Re-use of wastewater: preventing the recovery of pathogens by using medium-pressure UV lamp technology

B.F. Kalisvaart

Berson UV-techniek, P.O. Box 90, 5670 AB Nuenen, The Netherlands
(E-mail: *ben.kalisvaart@bersonuv.com*)

Abstract Ultraviolet (UV) light has become widely accepted as an alternative to chlorination or ozonation for wastewater disinfection. There are now over 2,000 wastewater treatment plants worldwide using either low- or medium-pressure UV technology. Recent studies investigating UV lamp technology, configuration, cleaning requirements and ageing, as well as long-term performance tests, have demonstrated beyond any doubt the effectiveness of UV in inactivating pathogens in wastewater. Research has also shown that, to ensure permanent inactivation and prevent the recovery of microorganisms following exposure to UV, a broad, "polychromatic" spectrum of UV wavelengths is necessary. These wavelengths inflict irreparable damage not only on cellular DNA, but on other molecules, such as enzymes, as well. Only medium-pressure UV lamps produce the necessary broad range of wavelengths; low-pressure lamps emit a single wavelength peak which only affects DNA. Polychromatic medium-pressure UV light is so effective because of the lamp's exceptionally high UV energy output at specific wavelengths across the UV spectrum. It has been shown, for example, that pathogenic *E. coli* O175:H7 was able to repair the damage caused by low-pressure UV, but no repair was detected following exposure to UV from medium-pressure lamps.
Keywords Disinfection; DNA; low-pressure; medium-pressure; microorganism; photoreactivation; reactivation; recovery; repair; re-use; UV lamp; wastewater; wavelength

Water Science and Technology **Vol 50 No 6 pp 337–344** © IWA Publishing 2004

Introduction

It is well known that bacteria and other microorganisms are capable of repairing their DNA following damage by ultraviolet (UV) radiation. Known as "reactivation" or "recovery" it is a natural defence mechanism that evolved over millions of years. Some microorganisms need visible light to repair their DNA (known as "photoreactivation") while others can repair it without light ("dark-repair"). This self-repair ability poses obvious problems when UV disinfection technology is used to treat potable water, swimming pool water, wastewater or other liquids.

There are two main types of UV disinfection lamp technologies currently in use: low-pressure and medium-pressure. Low-pressure UV lamps contain mercury gas at a low pressure (< 10 torr) which, when excited by an electrical charge, emits UV light at 254 nanometres (nm). Medium-pressure lamps contain mercury gas at much higher pressure (approximately 1,000 torr). These lamps produce UV of a higher intensity and over a broader range of wavelengths (200–400 nm) than low-pressure lamps.

Recent research comparing microbial DNA photoreactivation after exposure to UV from low- and medium-pressure lamps has shown that the DNA of pathogenic and non-pathogenic *E. coli* was repaired following low-pressure UV irradiation, but not after exposure to medium-pressure UV. These results are highly significant and could have important implications for wastewater treatment and other liquids.

Over 25 years ago Berson UV-techniek, based in the Netherlands, was the first company to introduce second-generation medium-pressure UV lamps for disinfection purposes. Today, medium pressure UV lamps are used for a wide range of disinfection applications including potable water, wastewater and industrial process water. At the end of the

twentieth century, Berson developed enhanced medium-pressure lamps. These new lamps combine the high UV efficiency of low-pressure lamps with multiple germicidal effects of medium-pressure lamps' broad-wavelength output.

UV disinfection

One of the earliest reports describing the germicidal effects of UV was by Downes and Blount (1877). They described the lethal effects of sunlight on a mixed microbiological population and assigned the cause of these effects to UV radiation. Early interest in the application of UV for disinfection was originally centred on potable water. Today, however, many different liquids are disinfected by UV light, including primary, secondary and tertiary filtered wastewater.

UV light has proved to be a very "clean" and effective alternative to those disinfection methods which use chemical agents such as chlorine, chlorine dioxide or ozone. Unlike these methods, UV does not produce any disinfection-by-products (DBPs).

More than 2,000 UV installations are now in operation throughout the world using both low- and medium-pressure UV lamp technology. All kinds of wastewater (primary, secondary and tertiary treated) have been successfully disinfected with medium-pressure UV lamp technology in the last decades. Laboratory and full-scale studies investigating the effects of UV lamp technology, configuration, cleaning requirements, ageing and long-term performance have demonstrated the effectiveness of UV in inactivating pathogens in wastewater.

Emission spectrum of UV lamps

As both the UV emission spectrum and UV intensity play an important role in killing microorganisms, medium-pressure lamps were designed to combine these elements. The improved performance of this type of lamp is achieved by: a broad emission spectrum in the UV area, ultra-high UV intensity, and improved UV efficiency, all of which cause multiple germicidal effects. The emission spectra and intensities of low and medium-pressure UV lamps are significantly lower than those of medium-pressure lamps.

Electromagnetic spectrum

In the electromagnetic spectrum, the wavelengths between 100 and 400 nm are known as the ultraviolet (UV) region. This region can be roughly divided into three areas (Jagger, 1967):

- extreme, or vacuum, UV : 100–190 nm
- far-UV (UV-C and UV-B) : 190–300 nm
- near-UV (UV-A) : 300–400 nm

Because water and air absorb all wavelengths below 190 nm (extreme or vacuum UV), only the wavelengths between 190 and 380 nm (far-UV and near-UV) can be used for biological effect (Harm, 1980). The International Commission of Light (CIE), subdivides the UV region into four areas: vacuum UV (100–200 nm), UV-C (200–280 nm), UV-B (280–315 nm) and UV-A (315–400 nm).

Absorption of UV light

UV lamps emit light in the UV region at the following wavelengths:

- monochromatic low-pressure UV lamps : 254 nm
- polychromatic medium-pressure UV lamps : 185–400 nm

Photons of light are produced at each specific wavelength. Each photon has its own energy content, which depends upon the wavelength. When the photon is absorbed, for example by a molecule within a microorganism, electrons in the atoms or molecules making up the material are excited.

The velocity of the photons is equal to the velocity of visible light (3×10^8 m/s), while the time needed for their absorption by atoms or molecules is about 10^{-15} seconds. The absorption of specific wavelengths can be shown in an "absorption line spectrum".

The larger and more complex the molecule (for example DNA or protein), the wider the range of wavelengths absorbed and therefore the wider the absorption line spectrum. Molecules – like atoms – are held together by bonds in the form of electrons shared between them. When the energy absorbed from the photons reaches a threshold known as ionisation energy (E_{ion}) , a bond can be broken, splitting the molecule ("reactant") into two or more parts ("products"). This photochemical reaction is called dissociation. Obviously only the photons or radiation energy absorbed by the material can be photochemically effective (Draper–Grotthus principle).

Absorption of UV light by DNA

Harm (1980) considers that for UV radiation to achieve biological effects, the wavelength range 190–380 nm (far-UV and near-UV) is essential. The majority of biological effects, especially in very small microorganisms, are due in the first place to photochemical reactions in the DNA. According to Von Sonntag (1986) the UV absorption curve shows maximum absorption at 200 nm. There is also an absorption peak at around 260–265 nm. Maximum absorption therefore does not occur at 254 nm, the wavelength often assumed to be the most effective for killing microorganisms.

In DNA, the "backbone" molecules of sugar (ribose) and phosphate do not absorb significantly above 210 nm. The absorption by DNA and RNA at these wavelengths is due instead to absorption by the nucleotide bases: adenine (A), guanine (G), cytosine (C), thymine (T) and (in the case of RNA) uracil (U). The absorption spectra of these nucleotide bases are found in the UV-C and UV-B regions. The absorption of photons by the nucleotide bases results in the formation of photo-products, the most common of which are thymine dimers, formed when two adjacent thymine bases become covalently joined by cyclobutane (Adams, 1986). When DNA is damaged in this way it cannot replicate, so the bacterial cell is unable to multiply and is effectively dead (Lehninger, 1976).

Absorption of UV light by proteins and enzymes

In addition to DNA and RNA, photochemical reactions in proteins, enzymes and other molecules are also important, particularly in the case of larger microorganisms such as fungi, protozoa and algae, which have dimensions of tens or hundreds of microns. UV may be unable to penetrate far beneath the surfaces of these organisms, leaving the critical component – the DNA – scarcely affected.

Jagger (1967) suggests that it is probably no accident that, precisely at the point where solar radiation falls off below 300 nm, proteins and nucleic acids (DNA/RNA) – molecules which are of prime importance to life – begin to absorb and be damaged by UV radiation.

The absorption spectra of proteins show, in general, a maximum peak at around 280 nm. The peptide bond (–CONH–) in proteins displays some double bond characteristics and is relatively weak at absorbing UV, the only significant absorption occurring below 240 nm. However, as there is a peptide bond for every amino acid residue in a protein, UV absorption below 240 nm is nonetheless significant.

Bensel (1977) claims that absorption of UV light by the amino acid cysteine makes proteins and enzymes unstable. If the dissociation energy for the disulfide (S-S) bond between cysteine molecules is reached, dissociation of the tertiary structure of the amino acid takes place, resulting in denaturation of the molecule and a loss of biological activity. If, for example, denaturation of the enzyme polymerase takes place, the microorganism loses its ability to multiply; while in the case of the enzyme photolyase, the microorganism loses its

ability to repair UV damage. Because of the high concentration of proteins in micro-organisms (about 50% of the dry weight), absorption of UV light can influence their role in nucleic acid synthesis and chromosome structure (Ingraham, 1994).

Absorption of UV light by other molecules

In addition to DNA, proteins and enzymes, other molecules with unsaturated bonds may be sensitive to the deactivation effects of UV radiation. Important examples include coenzymes, hormones and electron carriers.

Molecules absorbing far-UV (below 300 nm)

Jagger (1967) claims that, generally, the most important UV absorbers are those with conjugated bonds (alternating single and double bonds). Structures containing conjugated rings are usually good absorbers of far-UV light (below 300 nm). Absorption of far-UV by aromatic acids may result in decarboxylation, deamination or breaking of ring structure.

Some far-UV absorbing molecules are:

- six-membered carbon rings – benzene, toluene, phenol
- rings containing nitrogen – pyridine, imidazole, pyrimidine, cytosine, thymine, uracil
- double rings – naphthalene, purine, adenine, guanine
- triple rings – anthracene, riboflavin
- quadruple rings – steroids, porphyrins
- amino acids – tryptophan, tyrosine, phenylalanine, cystine, cysteine
- other molecules – nicotinamide adenine dinucleotide (NAD)

Molecules absorbing near-UV (above 300 nm)

Molecules absorbing UV above 300 nm include:

- triple rings – riboflavin
- four rings – porphyrins, steroids
- long-chain conjugated molecules – carotenoids
- other molecules – NADH2 (reduced form of NAD), isoprenoid quinones, vitamin K, pterins, vitamin A, flavins, cytochrome.

Germicidal effects of visible light

The ability of wavelengths in the solar spectrum above 300 nm to kill small bacteria (< 10 microns) has been known for a long time (Ward, 1893). However, the affected biological molecules, or chromophores, have yet to be identified. Since proteins and nucleic acids show little or no absorption above 340 nm there must be other chromophores with sufficient absorbency to result in the death of small microorganisms.

In 1952 it was discovered that wavelengths above 300 nm and in the adjacent visible spectrum destroy the capacity of microorganisms to multiply. Radiation between 350–490 nm has been shown (Bruce, 1958), for example, to cause a leakage of ions.

It has often been assumed that the killing of microorganisms above 300 nm occurs because the damage caused by these wavelengths is less effectively repaired than at 254 nm (UV-C). This suggests that, in addition to the formation of pyrimidine dimers, exposure to sunlight can result in the sort of poorly repairable – or even non-repairable – lethal damage which is rarely caused by low-pressure lamps producing UV at 254 nm. Lethal damage of this sort, however, is well within the capacity of third generation, medium-pressure lamps.

It has been shown experimentally that, by filtering out the shorter wavelengths below 360 nm, non-repairable, lethal damage can be inflicted. The germicidal effects of solar radiation are almost entirely due to the formation of oxygen radicals in the cytoplasm

(Tortora, 1995). Experiments with solar light indicate that effects on molecules other than DNA and proteins can also cause lethal damage to microbiological cells.

Wavelengths other than UV-C may become even more important in killing larger, more UV-resistant microorganisms (e.g. *Cryptosporidium parvum* oocysts), as large cells tend to be less transparent to UV-C. The penetration of larger cells by wavelengths below 295 nm is a major problem which increases the importance of other wavelengths. It is assumed that absorption by organic molecules in the outer cell wall is a major contributor to the killing of larger microorganisms.

Oguma (2002) observed that wavelengths between 300 and 580 nm play an important role in reducing the subsequent recovery of microbial colony-forming ability by inducing damage other than that to the pyrimidine dimers in the DNA. Therefore, it was found that light at a broad range of wavelengths effectively reduced the subsequent photoreactivation, which is an advantage that medium-pressure lamps have over conventional low-pressure UV lamps.

Recovery from UV damage

The need to recover from or repair UV damage is common to all organisms. Known as reactivation, the process can take place in both dark and light conditions and is consequently described as either dark repair or photoreactivation (Schlegel, 1992). The method of reactivation varies significantly according to the level of biological organisation and the kind of UV damage inflicted.

The repair mechanism (EPA, 1986) in microorganisms is not universal and there is no clearly defined set of characteristics to determine which species have the ability to repair themselves and which do not. If wastewater is re-used, it is especially important that recovery as a result of exposure to visible (sun) light (photoreactivation) does not occur.

Organisms shown to be capable of photoreactivation include: *Streptomyces* spp., pathogenic and non-pathogenic *Escherichia coli, Saccharomyces* spp., *Aerobacter* spp., *Micrococcus* spp., *Erwinia* spp., *Proteus* spp., *Penicillium* spp., *Neurospora* spp., *Enterobacter cloacae, Citrobacter freundii, Enterocolitica faecium, Klebsiella pneumoniae, Mycobacterium smegmatis, Pseudomonas aeruginosa, Salmonella typhi, Salmonella typhimurium, Serratia marcescens, Vibrio cholerae, Yersinia enterocolitica.*

In the microbial cell the most vulnerable component is the genetic material contained in DNA and RNA. This is due not only to its uniqueness, but also because of the molecule's complex structure and huge size. It is hardly surprising, therefore, that all known types of molecular repair processes have evolved to act upon the macromolecular nucleic acids, particularly the DNA. The effects of monochromatic UV light on the DNA molecule and the enzymatic repair processes are well described in the literature (Ingraham, 1994). Photoreactivation (EPA, 1986) is a phenomenon which can influence the performance and design of UV treatment systems – to prevent enzymatic repair, UV irradiation must damage a wide variety of molecules, including the DNA.

Damage caused by low-pressure UV lamps, which produce monochromatic 254 nm UV light, can be repaired relatively easily using active enzymes like photolyase (Ingraham, 1995). Oguma (2002) reported that wavelengths between 220 and 300 nm reduced the subsequent photorepair of ESS (endonuclease sensitivity site), by causing a disorder in endogenous photolyase.

However, it has been demonstrated that UV damage caused by high intensity, polychromatic medium-pressure UV lamps, cannot be repaired (Oguma, 2001, 2002; Zimmer, 2002, 2003). The repair processes in molecules other than DNA have not been identified.

Differences between low and high UV intensities

Microorganisms have the capacity to recover using active enzymes such as photolyase,

endonuclease, polymerase and ligase (Dressler, 1986). For the photo-reactivation process to take place, exposure times to light between the wavelengths 310 to 480 nm varying from a few minutes to several hours are needed, depending on the organism. It is likely that the repair systems in microorganisms are so efficient that, below the lethal UV dose, the potentially lethal effects of virtually all UV damage is avoided. As already mentioned, all known types of molecular repair process act upon the DNA because it is the most important molecule within microorganisms. Over 90% of the pyrimidine lesions (thymine dimers) caused by 254 nm UV irradiation can be repaired by active enzymes.

In studies of survival kinetics using biological materials, UV intensity – that is, the amount of UV energy absorbed – is often considered to be less important for disinfection than the UV dose – the total number of photons absorbed. The low intensity of low-pressure lamps should therefore produce the same UV dose as the high intensity of polychromatic medium-pressure lamps, as long as the exposure time is increased by the appropriate factor. This is correct from a photochemical point of view. The extent and nature of the damage caused should depend only on the UV dose, which is a product of UV intensity and exposure time as stated by the Bunsen–Roscoe reciprocity law:

$$\text{UV dose (mWs/cm}^2\text{)} = \text{UV intensity (mW/cm}^2\text{)} \times \text{time (s)}$$

However, since the effectiveness of UV disinfection depends not only on photochemical reactions but also on biological processes (namely repair), it has been concluded (Harm, 1980) that the Bunsen–Roscoe law may not actually apply here. UV intensity seems to play a very important role. Harm (1980) shows that the most lethal effect can be achieved by combining high intensities with short exposure times. He concludes that, at equal UV doses, significantly greater reductions in *Escherichia coli* are achieved using short-term, high intensity UV irradiation ("single exposure") rather than long-term, low intensity irradiation ("sector irradiation").

Wayne (1999) surmises that recent studies of photochemical reactions involve multi-photon processes, in which a single particle absorbs more than one photon. Such absorption only occurs at sufficiently high intensities.

Sommer *et al*. (1998) confirmed that, using *E. coli* strains (ATCC 25992, ATCC 11229 and isolate from sewage), high intensities during short times are more effective in the inactivation than low intensities during longer times at the same UV doses.

Practical experiments

Practical experiments carried out by Dutch KIWA-Institute (Kruithof, 1992) show that at equal UV doses, the indicator organism HPC 22°C is reduced more effectively using medium-pressure rather than low-pressure lamps. This increased effectiveness means that equal reduction rates can be achieved with a reduction in UV dose of about 35% when medium-pressure lamps are used. This is due to the "multiple" effects on microorganisms caused by the very high UV intensity and broad emission spectrum of the medium-pressure lamps. The higher energy consumption of these polychromatic lamps is offset by their increased efficiency in killing microorganisms.

Cryptosporidium parvum

Linden (2001) describes the results of exposing *Cryptosporidium parvum* oocysts to distinct wavelengths of UV from a medium pressure lamp across the germicidal wavelengths 210 to 295 nm. The highest reduction rates were reached at a wavelength of 271 nm, which was approximately 15% more effective than at 254 nm. Even 263 nm was shown to be more effective at inactivating *Cryptosporidium parvum* oocysts than 254 nm. Oguma (2001) investigated both photoreactivation and dark-repair of *Cryptosporidium parvum* with

endonuclease sensitive sites (ESS) assay, to determine UV-induced pyrimidine dimers in the genomic DNA. Exposure of *C. parvum* to fluorescent light after UV inactivation with low-pressure lamps (254 nm) showed continuous repair of UV-induced pyrimidine dimers in the DNA.

***Bacillus subtilis* spores**

Waites (1988) reported that the greatest kill of *Bacillus subtilis* is achieved using UV radiation around 270 nm. This suggests that the UV is not acting directly on the DNA of *Bacillus subtilis* spores, but rather on the dipicolinic acid (DPA), whose absorption peak is close to 270 nm. The absorption spectrum of DPA within spores of *Bacillus subtilis* shows that wavelengths at 270 nm are about 40% more effective compared to those at 254 nm.

Non-pathogenic *Escherichia coli*

Oguma (2002) investigated recovery of non-pathogenic *E. coli* using low-pressure and medium-pressure UV lamps. The medium-pressure lamp used in the experiments filtered out wavelengths below 300 nm so as to investigate the effect of shorter UV wavelengths. In photoreactivation experiments, more than 80% of the pyrimidine dimers were induced by the low-pressure and medium-pressure filtered UV. Photorepair treatment of DNA *in vitro* suggested that among the medium-pressure UV emissions, the wavelengths below 300 nm reduced the subsequent photorepair of endonuclease sensitive sites, possibly by causing a disorder in endogenous photolyase, an enzyme specific for photoreactivation. On the other hand, medium-pressure UV emissions of wavelengths between 300 and 580 nm were observed to play an important role in reducing the subsequent recovery of colony-forming ability by inducing damage other than that to pyrimidine dimers. Therefore, it was found that light at a broad range of wavelengths effectively reduced subsequent photoreactivation, which could be the advantage that medium-pressure UV has over low-pressure UV.

Pathogenic *Escherichia coli* O157:H7

Results of collimated beam studies (Zimmer, 2002, 2003) show that pathogenic *E. coli* O157:H7 underwent photorepair following exposure to low-pressure UV lamps, but no repair was detectable following exposure to medium-pressure lamps. The studies clearly indicate differences in repair potential under laboratory conditions between low-pressure and medium-pressure UV lamps. Oguma (2001) investigated photoreactivation in *E. coli* and *Cryptosporidium parvum* using low-pressure and medium-pressure lamps. UV-induced pyrimidine dimers in DNA were continuously repaired using low-pressure lamps, while none of these dimers were repaired when using medium-pressure UV lamps.

Conclusions

All types of wastewater, from low-transmission primary treated to high quality reverse-osmosis filtered wastewater, can be effectively disinfected with medium-pressure UV, with laboratory and full-scale investigations proving the effectiveness of UV technology compared with chemical treatment. Next generation medium-pressure UV lamps combine a broad emission spectrum with ultra-high UV intensity, which have been shown to cause multi-photon photochemical damage inside microbiological cells. In addition to far-UV (<300 nm), both near-UV (>300 nm) and visible wavelengths may also be effective in killing microorganisms, particularly larger, multi-cellular ones.

Most microorganisms can repair UV-damaged DNA with enzymes in light or dark conditions. Repair processes of UV-damaged molecules other than DNA, however, have yet to be discovered. The Bunsen–Roscoe reciprocity law may not be applicable for the destruction of microorganisms, as biological processes also play an important role.

Wavelengths of polychromatic medium-pressure UV lamps are more effective at killing larger and more UV-resistant microorganisms than low-pressure monochromatic UV lamps. For the inactivation of *Cryptosporidium parvum* oocysts and *Bacillus subtilis* spores, wavelengths in the region of 270 nm appear to be more effective than those at 254 nm.

Differences in photoreactivation were observed between low-pressure and medium-pressure lamps: UV damage caused by low-pressure UV lamps underwent photorepair, while no repair was detectable following exposure to medium-pressure UV lamps.

It was found that light at a broad range of wavelengths, up to 580 nm, effectively reduced subsequent photoreactivation, which is an advantage that medium-pressure lamps have over conventional low-pressure UV lamps.

If wastewater is to be re-used, preventing microbial recovery in light (photoreactivation) is vitally important. It is in instances such as this that medium-pressure, rather than low-pressure UV lamp technology offers the best option.

References

Adams, R.L. (1986). *The Biochemistry of the Nucleic Acids*, Chapman & Hall, 10th ed.

Bensel, J. (1977). *Ultraviolette Strahlen*, Walter de Gruyter, Berlin.

Bruce, A.K. (1958). Response of potassium retentivity and survival of yeast to far-ultraviolet, near-ultraviolet and visible, and X-radiation. *J. Gen. Physiol.*, **41**, 693–702.

Clancy, J.L. *et al.* (1999). *Evaluation of Inactivation of Cryptosporidium parvum Oocysts in Recreational Water by the Aquionics UVP 61 System.*

Downes, A. and Blount, T.P. (1877). Researches on the effect of light upon bacteria and other organisms. *Proc. Royal Soc. London*, **26**, 488–500.

Dressler, D. (1986). *Discovering Enzymes*, Scientific American Library, 1st ed.

EPA (1986). *Design Manual Municipal Wastewater Disinfection*, EPA/625/1–86/021.

EPA (2003). *Environmental Verification Report* UV disinfection for Re-use applications, Aquionics Inc. bersonInLine® 4250 UV System, September 2003.

Harm, W. (1980). *Biological Effects of Ultraviolet Radiation*, Cambridge Press, 1st ed.

Ingraham, J.L. (1994). *Introduction to Microbiology*. Wadsworth Publishing Company.

Jagger, J. (1967). *Introduction to Research in Ultraviolet Photobiology*, Prentice-Hall, Englewood Cliffs, 1st ed.

Kruithof, J.C. *et al.* (1992). Practical experiences with UV disinfection in the Netherlands. *J Water SRT-Aqua*, **41**(2), 88–94.

Lehninger, A.L. (1976). *Biochemistry*. Worth Publishers Inc., 2nd ed.

Linden, K.G. (2001). Comparative effectiveness of UV wavelengths for the inactivation of *Cryptosporidium parvum* oocysts in water. *Water Science and Technology*, **43**(12), 171–174.

McCarty, D.L. (2003). Comparing microbial repair mechanisms with different UV lamps. *WCP*, July 2003, pp. 62–64.

Meltzer, T.H. (1993). *High-purity water preparation*. Tall Oaks Publishing Inc., 1st ed.

Oguma, K. *et al.* (2001). Determination of pyrimidine dimers in *Escherichia coli* and *Cryptosporidium parvum* during UV light inactivation, photoreactivation and dark repair. Department of Urban Engineering, University of Tokyo. *Applied and Environmental Microbiology*, **67**(10), 4630–4637.

Oguma, K. *et al.* (2002). Photoreactivation of *Escherichia coli* after Low- or Medium-Pressure UV disinfection determined by an endonuclease sensitivity site assay. Department of Urban Engineering, University of Tokyo. *Applied and Environmental Microbiology*, **68**(12), 6029–6035.

Schlegel, H.G. (1992). *General Microbiology*. Cambridge Univ. Press, 7th ed.

Tortora, G. (1995). *Microbiology, An Introduction*, The Benjamin/Cummings Publishing Company, 5th ed.

Sommer, R. *et al.* (1998). Time dose reciprocity in UV disinfection of water. *Water Science and Technology*, **38**(12), 145–150.

Von Sonntag, C. (1986). Disinfection by free radicals and UV-radiation. *Water Supply*, **4**, 11–18.

Waites, W.M. *et al.* (1988). The destruction of spores of *Bacillus subtilis* by the combined effects of hydrogen peroxide and ultraviolet light. *Applied Microbiology*, **7**, 139–140.

Ward, H.M. (1893). Further experiments on the action of light on *Bacillus anthracis*. *Proc. Royal Society London*, **53**, 23–44.

Wayne, C.E. and Wayne, R.P. (1999). *Photochemistry*, Oxford University Press, 2nd ed.

Zimmer J.L. and Slawson, R.M. (2002). Potential repair of *Escherichia coli* DNA following exposure to UV radiation from both medium- and low-pressure UV sources used in drinking water treatment. *Applied and Environmental Microbiology*, **68**(7), 3293–3299.

Zimmer, J.L. (2003). Repair potential of selected microorganisms following UV irradiation used in drinking water treatment. *IOW Proceedings "Wasser Berlin 2003"*, pp. 113–130, 2002.