

Dissolved Ozone in Pharmaceutical Water Systems

Why Use and Measure Dissolved Ozone

Nissan Cohen (IN USA, Industry Marketing Manager)

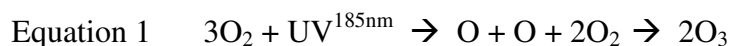
In collaboration with RP Bruno and Associates

Introduction

Charles W. Eliot was quoted as saying that “all business proceeds on beliefs or judgments of probabilities; and not on certainties.” This is also largely true of what we call science today. The analysis of dissolved ozone in USP or general pharmaceutical waters is no exception. There are risks and rewards for using the ambient sanitizer. This paper will seek to establish a fundamental understanding of dissolved ozone and its use in a pharmaceutical water plant.

What is dissolved ozone?

Ozone is a naturally occurring triatomic form of oxygen (O₃) and exists in the gas form in nature. Familiar sources of ozone are lightning in the atmosphere, the sun’s UV in the upper stratosphere creating the infamous “ozone layer”, and copy machines or laser printers. Ozone forms when oxygen comes in contact with ultraviolet (UV) energy wavelength of 185 nm. The UV energy splits the oxygen molecule which then reattaches to another oxygen molecule (see equation 1).



The resulting unstable ozone gas molecule wants to revert back to the stable diatomic oxygen molecule (O₂). In order to do this, it must react with another compound or transfer energy through another source. This makes ozone an oxidizer. In fact, it is one of the strongest oxidizers known to man (see below table). Dissolving this gas into water makes for a very potent antimicrobial solution, which can then be used as a sanitizing agent. Ozone is different than most sanitizers because it is a gas and remains a gas during the sanitization process. It does not metamorph into an ionic form like chlorine and therefore is much harder to stabilize in water.

Oxidant	Oxidation Potential eV
Fluorine (F ₂)	3.0
Hydroxyl radical (OH [·])	2.8
Ozone (O ₃)	2.1
Hydrogen peroxide (H ₂ O ₂)	1.8
Potassium permanganate (KMnO ₄)	1.7
Chlorine dioxide (ClO ₂)	1.5
Chlorine (Cl ₂)	1.4

Why use dissolved Ozone?

There are many types of systemic sanitizers: heat (>65°C), chemicals (acids/bases), oxidizers (ozone, chlorines, peroxides). Each has its peculiar advantages and risks. Heat

has been proven over the years to produce high quality, low microbial growth water, yet it is expensive to maintain and difficult to work around. Chemical sanitizers can be effective for removing biofilms, but must be rinsed out with excessive amounts of high quality water and involve hazardous chemical handling.

Dissolved ozone has the advantage of being able to reach into every part of the water system and then be easily removed. There is no “handling” required of ozone as it is generated from air or compressed gas and automatically injected. The ability to operate at room temperature obviates the need for expensive mechanically complex heating systems, and heat tracing on pipes.

The dissolved ozone mechanism is different than dissolved chlorine, the world’s most popular potable water sanitizing agent. Ozone attacks (oxidizes) all organic (carbon-carbon) bonds which include the cell walls and external structures of bacteria¹, spores and cysts. Since most biological structures are organic ozone is an equal opportunity biocide. If properly dosed dissolved ozone can cause complete destruction of the biological entity.

Ozone is also extremely fast at eliminating microbiological activity in the water at relatively low doses. One source has shown that 0.1 mg/L of ozone will destroy 60,000 colony-forming units (cfu) e.coli in one minute; whereas the same dose of chlorine will take up to 400 hours². As one USP water user and system designer relates; “zero counts over three years are hard to argue with”. This process engineer specifies water systems for his company and highly recommends ozone.

Comparatively, chlorine is an oxidizer that works through the mechanism of diffusion into the cell. It attaches to and denatures the protein structures that comprise the enzymes of a cell. This ultimately inactivates the organism inhibiting reproduction and proper functioning. Chlorine destroys from the inside out. The diffusion required to enter the cell is a delay that the ozone mechanism does not endure³.

Some of the additional benefits of ozone:

- adds no residual chlorine compounds;
- is easily removed by exposing to UV light (wavelength 254 nm) or by degasification;
- is an antistatic agent loosening particulates from vessel walls;
- can oxidize inorganic materials such as nitrites, sulfides, etc.⁴;
- acts as a clarifier actually removing color⁵;
- reduces THM, TOC⁶, endotoxin⁷ and endocrine disruptor⁸ levels;
- deactivates Cryptosporidium and Giardia cysts⁹.

The strength of ozone and the associated advantages lead to the conclusion that ozone use for sanitization can offer increased product quality and lower the risk of water-borne contraindicative components.

Why measure dissolved ozone?

Microbiological analysis is a requirement for all grades of USP and EP water and there are clearly defined limits for each type^{10,11}. Microbiological testing is a time consuming process. It can take from six hours to two weeks to perform impinging product acceptability. Since real time microbiological monitors do not currently exist, one solution is to correlate another more easily adaptable real-time measurement to the disinfection required. Ozone disinfection can be defined by the term “contact time” (CT). CT is the residual ozone quantity in a storage tank or loop multiplied by the time the ozone is in contact with the water. Therefore, a direct dissolved measurement of ozone can be validated to disinfection efficacy. In ozone sanitized system, the measurement of dissolved ozone by a real time instrument is a risk mitigation tool as the values can be correlated to assured ingredient quality.

Equal in concern for the user is the verification that ozone has been removed from the water. Ozone is strong. It must be removed before it comes in contact with other excipients or active ingredients. Else, there is a risk of product alteration or stability issues. It is best to choose a measurement technology based on its ability to measure at the very lowest level. Most users can adjust the residual confidence limits for just about any technology used to measure dissolved ozone. For instance, an analysis with a +/- 10 ppb accuracy would mean that the user who needed a 50 ppb residual could set its target value at 60 ppb and still have some room for variation. This is not so with post destruct measurement. For example, a value of 10 ppb with this same technology +/- 10 ppb could be zero or it could be 20 ppb. These are vastly different values. The assurance of removal is a concern because of ozone's speed and power. Therefore in some systems redundant destruct mechanisms and measurement systems are in place to ensure destruction before product is released.

How is dissolved ozone made?

Ozone is made onsite at all facilities. In most US based pharmaceutical companies, ozone is generated by passing air or oxygen between two electrically charged plates commonly known as the Corona Discharge method (also known as dielectric barrier discharge) which simply means generating a high voltage electrical field and passing air through it. Passing air through the corona converts approximately 2% of the oxygen into ozone. The gas must then be dissolved in the water. Usually via a venturi injector¹² system, the ozone is administered into a moving stream. Sometimes injection is administered by bubbling into a filled water tank or reservoir.

In European pharmaceutical companies the preference is to create dissolved ozone electrolytically from the water itself. In this case, a strong potential is applied to a set of electrodes and either the water is split or dissolved oxygen is converted and ozone made. The ozone is created as a dissolved species. There are of course advantages and disadvantages to each method. The traditional difference seems to be the corona discharge method produces a higher concentration than the electrolytic method. For applications requiring higher levels of dissolved ozone, corona discharge would be generation method of choice.

What are the operating costs?

The main operating expenses are electricity and equipment maintenance. In some cases, liquid oxygen (LOX) (in only very large water systems when generation of over 200 grams to kilos of ozone are needed per day) is used with the corona discharge method to gain a higher ozone weight. Electricity is by far the largest and most predictable expense. Wasted or unnecessary ozone production increases operating costs and also reduces system component longevity. Excess levels of ozone will rapidly degrade non-resistant seal materials. Therefore monitoring dissolved ozone levels can be used as a feed back control to adjust the generator output. This is cost effective and important when designing a system with variable water usage.

A quick review of potentially negative impacts:

- Ozone is electrically expensive to produce. It is therefore important to consider a feedback control mechanism involving the dissolved monitor.
- Dissolved ozone is aggressive on materials and has been known to destroy piping, seals, and components that have been poorly chosen. A small materials selection chart is located in the appendix, aiding the user in deciding on common materials of manufacture.
- Because dissolved ozone desires to be in its gaseous state and can be a human health hazard¹³ it is advisable to have ambient monitors with alarms in the vicinity of the water system and ozone generator.

Are there regulatory concerns?

The FDA's early reluctance to endorse ozone usage is changing as ozone has been shown to reduce the risk of microbial contamination. This is evidenced by the FDA approval of dissolved ozone as food contact disinfectant in 2001 (21CFR part 173). It is also consistent with the recent 21st Century Initiatives including the move toward risk-based decisions¹⁴ and assuring production quality versus testing for it¹⁵.

For users making USP water, USP 29 states Water For Injection (WFI) will contain "no added substance"¹⁶. The FDA, depending on the auditor, may consider ozone to be an "added substance". Therefore, it must be shown to be removed.

For Purified Water (PW) the manufacturer is required to label ozone as an added ingredient or prove that the ozone has been removed from the water. USP users should remove ozone before formulation of the final product. The user should then verify that ozone has been removed.

Some exceptions would be sterile bottled waters, dialysate buffers and saline preparations where the ozone actually acts as a package sanitizer. In these cases, packaging studies performed would need to show the ozone had naturally dissipated or a small residual was intact.

Instrumentation for monitoring and controlling ozone in water sanitization systems for pharmaceuticals

There are two common methods for monitoring and subsequently controlling ozone within an ozonated water sterilization system. A first method utilizes electrochemical technology where an electrolyte is embedded within a solid state electrical probe. This method is also referred to as “amperometric” or “polarographic”. A second method utilizes UV light absorption, commonly referred to as “photometry”, which employs a UV light source and a UV sensor.

Electrochemical Method

In the electrochemical (EC) method an electrical potential is applied between the electrodes to reduce ozone that is driven through the membrane by a partial pressure gradient. The resulting electrical current is proportional in magnitude to the concentration of interest. This is measured by the indicating instrument, scaled, displayed and converted to analog and digital outputs.

UV Absorption Method

UV absorption has become a common, proven optical method to determine ozone concentrations within liquids and air. Because ozone absorbs UV light predominantly at a wavelength of 254 nanometers (nm), an absorption spectrophotometer can be “tuned” to that specific wavelength to routinely measure the presence of ozone down to concentrations to single parts per billion (ppb). (Ref. Fig 1).

The appeal of UV absorption photometers for dissolved ozone measurement derives from a number of factors including: a) the strong absorption at 254 nm lends to instruments with small-geometries that can measure a very wide range of ozone densities, b) a fairly good selectivity to ozone that can be achieved at this wavelength for most applications, c) the demonstrated ability that in the single PPBw levels, especially in the post destruct applications, the measurement by UV offers a more stable reading, and d) the reliable light sources that produce the desired discrete spectrum are available in as commercially available mercury vapor lamps.

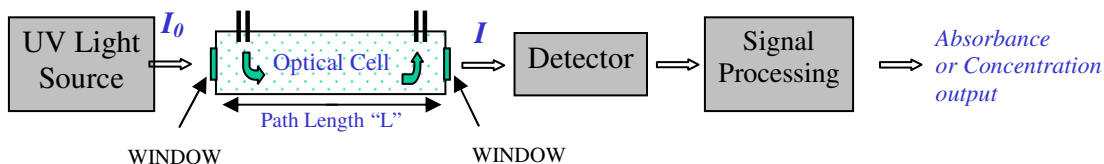


Figure 1 Single Path UV Absorption Platform for Ozone Measurement in Gas or Liquid phase (courtesy of IN USA Inc.)

The initial cost of UV absorption monitoring and control instrumentation has been found to be significantly less expensive than the most commonly installed EC instrumentation. Also, the overall cost of ownership of EC instrumentation has shown to be significantly higher than that of UV absorption for the following reasons:

- 1) EC probes require frequent replacement of electrolyte and membrane due to the short lifetime of the electrolytes (consumables). Replacement is a labor intensive process resulting in costly system downtime. Also, electrolytes tend to leach over time which can add to the frequency of replacement,
- 2) EC probes require frequent calibration which is labor intensive resulting in costly system downtime. This may be due to the fact that EC probes are calibrated in air (not in ozone) and are therefore prone to cross interference from dissolved gases resulting in questionable data.

Whereas UV absorption photometers require only the UV lamp to be replaced about every eighteen months. No recalibration is required since the measurement is ratiometric and in accordance with Beer Lambert law;

$$C = \frac{\ln\left(\frac{I_0}{I}\right)}{\epsilon \times L}$$

where the intensity of light of the reference I_0 over the intensity of light from the sample gas I is being measured, C is the resulting ozone concentration, L is the optical path length (as in Fig 1) and ϵ is a constant.

Conclusion

Dissolved ozone is a powerful and effective ambient water loop sanitizer gaining in popularity with pharmaceutical companies. The properties of this dissolved gas are unique and may be somewhat unfamiliar to the pharmaceutical professionals who normally deal with temperature or liquid chemical sanitization methods. Dissolved ozone measurement is an essential indicator for quality control use.

APPENDIX

Dissolved Ozone Materials Compatibility Chart (@20°C)

Material	Durability Grade
Silicone	D
EPDM/EPR	C
Buna N (Nitrile)	D
Viton	B
Kalrez/Simrez	AA

Teflon (PTFE)	A
PFA	A
PEEK	A
PVC	B
Delrin	D
Brass	B
Carbon Steel	C
316L SS	A
Hasteloy C	A
Monel	A
Titanium	AA

Legend:

AA= exceptional - never replace due to ozone damage.

A= excellent – replace only as needed or very seldom.

B= good – replace at defined time intervals.

C= susceptible to damage, replace at short time intervals or requires monitoring.

D= generally not acceptable, short term exposure only.

¹ Sweeting, Linda M., Oxidizing Agents, <http://pages.towson.edu/ladon/orgrxs/reagent/oxidizer.htm>, 1998

² ISPE Baseline Series, Vol. 4, Water and Steam Systems, Appendix to First edition, 2001, section 11.8.6.2 comparisons with chlorine, pg 59.

³ Fetner R.H.; Ingols, R.S., A comparison of the Bactericidal Activity of Ozone and Chlorine Against *Escherichia Coli* at 1°, J. Gen. Microbiol., 15, pp 381-385, 1956

⁴ "Chemical Synthesis with Ozone". *Ozone-Information.com*. http://www.ozone-information.com/Chemical_Synthesis_Ozone.html. Retrieved 2008-05-17

⁵ Delimpasis, K.J., http://www.ozonesolutions.com/Ozone_Color_Removal.html

⁶ 58th Annual International Water Conference, November 3-5, 1997, Pittsburgh, PA, W.R. Harrison, et. al., Paper titled "Reduction of TOC/THM Contaminants out of a UF-RO-EDI Membrane System at PECO's Limerick Generating Station"

⁷ J. Parenteral Sci. Technol., Vol. 45, No. 4, 1991, pp. 183–186, Lee, M.G., and P.B. Hunt, J. Vallor, "The Rate of Endotoxin Destruction during Water Treatment Using a Combination of Ozone and Ultraviolet Radiation."

⁸ UPW Magazine, Vol. 22 No. 7, Oct. 2005, Page 8, "Study finds ozone effectively removes endocrine receptors."

⁹ Water Quality Association, 1998 Annual Conference, R.G. Rice, PhD, et al, Paper titled "Ozone and the Safe Drinking Water Act"

¹⁰ United States Pharmacopoeia, Volume 28, valid January 1, 2005, Water for Injection and Purified Water Monographs.

¹¹ European Pharmacopoeia, Fifth Edition, Issue 5.2, valid July 1, 2005, Water for Injections, Purified Water, Highly Purified Water Monographs.

¹² Mazzei® type injector is one variety, www.mazzei.net, Mazzei Injector Corporation, Bakersfield, CA

¹³ OSHA temporary and permissible exposure limits are detailed at www.osha.gov reference 29 CFR 1910.1000

¹⁴ FDA CDER's Risk Based cGMPs for the 21st Century, www.fda.gov/cder/gmp/

¹⁵ FDA CDER's PAT Initiative, www.fda.gov/cder/OPS/PAT.htm

¹⁶ United States Pharmacopoeia, Volume 29, January 1, 2006, Water for Injection and Purified Water Monographs.