

Light Engines for the Life Sciences

Life science instruments present unique challenges to suppliers of light-generation equipment: Inexpensive, high-intensity, uniform, monochromatic light across a broad UV-VIS-IR spectrum is required. Competition is keen within today's \$50–60 billion biophotonics marketplace¹ to find commercial lighting solutions that provide for all of these requirements with a low-cost and durable emitter that, ideally, could be miniaturized for portability.

Today's commercial light sources include lamps, lasers, and light-emitting diodes (LEDs). Arc and tungsten lamps offer white light for a broad range of uses. Lasers are the brightest and most spectrally pure light sources, while LEDs offer by far the lowest-cost light source option. Manufacturers of lighting equipment have engineered solutions for multicolor emissions utilizing lamps based on these technologies: externally filtered lamps, bundled lasers, and LED arrays. However, no competitive technology employing these techniques provides a cost-effective solution for the range of bioanalytical tools used in today's life science marketplace.

While each of these solutions has advantages, some fundamental limitations are not addressed. Lamps are notorious for their spectral drift and short lifetimes. Lasers are restricted to particular wavelengths and require significant safety precautions. LEDs have limited spectral outputs and although considered an inexpensive lighting solution, when bundled to generate adequate intensity, costs associated with their implementation soar. Currently available commercial light sources compromise throughput and sensitivity, lead to frequent instrument calibration, and generate high cost of ownership. Consequently, instrument performance is constrained and overly priced.

Based on well-understood light pipe architectures, a light engine solution based on luminescent pipes has been developed (Lumencor, Inc., Beaverton, OR) to fill a void and satisfy the growing bioanalysis market. Lumencor light engines (patents pending) utilize solid-state luminescent materials, excitation sources, and optimized light management techniques. In order to best appreciate the relative merits of these lighting tools, a more detailed consideration of the competitive technical and business landscape follows.

Lamps

The primary light sources for PCR and Sanger sequencing instruments are tungsten halogen lamps, arc lamps, and lasers. The tungsten halogen lamp is a mature product, having been optimized for display and lighting applications. Improvements in its low light-collection efficiency or mitigation of high thermal emissions are unlikely. Durability and stability issues are also inherent and difficult to resolve. Its cost could come down, but only if driven by volume applications. This white light source with a width of over 1000 nm must be heavily filtered to create the 10–30 nm bands required in many bioanalytical instruments. As such, light output to the sample volume is quite low, on the order of a few hundred mW/cm², which relegates use of this lamp to low-throughput applications.

To overcome this limitation, instrument designs employ arc lamps. Figure 1 shows the typical emission curve of a metal halide arc lamp.² These high-power lamps generate large light levels. They also

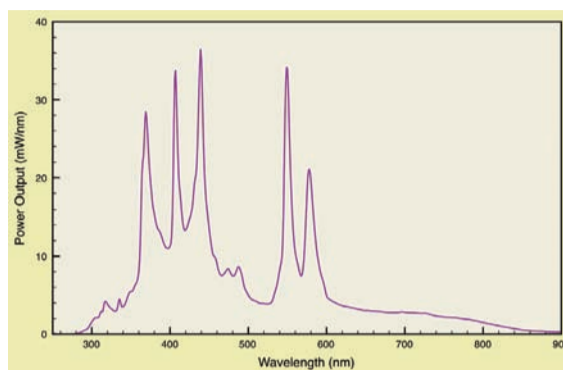


Figure 1 Typical metal halide arc lamp spectrum.

generate very broad white light that must be heavily filtered. Additionally, they create significant heat loads that must be managed to avoid compromising bioanalytical chemistry and instrument performance. Power instability due to the changing arc over its lifetime is a notorious problem. Arc lamps have been significantly improved due to proliferation in newer microdisplay-based projection televisions and business projectors. However, the availability of low-cost LEDs has diffused efforts to improve the arc lamp. Typically arc lamps, with their associated filters and spectral wheels, mirrors, controllers, and shutters, add as much as \$10,000 to the cost of a \$3000–\$5000 bulb.

Lasers

A common alternative to broadband lamps is the laser. One of the most frequently employed in bioanalyses is the air-cooled argon ion laser. A typical laser, for capillary-based sequencing, for example, provides 10–25 mW in a collimated, multiline beam. However, the laser color balance is inherently unstable; the power output of individual wavelengths varies over time and from product to product. Additionally, lasers are limited in the number of wavelengths available, and their speckle patterns are an obstacle for many applications in which field uniformity is important. They are nonlinear devices built to be run in a continuous mode. They are not readily modulated except by external mechanics. The failure mechanism of lasers is a catastrophic event. Typical replacement is required every 6–8 months. Replacement costs are a function of the wavelength generated and can range from a few hundred dollars to \$10,000, resulting in high annual maintenance costs. Serious safety concerns exist about their use by inexperienced users and in nontraditional testing environments.

As a consequence of the aforementioned shortcomings, competition from improved light sources will likely come from diode lasers and LEDs. Significant improvements in these semiconductor products have already resulted in

enhanced brightness, increased wavelength range, and cost reductions. However, diode lasers are primarily targeted for telecommunications and do not readily provide wavelengths in the visible and UV ranges of the spectrum, those required for bioanalyses. Techniques typically employed to generate these wavelengths, including frequency doubling, are unlikely to be employed for low-cost light engine designs. Difficulties with beam shape, power stability, and reliability are other issues yet to be resolved.

One exception receiving significant attention in newer instrument applications is the up-converted IR diode laser, the so-called diode pumped solid-state laser (DPSS). These lasers are less expensive, more powerful, and longer-lived than their predecessors. However, they too demonstrate certain limitations, namely, a limited number of emission lines and the need for external modulation, and they remain prohibitively expensive for many applications.

LEDs

LEDs are now available in a relatively wide range of wavelengths. However, their output is significantly broad so as to require filtering to achieve usable bandwidths on the order of 10–30 nm. Additionally, output in the visible spectrum is profoundly reduced in the yellow and green. This gap in usable wavelength range compromises their utility for bioanalyses where multicolor excitation throughout the visible spectrum is required. In many life science applications, sufficient light intensity cannot be obtained from individual LEDs, or from arrays of LEDs. Designs based on LED arrays lead to uniformity, stability, and durability issues. The spatially broad output results in lower collection efficiencies and contrast ratios. Their main advantage, low unit cost, is often offset by the cost and complexity resulting from additional components required for their implementation.

Light engines based on light pipes

Most important in the above discussion of traditional light technologies is that they cannot be readily improved for bioanalytical applications. The associated light engine market does not justify the investment necessary to overcome fundamental performance limi-

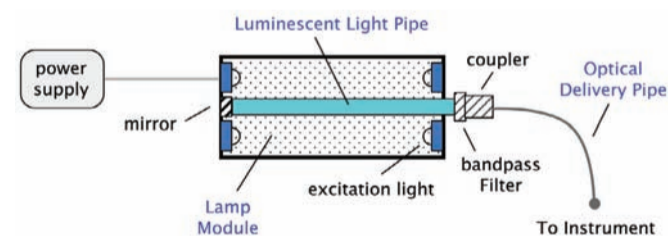


Figure 2 Schematic of a single-color light engine.

Table 1 General comparison of various sources used in light engines for bioanalyses

Light engine	Usable light	Uniformity	Temporal response	Heat generation	Durability	Cost
Arc lamp	Medium	Poor	None	High	Low	High
Laser	High	Poor	None	Low	Low	Very high
LED	Low	Poor	Fast	Low	High	Medium
Tungsten	Low	Poor	None	Medium	Low	Medium
Light pipe	High	High	Fast	Low	High	Low

TECHNICAL ARTICLE

Table 2 Lumencor light engine output vs light engines based on traditional sources for bioanalytical applications*

Specification	Sanger sequencing		Q-PCR		Flow cytometry		Fluorescence microscopy	
	Light pipe	Ar ion laser	Light pipe	Metal halide	Light pipe	Lasers	Light pipe	Metal halide
Intensity W/cm ²	~150–250	~150–250	0.5–1	0.2–1, very λ specific	~150–250	~150–250	<50	1–50, very λ specific
Wavelength	505 nm	Multiline	Four colors		Two colors		Four colors	
Bandwidth	10–30 nm	~26 nm	10–30 nm	~15 nm	10–30 nm	<5 nm	10–30 nm	~15 nm
Stability	0.1%	>1%	0.1%	>1%	0.1%	>1%	0.1%	>1%
Switching, msec	1	1–10, external shutter	1	40, external shutter	1	1–10, external shutter	1	40, external shutter
MTBF, hr	>10,000	<4000	>10,000	<1000	>10,000	<4000	>10,000	<1500
Price*	<\$3000	>\$5000	<\$7500	>\$10,000	<\$5000	>\$5000	<\$7500	>\$10,000

tations. As a result, in today's environment, analytical instrument performance and price are constrained by the light source. Moreover, the numerous manufacturers of lamps and lasers provide only a light source, not an integrated light engine. The **Lumencor** light engine is designed to provide a well-integrated, cost-effective subsystem that simplifies light subsystem implementation and does so with enhanced performance.

Light engines are light delivery subsystems that consist of a light source and delivery optics. A schematic is shown in *Figure 2*. Light is generated by exciting luminescence in a rod or light pipe, which may be driven by an e-beam, LEDs, or UV bulbs. The light pipe geometry integrates a significant fraction of the light, resulting in high external efficiencies. Efficiency is optimized by the design of the lamp module, including the excitation source and the geometric shape of the pipe. Increased power and intensity are obtained by scaling the light pipe and associated excitation.

The pipe, which consists of any of a variety of luminescent materials (e.g., rare earth doped glass, organic doped plastic, doped single-crystal fiber), is tailored to emit light in the UV through the IR range. In the case of the rare earths, emission is inherently narrow due to their atomic-like energy structure. Unwanted light is removed with a bandpass filter. The light output from the luminescent pipe is fed into an optical delivery pipe and directed into the analysis instrument or tool. Fast luminescence decay results in rapid switching, with no appreciable warm-up time for these long-lived, durable materials.³

Each color produced within a light engine requires a unique combination of light pipe and excitation source. Moreover, each application requires one of several different approaches to coupling light from the delivery pipe into sample volumes. As an example, the light in a Sanger sequencer is coupled to a row of 50- μ m-i.d.

capillaries. Quantitative PCR (Q-PCR) tools typically require uniform excitation in an array of wells within a multiple-well plate with no illumination of the interstitial spaces. Fluorescence microscopes need uniform light piped through the objective. Microfluidic chips employ unique detection capillaries and/or flood illumination of a plurality of channels, spots, or wells. To satisfy the range of instrument architectures, imaging and nonimaging optics are utilized.

While no one lighting solution can best satisfy every instrument architecture, light pipes meet or outperform the traditional technologies listed in *Table 1* on the basis of all figures of merit for all individual wavelengths. Single outputs, such as red from a diode laser, may be competitive. However, no family of outputs can be assembled that bests the pricing of the light engines described here. As an example, a **Lumencor** light engine can emit narrowband light exceeding 500 mW/color with intensities above 100 W/cm². Bandwidths as narrow as 10 nm are achievable. While such output power and overall emission intensity are impressive, the most significant figure of merit for quantifying the value of any lighting subsystem for bioanalytics is the intensity of high-quality illumination provided to the sample. This is dictated by the instrument design and sample volume and is clearly very application specific, e.g., fluorescent microscopy for live cell imaging vs for reading a chip designed for gene expression analysis.

A more detailed comparison of present-day light engines employed in a number of bioanalytical instruments with that of a light engine based on light pipes is shown in *Table 2*. Significantly, the relevant comparison is among proprietary lighting subsystems, not published specifications for a particular light source. Here, the most important figure of merit is intensity. The amount of fluorescent signal generated for a given analyte in each tool is defined by the S/N of the instrument. This ratio is optimized by maximizing the number of photons into the analysis volume and therefore by the illumination intensity delivered to the analyte in mW/cm² with the important caveat that the illumination not degrade the sample itself. Necessarily, performance is a function of instrument architecture. Examples include Sanger sequencers, which use individual argon ion lasers to simultaneously irradiate an array of up to 96 capillaries (50 μ m i.d.) fed electrophoretically from a multiple-well plate. Tools for Q-PCR most often require multicolor illumination of high-density multiple-well plates using mechanical shutters to switch among wavelengths. Multiplexed chip technologies and high-density microchannel approaches are even more demanding in terms of illumination intensity. Flow cytometry is typically carried out in serially addressed and spatially separate aliquots of a continuous flowing, focused stream. Finally, optical

microscopes for fluorescence detection require uniform, stable light, and their intensity requirements are essentially defined by the objective and the illumination area under interrogation.

Intensity values differ by orders of magnitude. This is a direct consequence of the instrument architecture and the corresponding illumination area dictated by the sample volume. As a result, highly multiplexed analysis like those performed using microarrays require very high intensities to satisfy numerous small illumination volumes. It should also be noted that intensity values have a wavelength dependence; this is much less an issue for light pipes. For example, while it is possible to achieve W/cm² intensity values for a band of light centered at 546 nm out of the metal halide lamp, much lower intensity values are produced in other colors such as cyan and red. Similarly, in the case of light engines based on LEDs, insufficient power is available between 520 and 590 nm. Relatively, light pipes achieve nonwavelength-dependent emissions due to the plethora of materials that are available to produce bands throughout the UV, VIS, and IR regions of the spectrum; not every band is possible, but as many as eight unique colors can be produced with comparable intensity and brightness. It is possible to integrate the luminescence for long rod lengths, yielding high emissions. Finally, because these bands are produced by individual emitters designed within independently controllable subunits, light engines have unique temporal and spatial properties that should enable new instrument designs and capabilities. Each color is uniquely operable such that modulation between colors and simultaneous irradiation by multiple colors is possible. These light engines make it feasible to provide controlled excitation for multiplexed analyses with no external mechanics, filters, or shutters. Exploitation of this solution for life science applications promises to enable highly multiplexed analyses in the next generation of low-cost, portable, high-density instrument platforms.

References

1. Lee, G.; Chu, K.; Conray, L.; Fix, L.; Lui, G.; Truesdell, C. *Optik & Photonik* June 2007, 2, 30–5.
2. www.exfo-xcite.com.
3. Bioanalytical instrumentation using a light source subsystem, US-2007-0281322-A1.

Mr. Conner is Vice President, Engineering; Dr. C. Jaffe is Vice President, Business Development; and Dr. S. Jaffe is President, Chief Executive Officer, **Lumencor, Inc.**, 15455 NW Greenbrier Pkwy., Ste. 210, Beaverton, OR 97006, U.S.A.; tel.: 503-530-1008; fax: 503-536-6741; e-mail: claudia.jaffe@lumencor.com.