenses are widely used for vision correction. Hence, leveraging this platform to treat other diseases and conditions is advantageous. Contact lens sensors can diagnose diseases and conditions ranging from diabetes to glaucoma, where minimal invasiveness and continuous monitoring are required. The ability to pair the sensor with mobile medical applications may allow real-time data logging and transfer to clinicians for efficient diagnosis. This platform is not limited solely to diagnostics, and in the future, we expect this platform to integrate treatment capabilities such as drug delivery.

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