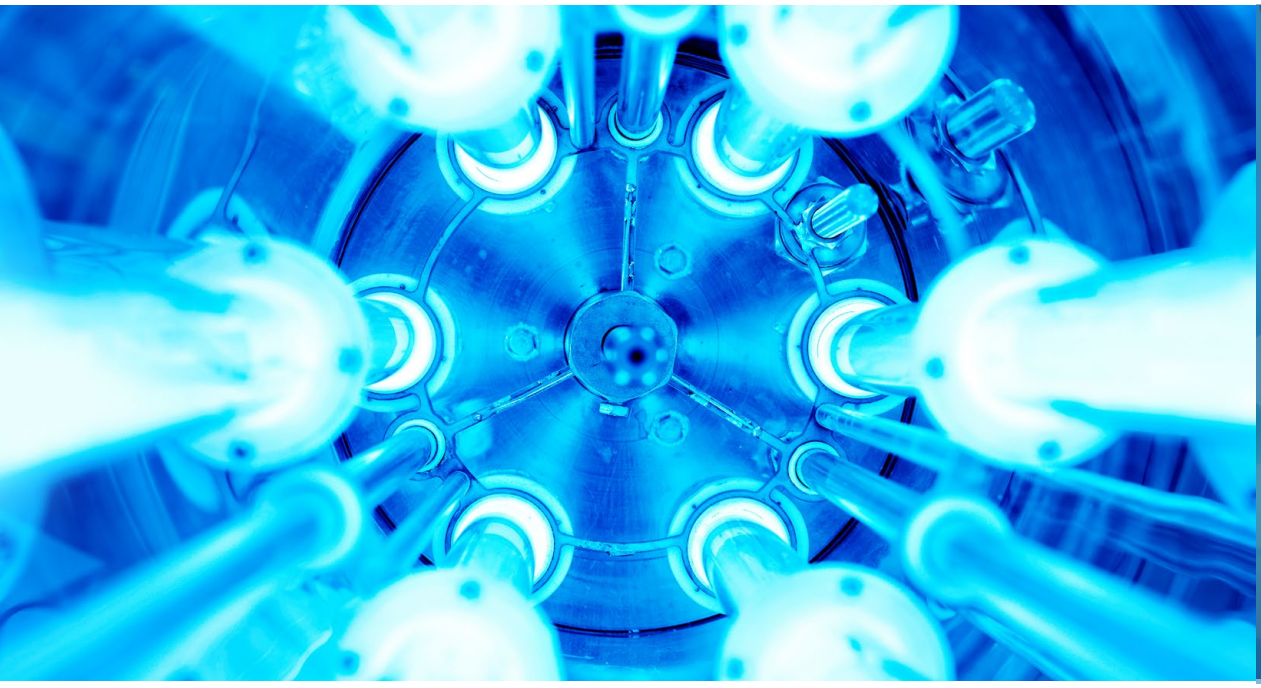
The Evolution of Germicidal UVC Efficacy Testing in the Post-COVID-19 Marketplace

A brief history of ultraviolet germicidal irradiation (UVGI) pre-pandemic



Microbiological testing has always played an important role throughout the lifecycle of various products in the market, spanning from basic electrical components and materials to complex medical devices and more. That market has expanded drastically since the pandemic as consumers are becoming much more conscious of microbiological claims. Prior to the pandemic, most testing that occurred was largely built around rather conventional technologies such as antimicrobial chemicals and mechanical filtration. This was mainly to confirm antimicrobial claims in textiles, or for EPA FIFRA market claim verification of air purifiers that used filtration. Other microbiological testing has always existed in the food industry, pharmaceuticals, and medical. However, new technologies rapidly expanded and gained prominence during the start of the pandemic. One of those rapidly growing technologies was ultraviolet germicidal irradiation (UVGI).

Throughout history, it was well known that microorganisms responded to light in various ways. However, it was not until 1877 that scientists noticed that sunlight-maintained sterility inside test tubes of Pasteur's solution. Shortly thereafter, wavelengths of light were narrowed down to focus on the shorter wavelengths of light within the UV range. Once the general principles of

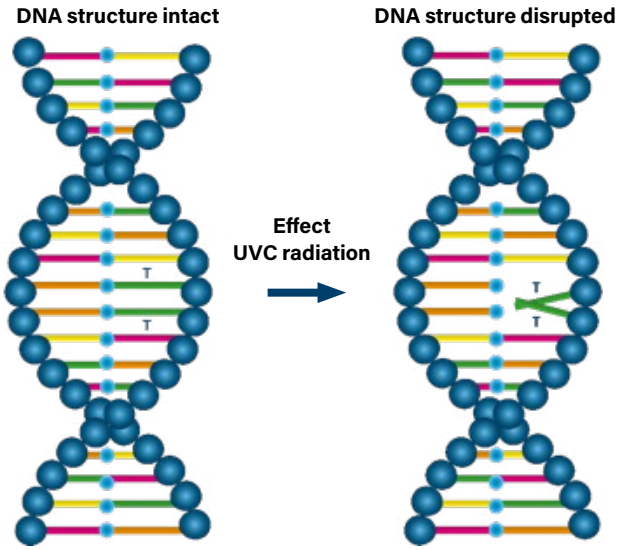
the UVC (100-280) were understood, research began on how to deliver the most precise dose of UVC radiation for the lowest energy cost. Accomplishing this has been one of the main tasks the industry has been working on since its inception. With the advent of LED technology, UVGI has become effectively added to many different applications that were difficult to reach. Prior to LED, more traditional technologies were used, such as Xenon-arc and mercury vapor lamps. Each one of these technologies has both strengths and weaknesses that can be leveraged based on the specific application.

Excluding photobiological measurements, very few specific microbiological tests existed for UVGI technology. The germicidal effect of UVC is determined primarily by the dose of energy delivered to the target substrate, ideally the DNA/RNA of the microorganism. Manufacturers can easily test this at various distances and angles using a radiometer and compare the dose given to the published data on different types of microorganisms to determine the time necessary to eliminate the organisms. However, while this approach makes sense on many levels, from the EPA's perspective, it is necessary to conduct tests using real data against the various microorganisms to provide claims substantiation. This is where microbiological testing and UVGI become critical for validation.

Mechanisms of Germicidal Ultraviolet Light

DNA and RNA contain basic building blocks known as bases. These bases are broken up into two different categories known as purines and pyrimidine. The category of purines contains the bases of adenine and guanine while the pyrimidines contain cytosine, thymine, and uracil (found only in RNA). UVC will directly disrupt the bonds between pyrimidines at the sub 300nm wavelength and cause something known as pyrimidine dimerization, which is when the bases will crosslink intrastrand, as shown in the illustration below.[i] Once this crosslink occurs, the cell's function is disrupted, and sometimes even fatal mutations will occur. Fortunately, for more advanced forms of life, mechanisms exist to repair this damage continuously, preventing cellular disruption simply with exposure to sunlight.

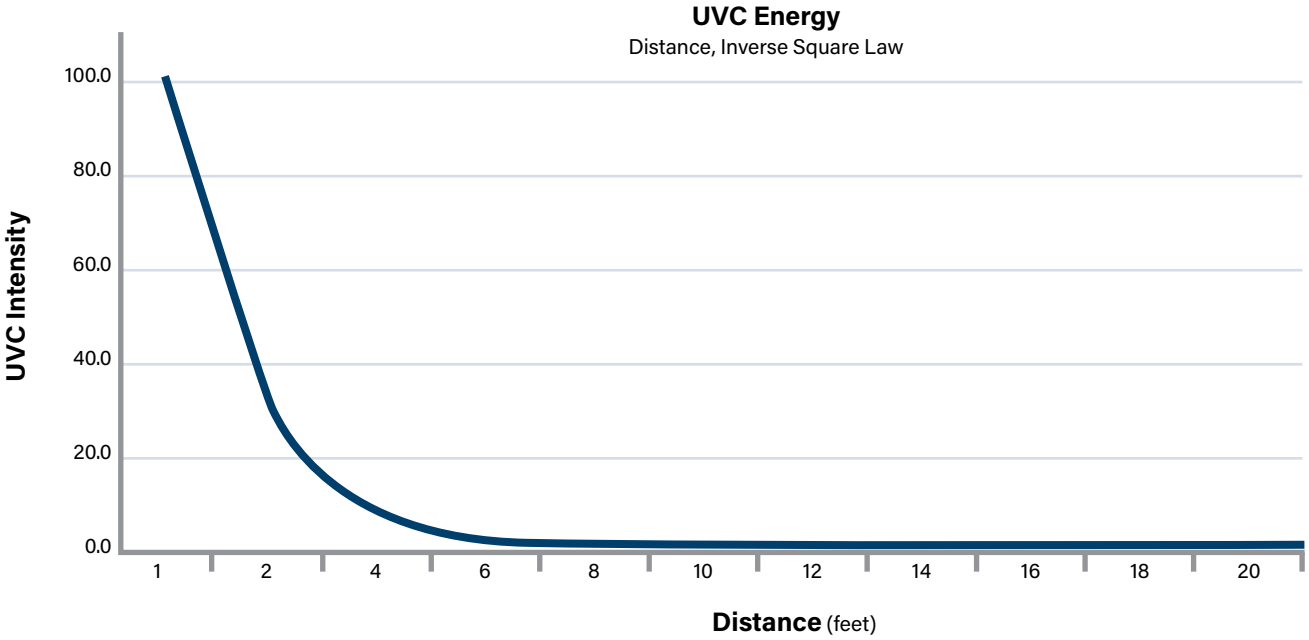
Figure 1: UVC Effect on Microorganism, SGL System



Knowing what is being targeted by the UVGI lighting, the industry has long been working on creative solutions to attack these bonds utilizing UVC light, but also doing it at the lowest energy cost for the largest amount of Micro-Watts per square centimeter or uW/cm^2, which is typically how UVC dosage is represented in literature. This presents some unique challenges to manufacturers, as UVC energy is subject to a particular troublesome rule of physics known as the Inverse Square law. This law specifically states that irradiation is inversely proportional to the square of the distance, as shown in the illustration below from Diversey.[ii] Essentially, this means that the drop off in energy is not linear in fashion. For instance, using a UVC lamp as an example, if we measured the intensity, and it was 10 uW/cm2 and we doubled the distance, you may expect the intensity to be about 5 uW/cm^2 if it followed linear rules. The real value is 2.5 uW/cm^2 – as you doubled the distance, the energy is actually 1/4th of the original intensity. Triple the distance? 3^2 = 1/9th of the energy.

As previously mentioned, to effectively eliminate a microorganism by a certain log value, a specific dosage must be delivered to it. This dosage is dependent on four main factors:

Figure 2: Inverse Square Law, *Diversey*



1. **Distance:** The separation between the source and the target microorganism.
2. **Intensity:** The strength (Radiant Power) of the source emitting the treatment.
3. **Duration:** The length of time that the target is exposed to the treatment.
4. **Wavelength:** The specific wavelength of the treatment source needed to achieve the desired elimination level.

Please note the emphasis on "main" factors, as secondary factors like reflectivity, geometry, and even soiling can significantly impact efficiency. These secondary factors will be discussed later in this paper. Now, understanding these factors will impose certain design and engineering constraints on how the product is used and, of course, what types of claims it can make. One additional consideration, which this whitepaper does not delve into, is safety. When it comes to calculating the dosage necessary to eliminate a particular microorganism, the following equation can be utilized, courtesy of Klaran.[iii]

Figure 3: Dosage Calculation Formula, *Klaran*

$$\text{Dose}\left(\frac{\text{mJ}}{\text{cm}^2}\right) = \text{irradiance}\left(\frac{\text{mW}}{\text{cm}^2}\right) * \text{time (s)}$$

This formula can be adjusted to determine the time necessary at a particular energy and wavelength to achieve a certain dosage. This is often necessary to calculate before sending it to a microbiology testing laboratory to save time and cost, as each timepoint from a laboratory test could equate to thousands of dollars. The excerpt of the table below was taken from Crystal IS but can be verified by the referenced studies.[iv]

The dosage required to achieve a 2-log reduction is provided as 5.4 mJ/cm^2. This is based on mercury vapor lamps, and therefore the manufacturer has made a correction based on the utilization of their LED technology, as depicted in the equation below.[iii]

Table 1: Dosage Calculation Workflow, *Crystal IS*

Bacterium	Lamp Type	UV DOSE (FLUENCE) (MJ/CM²)*							REFERENCE
		1	2	3	4	5	6	7	
Shigella sonnei ATCC9290	N/A	3.2	4.9	6.5	8.2				Chang et al. 1985
Staphylococcus aureus ATCC25923	N/A	3.9	5.4	6.5	10.4				Chang et al. 1985
Streptococcus faecalis ATCC29212	N/A	6.6	8.8	9.9	11.2				Chang et al. 1985

Figure 4: Dosage Calculation Adjustment for LED Technology, Klaran

$$\textbf{time} = \left(\frac{\text{dose (at 265 nm)}}{\text{irradiance}}\right) = \left(\frac{5.2\ \text{mJ/cm}^2}{0.075\ \text{mW/cm}^2}\right) = \textbf{1 min 10 seconds}$$

Achieving a dosage of 5.2 mJ/cm² at a particular watt density of 0.075 mW/cm² (which can be calculated using an intensity map or via source/distance calculation). Once this calculation is complete, the result is 69.333 seconds, or if rounding up for caution's sake, 1 minute and 10 seconds.[iii]

A wealth of dosage data is accessible through standard online literature searches. While the online data primarily pertains to the 254nm wavelength, information for other wavelengths can also be found with more diligent effort. It is crucial to compare data from multiple sources since various experimental variables can influence the reported dose-kill relationship. The Environmental Protection Agency (EPA) has published an extensive meta-analysis, offering a substantial amount of data on the UV dose inactiviation required for various microorganisms. Below are excerpts from this published meta-analysis, illustrating the diverse dosages necessary to inactivate not only different pathogens but also the same pathogen as reported in various studies.[v]

Implementation of Germicidal Ultraviolet Light in Products

Prior to the pandemic, and due to many factors, UVGI was primarily used in the water purification industry. High-intensity UVGI could be kept outside of any direct human exposure behind an enclosure, and the target (the water) could be in near contact with the source, allowing for an immense amount of UVC radiation to bombard the liquid. Not only that, but whereas surface disinfection will utilize only a fraction of a UVC source's energy based on the flat surface geometry, water disinfection can use the full 360-degree arc of a UVC bulb's energy distribution. The figure below shows a basic schematic of a UV unit with a bulb, sourced from Agriculture and Agri-Food Canada.[vi]

Figure 5: Basic Schematic of UV Unit with Bulb, *Agriculture and Agri-Food Canada*

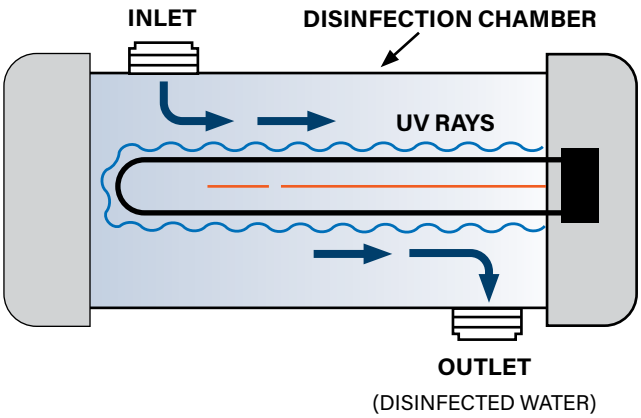


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Table 2: Dosage Variability Across Microorganisms, *EPA*

MICROORGANISM	TYPE	UV DOSE (mJ/cm²) in activation indicated				REFERENCE
		1-log	2-log	3-log	4-log	
Aeromonas hydrophila	Bacteria	1.1	2.6	3.9	5	Wilson et al. 1992
Campylobacter jejuni	Bacteria	1.6	3.4	4	4.6	Wilson et al. 1992
***Escherichia coli* O157:H7**	Bacteria	1.5	2.8	4.1	5.6	Wilson et al. 1992
Legionella pneumophila	Bacteria	3.1	5	6.9	9.4	Wilson et al. 1992
Salmonella anatum	Bacteria	7.5	12	15	-	Tosa and Hirata 1998
Salmonella enteritidis	Bacteria	5	7	9	10	Tosa and Hirata 1998
Salmonella typhi	Bacteria	1.8	4.8	6.4	8.2	Wilson et al. 1992
Salmonella typhimurium	Bacteria	2	3.5	5	9	Tosa and Hirata 1998
Shigella dysenteriae	Bacteria	0.5	1.2	2	3	Wilson et al. 1992
Shigella sonnei	Bacteria	3.2	4.9	6.5	8.2	Chang et al. 1985
Staphlococcus aureus	Bacteria	3.9	5.4	6.5	10.4	Chang et al. 1985
Vibrio cholerae	Bacteria	0.8	1.4	2.2	2.9	Wilson et al. 1992
Yersinia enterocolitica	Bacteria	1.7	2.8	3.7	4.6	Wilson et al. 1992
Adenovirus Type 40 [2]	Virus	30	59	90	120	Meng and Gerba 1996
Adenovirus Type 41 [2]	Virus	22	50	80	-	Meng and Gerba 1996
Coxsackievirus B5	Virus	6.9	14	21	-	Battigelli et al. 1993
Hepatitis A HM175	Virus	5.1	14	22	30	Wilson et al. 1992
Hepatitis A	Virus	5.5	9.8	15	21	Wiedenmann et al. 1993
Hepatitis A HM175	Virus	4.1	8.2	12	16	Battigelli et al. 1993
Poliovirus Type 1	Virus	4.0	8.7	14	21	Meng and Gerba 1996
Poliovirus Type 1	Virus	6	14	23	30	Harris et al. 1987
Poliovirus Type 1	Virus	5.6	11	16	22	Chang et al. 1985
Poliovirus Type 1	Virus	5.7	11	18	13	Wilson et al. 1992
Rotavirus SA11	Virus	7.6	15	23	-	Battigelli et al. 1993
Rotavirus SA11	Virus	7.1	15	25	-	Chang et al. 1985
Rotavirus SA11	Virus	9.1	19	26	36	Wilson et al. 1992
Cryptosporidium parvum [2]	Protozoa	<2	<3	<5	-	Shin et al. 2001
Cryptosporidium parvum [2]	Protozoa	-	<3	<6	-	Clancy et al. 2000
Giardia lamblia [2]	Protozoa	<1	-	-	<2	Linden et al. 2002a
Giardia lamblia [2]	Protozoa	<1	<3	<6	-	Mofidi et al. 2002

[1] Adapted from Wright and Sakamoto 1999

References:

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[viii] United States Environmental Protection Agency (EPA). EPA Regulations About UV Lights that Claim to Kill or Be Effective Against Viruses and Bacteria. (online). https://www.epa.gov/compliance/compliance-advisory-epa-regulations-about-uv-lights-claim-kill-or-be-effective-against

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