TRENDS IN OPTICS FOR LIFE SCIENCE

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INTRODUCTION

The impact SARS-CoV-2 has had on the medical, pharmaceutical and life science industries in the last few years is evident. Though it has created a huge boom in the medical diagnostics, the life science and medical devices sector was already a growing market. The drive in recent years towards less invasive, digitalized, and accessible technologies has increased due to globally growing populations, ageing populations in developed countries, and as societies become progressively more health-conscious. Many of the technologies in this sector rely heavily on optics and photonics principles, and as their application requirements and markets change and evolve, so do those for the optical elements within them. From portable analysis equipment, to luxury wearables, to smartphone-based diagnostics, optics remain an integral part of innovation.

MINIATURIZATION IN MEDICAL AND HEALTH DEVICES

qPCR

The medical diagnostics market was kicked into a new gear level with the arrival of COVID-19. Suddenly, fluorescence-based technologies like Polymerase Chain Reaction (PCR) testing and ELISA antibody testing became crucial and required large-scale operations to be able to handle the sheer volume of tests being conducted daily. PCR techniques amplify small samples of DNA in order to analyse them in detail to help identify infectious agents. Many quantitative PCR (qPCR) screening systems rely on fluorescence principles in order to detect specific nucleic acid sequences such as those of pathogens. Most of these PCR machines nowadays are about benchtop size, but are not very easily transportable. Portable PCR machines, though already becoming commercialised in the mid-2010s, have become even more important for use in the field in the current global health climate, with accessible, low-cost versions required for developing countries[1].

In qPCR, fluorescent dyes are used to tag biological structures with high specificity. Each fluorescent dye has its own excitation wavelength where if exposed to light at this frequency, it will in turn emit longer wavelength light specific to the dye. The optical systems typically present in fluorescence detection PCR are very similar to a typical fluorescence imaging system, as outlined in **Fig. 1**, and involve excitation and emission filters centred on their respective wavelengths to purify the light signals going to the sample under observation and back to the detector, dichroic filters reflective on the excitation wavelength and transmissive on the emission wavelength to correctly direct the light signals, lenses to focus and shape any light source beams, and mirrors for alignment and beam steering.

Similarly, other medical technologies such as pulse oximeters, dermatoscopes, and ophthalmoscopes are also beginning to shrink in size. Newer models coming out on the market are becoming increasingly lightweight and more portable.

Pulse Oximeters

Optical pulse oximeters are used to measure the oxygen concentration in blood based on the principle that the absorption of light differs between oxygenated and deoxygenated blood. Oxygenated blood absorbs more IR and reflects more red light than deoxygenated blood. The reverse is also true, where deoxygenated blood absorbs more red light and transmits more IR light. Using this principle, most current pulse oximeters feature two LEDs of differing wavelengths, namely 660 nm and 880-940 nm. These are mounted opposite a photodiode between which a translucent part of the body (such as a finger or ear lobe) is placed (**Fig. 2**). The signal of red and IR light that is passed through the translucent tissue is read by the photodiode and from there, the level of oxygen is calculated. More recently however, some newer technologies have been developed whereby a CCD coupled with colour filters are used instead, to narrow down the spectral band detected, filter out any noise, and reduce uncertainty in readings[2,3].

Figure 1: Typical Fluorescence Imaging Beam Path

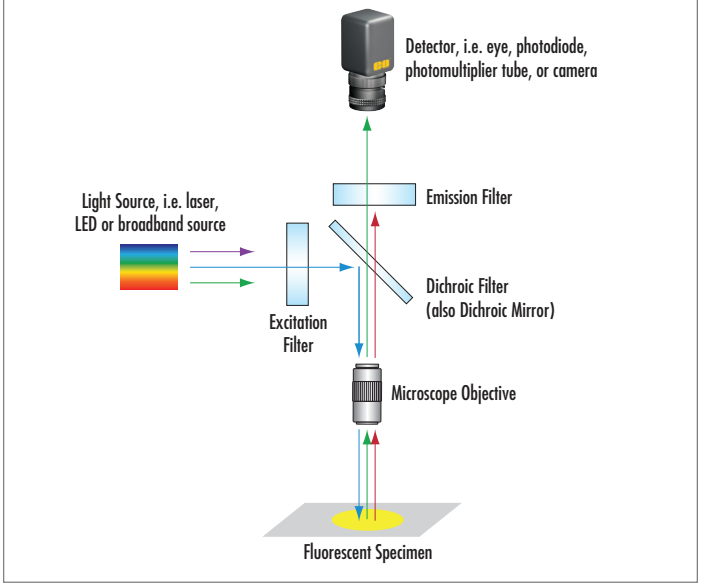

Figure 2: A portable oximeter is used to measure oxygen levels and pulse rate. Portable, low-cost life science devices particularly benefit from incorporating colour glass filters.

Dermatoscopes

Dermatoscopes, which are used to inspect and aid in the diagnosis of different skin complaints including melanomas, carcinomas, and dermatitis, work based on epiluminescence microscopy whereby the skin is magnified, illuminated, and visually inspected.

Typical dermatoscopes feature achromatic lenses for magnification, though newer models also include cross-polarizers to reduce glare and filter out any peripheral stray light. Whilst traditionally dermatoscopes have been handheld devices, miniaturisation has allowed for the creation of lightweight clip-on modules for digital cameras and even mobile phones, allowing for image capture and documentation[4].

Ophthalmoscopes

Additionally, ophthalmoscopes and other diagnostic devices that have traditionally also been large and bulky are experiencing similar treatments as dermatoscopes as far as miniaturisation and smartphone integration. Smartphone attachment ophthalmoscopes for diagnosing various eye ailments have been cropping up in the last few years[5]. Traditional "handheld" ophthalmoscopes consist of direct and indirect (binocular) versions, both of which consist of a co-axial light source and beamsplitter to shine light into the back of a patient's eye through the pupil and examine their optic nerve and retina.

Indirect versions also feature ulterior lenses to direct entering coaxial light at such an angle that it minimizes reflections and backscatter. However, the major drawback of these conventional devices in a 21st century world is their lack of digital storage and retrieval capacity. With larger and more complex models, ulterior lenses and filters are added in order to increase the diagnostic image resolution, and image capture is possible, but they are too large and heavy to be comfortably portable. An example image produced by such an ophthalmoscope is shown in **Fig. 3**.

In this vein, various attempts have been made at introducing smartphone-based systems with portable, clip-on attachments as a better, more accessible alternative to their analogue ancestors[6,7]. These systems would thus be able to capture and store diagnostic pictures and data, even in the field.

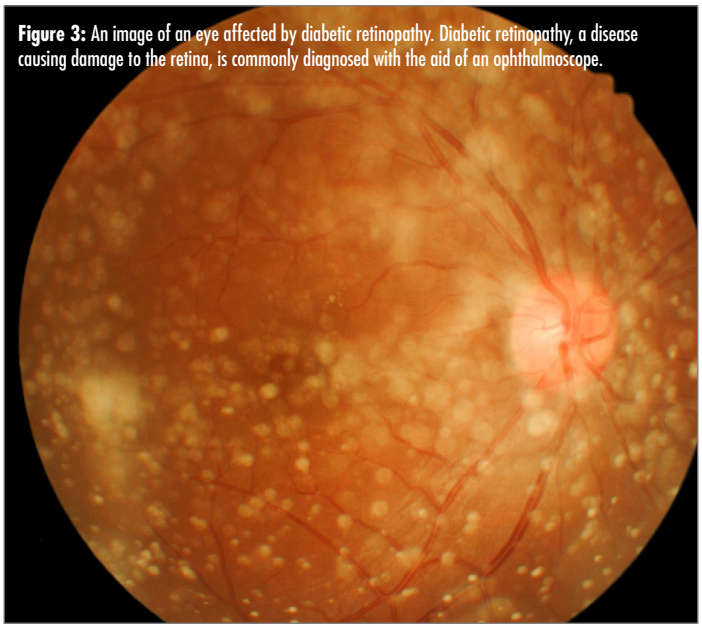

Figure 3: An image of an eye affected by diabetic retinopathy. Diabetic retinopathy, a disease causing damage to the retina, is commonly diagnosed with the aid of an ophthalmoscope.

Considerations on Miniaturisation of Optical Components

Of course, in all these cases, portability, size and weight factors are crucial. The smaller and lighter a machine, the smaller the optical component requirements. For researchers, but especially for original device manufacturers (OEMs) making these miniaturised devices, it is crucial to consider the implications of a smaller system on the optics involved.

One such consideration to make is around the clear aperture of the optic. Whilst larger optics can comfortably achieve clear apertures of 75-90+%, on a smaller scale this percentage may become reduced due to manufacturing challenges, decreasing the proportion of the useful area on the optic. The key to the solution is knowing when to cut and when to coat. Properly coating an optic of just a few millimetres in diameter can be difficult, so core drilling a larger, already coated part can be an alternative option. Laser cutters can also be used on polymer optics. The edge-melting effects this type of cutting induces, however, though not generally too much of an issue on a large part, is problematic on a small optic. On components of just a few millimetres in size, the edge surface area represents a higher overall proportion of the optic compared to parts even just half an inch in diameter. In this case, other cutting technologies that cause less material erosion along the edges, such as water jet cutting, may also be considered depending on the part being processed.

Additionally, handling, packaging, and cleaning become more problematic at smaller optic scales, particularly when the optic is but a few millimetres big. Here, the human eye struggles to visualise the optic to ensure proper handling and appropriate cleaning. Magnifiers and tweezers or vac-pens may be used, but this becomes fiddly and annoying, especially when handling a high volume of components. In this respect, more automation may be needed to improve speed and accuracy of inspection and packaging.

Thus, when developing miniaturized systems, especially for a high-volume market, it is important to find a well-established component supplier who can deliver the required quality and precision of products on a high-volume scale. It may be also prove beneficial to test out an off-the shelf component for testing beforehand to make sure the optics' properties align with requirements, and then ensure that the manufacturer has appropriate manufacturing capacities to repeat this quality on a volume scale.

CONSOLIDATION OF OPTICAL ELEMENTS

In recent years, and especially following the onset of the coronavirus/ SARS-CoV-2 pandemic, speed and ease-of-use aspects of medical devices have grown in importance. Demand for plug & play solutions with a high degree of "user friendliness" have increased , and so have the requirements to simplify the optical path thus reducing the overall number of optical elements present. One notable example of such a technology is flow cytometry; it plays a significant role in many applications such as infectious disease monitoring, virology, and immunology. Flow cytometers use fluorescent parameters to quickly analyse, count, or sort particles in solution. Single dissociated cells or particles are tagged with fluorophores or dyes and suspended in salt-based solutions. The cells flow past one or more excitation-wavelength lasers, and the resulting scattered light is directed by a set of dichroic filters to a specific detection channel with a bandpass filter centred on a specific emission wavelength in order to get a specific fluorescence readout. The light collected by the detector, usually a photomultiplier tube or photodiode, is then utilized to measure the

size or assess the granularity of the cells. For each different parameter that requires analysis, a different fluorescent dye is used. This means flow cytometers used in multicolour analysis will sometimes require multiple laser sources, possibly multiple detectors, but certainly multiple optical components.

Whilst it is possible to use one filter for every dye in the detection channel, switch each filter around, and look at each channel individually, this takes time. Using multi-band emission filters, filters with multiple bandpass peaks at different wavelengths, it is possible to reduce the quantity of components required and speed up the detection as it allows multiple channels to be viewed at the same time. This is applicable not only to flow cytometry, but to any multicolour fluorescence detection system including qPCR, fluorescence microplate readers or fluorescence microscopes[8].

Optimizing the system using fewer multi-band filters instead of multiple others also reduces the size, weight, and overall cost of the system. Another consideration to make, is though multi-band are generally more expensive than single-band filters due to the complexity of their coating, one multi-band remains cheaper than several single-band filters. It is also important, when selecting multi-band filters, to have clear separation between each channel so that the signals do not overlap (bleed through), and each channel needs to be carefully optimized for the fluorophores utilised which usually means that a custom optic will be required rather than a standard off-the-shelf part. Alternatively, it is also possible to reduce the number of optical elements within a beam path by replacing multiple spherical lenses with a single asphere. This will achieve a lighter, more portable design whilst achieving the same results as multiple spherical elements.

WAVELENGTH RANGE CUSTOMISATIONS

When working with fluorescence, applying a fluorophore to a molecule, antibody, or cell almost always results in some kind of interference with the biological processes of the sample. There are constantly new developments in fluorescence labelling such as the use of fluorescently-labelled nanobodies to interfere less with biological processes, and the development of dyes that excite and emit further in the NIR and IR. These constant developments are driving the design of new filter coatings to efficiently image and detect these newer dyes.

While filters for well-established fluorophores and dyes such as GFP are already commercially available off-the-shelf for fast system integration, the newer fluorophore wavelengths are not yet widely addressed in the market. In applications where these are crucial, it may be advantageous to consider wavelength customisation as an option, particularly for applications with volume quantities. Additionally, due to some fluorophores' excitation and emission spectra being very close together, it is important to select very steep or narrow bandwidth filters in order to effectively differentiate between the two or more signals for more accurate spectral readings. It is possible to find very narrow bandwidth or steep band filters commercially, though these are typically centred on other laser wavelengths like 488, 532 nm and 633 nm. As always, considerations must be made when deciding to go down a made-to-order solution. Firstly, it is important to consider what spectral transmission range is required. Although broadband filters are highly useful due to their flexibility, there is such a thing as "too much broadband". Very broad spectral ranges (for example, from UV all the way to mid-IR) are quite challenging to manufacture, and can become cost-prohibitive very quickly. Really, going to any extreme- range requests too deep into the UV or too far into the IR, or ultra-narrow bandwidths of just a few single nanometres are equally as tricky and expensive. The same principles can equally be applied to the blocking properties of a filter as "too broadband a blocking range" is just as difficult to achieve. A common misconception is that outside of the passband, all other wavelengths are blocked in both directions when in reality, this is never the case. At one extreme or the other, the blocking power of the filter will deteriorate, so it is important to consider at what spectral range a higher blocking level is most crucial, and to remain flexible on wavelengths outside of this range.

It is also important to consider bandwidth and wavelength when using multi-band bandpass and notch filters. If an application uses dyes with bands that are very close together and so requires the filters' coatings to reflect this, the complexity of the design of the coating is likely to be very time-consuming to the designer, and ultimately costly to the client. In these situations, it may be easier to use different, more discrete fluorophores instead.

CONCLUSION

Finally, it is evident that developments in life science and medical devices are placing tougher constraints and requirements on the optical systems and components within them. As this market continues to grow, the optics, photonics, and imaging sectors will follow. Equally, as advancements in optical technologies are made, they will, in turn, continue to drive further innovation in those same life science markets, inevitably demonstrating that the future truly depends on optics.

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