

Photochemical Sterilization

Chemical bond dissociation induced by light occurs when a photon's wavelength dependent quantum energy is equal to or greater than the energy of the molecular bond upon which it is incident.

Photochemical damage to a life-critical biological structure can be induced by irradiation with photons having energy levels corresponding to the bond energies of biomolecular chemical bonds. Upon UV photon absorption, excited states and reactive species are created which react to form biologically non-functional reaction products.

The history of photochemical microbial inactivation dates back to the discovery in 1877 by Downes and Blount (1) that ultraviolet light can damage microorganisms. In 1928 F.L. Gates (2) made the formal discovery that

specific monochromatic wavelengths of UV light are bactericidal. The physical mechanism connecting specific wavelengths of light with specific molecular bonds was finally revealed by quantum mechanics, developed by Planck, Einstein, Bohr, Sommerfeld, de Broglie, Heisenberg, Dirac, Pauling and others during the first half of the 20th century. Biochemical research since then has shown that the most effective wavelengths, 250 nm to 280 nm, coincide with the peak absorption spectra of nucleic acids (3). On the basis of this correlation, and the observation that the majority of damage to inactivated microbes is found in their genetic material, the primary mechanism in UV induced microbial inactivation is now known with certainty to



be biomolecular damage to DNA and RNA nucleic acids.

Photochemical sterilization of microorganisms is achieved by high flux UV irradiation in the germicidal wavelength range from 200 nm to 320 nm. Absorbed UV photon energy dissociates C, N, O, and H covalent bonds resulting in irreversible molecular damage to nucleic acids and cell death. Xenon Corporation's *SteriPulse-XL*[™] photochemical sterilization systems generate the most effective form of germicidal UV light known - high flux broad

spectrum pulsed xenon arc radiation - and achieve high assurance USP sterility levels for all known microorganisms.

UV radiation damage to DNA includes formation of cyclobutane pyrimidine dimers (CPDs) and pyrimidine-pyrimidone 6-4 photoproducts (6-

4 PP's) (4, 5, 6, 7), as shown in Figure 1. CPDs are formed by covalent bonding between two same strand adjacent pyrimidines. UV irradiation usually generates thymine dimers in the greatest quantity, cytosine dimers in low quantity, and mixed dimers at an intermediate level (8). In UV irradiated RNA viruses, the nucleotide uracil forms pyrimidine photoproducts. At irradiation flux magnitude and total dose high enough to irreversibly inactivate nucleic acid repair mechanisms, nucleic acid damages result in irreversible mutations, impairment of replication and gene transcription, and eventual death of the organism.

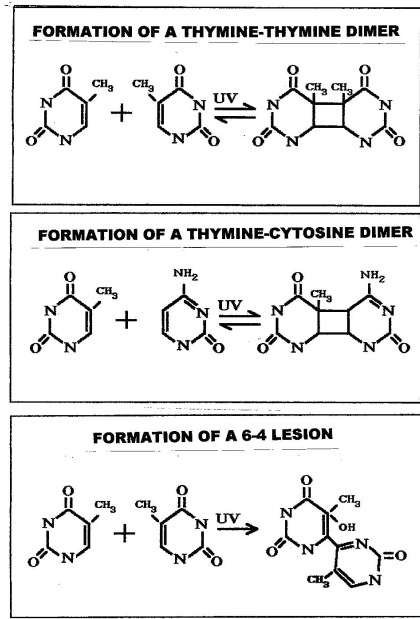


Figure 1 Formation of Dimers and 6-4 Lesions

Conventional low pressure monochromatic and medium pressure polychromatic mercury vapor arc UV lamps are inherently low radiance sources and cannot practically deliver radiant flux magnitudes required for irreversible inactivation of nucleic acid repair mechanisms. Pulsed xenon arc sources are inherently high radiance, high power devices which easily and practically yield high over-kill radiant flux magnitudes sufficient to cause irreversible inactivation of all nucleic acid repair mechanisms and consequent lethal nucleic acid injury.

The inherent advantages of high peak pulsed UV, when compared to mercury vapor arc exposure, have established pulsed UV technology as the UV source of choice for high sterility sterilization systems. Xenon Corporation, the acknowledged leader in pulsed UV technology, is the supplier of choice for high sterility pulsed UV photochemical sterilization systems.

For a more complete discussion on the photochemistry of UV induced damage to nucleic acids, and on repair mechanisms, the reader is referred to the excellent treatment by Blatchley and Peel (9).

Sterilization results on *Bacillus subtilis*, shown in Figure 2, demonstrate the remarkable rapidity and effectiveness with which high flux pulsed UV light eradicates microorganisms. Only three 360 microsecond pulses at 1.2 joules/cm²-sec yield a greater than log 6 kill.

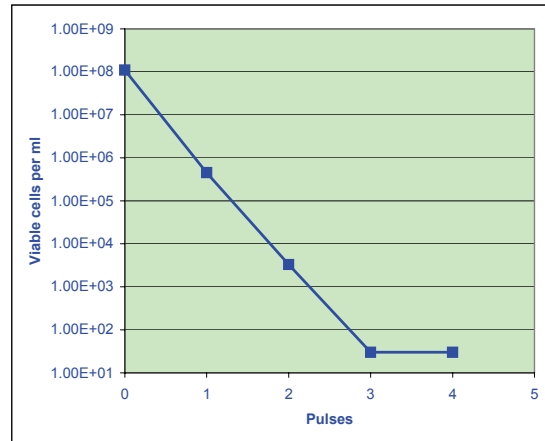


Figure 2 Example of the lethality of Pulsed UV exposure on *Bacillus subtilis*. Determined experimentally by Abraham L. Sonenshein, PhD, Tufts University School of Medicine and Xenon Corporation.

High peak power pulsed UV creates excited states and reactive species in very large concentrations. One joule contains on the order of 10^{18} UV photons. Delivered at high power in microseconds, these photons produce far more biological damage than they would if delivered by a mercury vapor source at low power over longer time. For equal total energy absorbed, bactericidal damage is inversely related to duration of energy – a work-time relation which is consistent with the finite characteristic energy dissipation rates typical of molecular structures in general and nucleic acids and their repair mechanisms in particular, and which underscores the virtue of high peak power pulse UV energy.

References

1. Downes, A., Blount, T. Research on the effect of light upon bacteria and other organisms. *Proc R Soc London* 1877;26:488
2. Gates, F.L. 1928. "On nuclear derivatives and the lethal action of UV light." *Science*, 68, 479-480
3. Davidson, J.N. *The biochemistry of nucleic acids*, 5th ed. London: Methuen, 1965.
4. Setlow, R.B., Cyclobutane-type pyrimidine dimmers in polynucleotides. *Science* 1966;153:379-386
5. Lippke, J.A., Gordon, L.K. Brash DE, et al. Distribution of UV light-induced damage in a defined sequence of human DNA:detection of alkaline-sensitive lesions at pyrimidine nucleoside-cytidine sequences. *Proc Natl Acad Sci USA* 1981;78:3388-3392
6. Friedberg, E.C., Walker, G.C., Siede W. *DNA repair and mutagenesis*. Washington DC: ASM Press, 1995:24-108
7. Yasui, A., McCreedy, S.J. Alternative repair pathways for UV-induced DNA damage. *Bioessays* 1998; 20:291-297
8. Setlow, R.B., Carrier WL. Pyrimidine dimmers in ultraviolet-irradiated DNAs. *J Mol Biol* 1966;17:237-254
9. Blatchley, Ernest R. III and Peel, Margaret M. "Disinfection by Ultraviolet Radiation." In *Disinfection, Sterilization, and Preservation*. Edited by Seymour S. Block, Philadelphia: Lippincott Williams & Wilkins, Fifth Edition, 2001. Page 828
10. Panico, L.R., 1999, "Pulsed UV Curing for Medical Devices", *Medical Device & Diagnostic Industry*, October
11. Panico, L.R., 1997, "Pulsed UV Curing Provides User-friendly Solutions to Tough Problems", *Adhesive Age*, January
12. Blank, E., 1980, "Flash Xenon UV Curing - An alternative System", *J. Radiation Curing*, October
13. Panico, L.R., 1972, "Simulating the Sun with Pulsed Light", 6th Space Simulation Conference, May
14. Marshald, I.S., et al. 1959, "New Flashtubes", *Uspekly Naoteshnoy Fotograf*, 6