# Research on ozone application as disinfectant and action mechanisms on wastewater microorganisms

**M. N. Rojas-Valencia**[1]

[1] National Autonomous University of Mexico, Institute of Engineering, Coordination of Environmental Engineering, Post Box 70-472, Coyoacán 04510, Mexico, D.F. Mexico. Tel. (52) (55) 56233600. E-mail: nrov@iingen.unam.mx.

Although the use of ozone in wastewater treatment plants is not a common practice, it has been known for over one hundred years. The ozone capacity as wastewater disinfectant was acknowledged in 1886 by Meritens. The first industrial application of ozone in water treatment was performed in 1893 in Holland. Since that time, its use has spread in Europe and the USA. Owing to its oxidizing properties, ozone is currently known as one of the most efficient and fastest microbicides. The evidence has shown that it can break cell membrane or protoplasm, making it impossible to activate bacteria, virus and protozoa cells, removing up to 99% of bacteria and viruses at 10 mg/l in 10 minutes. It attacks mainly unsaturated fatty acids, lipid fatty acids, glycoproteins, glycolipids, amino acids and sulphydryl groups of certain enzymes; DNA is not ozone-resistant. Different studies have shown that ozone can destroy pathogenic and non-pathogenic microorganisms such as: viruses, bacteria, fungi, spores, protozoa, nematodes (helminth eggs) and algae. In this chapter, the results of studies on the application of ozone as disinfectant and its action mechanisms for destroying pathogenic microorganisms are detailed.

**Keywords:** action mechanisms; disinfectant; ozone; wastewater.

## 1. Introduction

The capacity of ozone as contaminated water disinfectant was acknowledged in 1886 by Meritens [1]. In 1889, the French chemist Marius Paul Otto started studying ozone at the University of Paris [2].

In the United States of America, the installed disinfection plants (25 in 1992) have two points of application: as primary disinfection (for treating trihalomethanes by-products) and as secondary disinfection (for microorganism removal). In Europe, particularly in France, ozone is used for disinfection at the end of the treatment train and chlorine can be added for presenting microbiological or algae growth in pipes.

Because of its oxidizing properties, ozone is considered one of the fastest and most efficient known microbicides. It can break cell membrane or protoplasm, inhabilitating cellular reactivation of bacteria, coliforms, viruses and protozoa, removing up to 99 % of bacteria and viruses at 10 mg/L in 10 minutes, attacking mainly unsaturated fatty acids, lipid fatty acids, glycoproteins, glycolipids, amino acids and sulfhydryl groups of some enzymes; DNA is nor ozone resistant [1, 3, 4, 5].

Genera such as: *Pseudomonas, Flavobacterium, Streptococcus, Legionella*, etc. are some of the bacteria removed through ozone treatment while among fungi, *Candida aspergillus* can be mentioned. Hereinafter, the results of studies on ozone application on viruses, bacteria, fungi, protozoa, helminths and algae are detailed.

## 2. Viruses

Viruses are small particles considered borderline between live beings and inert matter that can only live and reproduce parasiting cells and causing their destruction.

Contrary to bacteria, viruses are always harmful and cause diseases such as influenza, cold, measles, small pox, chicken pox, German measles and poliomyelitis.

Ozone acts on viruses oxidizing the proteins of their envelope and modifying their three-dimensional structure. When this occurs, the virus cannot anchor itself onto the host cell and thus cannot reproduce and dies. Type II and III viruses are less resistant and their destruction is complete [6, 3].

Ozone viricide action is observable at lower concentrations than its bactericide action because the viral envelope is less complex than the bacterial wall. Viruses are generally more ozone resistant than vegetative bacteria but no more than the sporulated forms such as *Mycobacterium*.

Table 1 shows the results of ozone application at different doses, temperatures and pHs, contact time (CT) values can also be seen for inactivating viruses, taken from the US EPA Guideline Documents [7].

**Table 1** Results of ozone application on viruses.

| Organisms | Dose $O_3$ (mg/L) | Time (min) | Temperature °C | | | | | pH | Log | Reduction (%) | References |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | 5 | 10 | 15 | 20 | 25 | | | | |
| *Polio I* | 0.4 | 3<br>0.4 -1.5 | --- | --- | --- | --- | --- | 7.2 | --- | 99 | [3] |
| *Polio II* | 4-8.5 | --- | --- | --- | --- | --- | --- | 7.2 | 3 | 99 | [8] |
| *Polio III* | --- | --- | --- | --- | --- | --- | --- | 7.4 | --- | 99.5 | [5] |
| *Rota SA II* | --- | 0.12- | <5 | --- | --- | --- | --- | 6.8 | --- | 99 | |
| *V. aughn* | | 0.19 | | | | | | | | | |
| *V. EEE* | 0.25 | 10 | --- | --- | --- | --- | --- | --- | --- | --- | [6] |
| *canine distemper virus* | 0.43 | 5 | --- | --- | --- | --- | --- | --- | --- | --- | |
| *V. coxsackie* | 0.51-33 | --- | --- | --- | --- | --- | --- | --- | 3 | --- | [3 , 5] |
| *V. porcine* | 0.024 | --- | --- | --- | --- | --- | --- | --- | --- | --- | [3] |
| *Virus inactivation* | 0.6 | --- | 0.6<br>0.9<br>1.2 | 0.5<br>0.8<br>1.0 | 0.3<br>0.5<br>0.6 | 0.3<br>0.4<br>0.5 | 0.15<br>0.25<br>0.3 | --- | 2.0<br>3.0<br>4.0 | --- | [7] |

*Virus EEE*= encephalomyelitis virus. --- = Not reported

## 3. Fungi

Some types of fungi can cause diseases to human beings. Many others can cause food alteration, turning them unacceptable for consumption, such as molds, among others. It is thus advantageous to handle and eliminate said pathogenic forms, the spores of which are found in all types of environments. Ozone eliminates them through its oxidizing action causing them an irreversible cellular damage as shown in Table 2 [3, 6, 9].

**Table 2** Results of ozone application in fungi.

| Organisms | Dose $O_3$ (mg/L) | Time (min) | Temperature (°C) | pH | Log | Reduction (%) | References |
|---|---|---|---|---|---|---|---|
| *Candida albicans* | 1.5 | 10 | 25 | 7.2 | ---- | 99 | [10] |
| *Candida parapsilosis* | | ---- | ---- | ---- | ---- | ---- | |
| *Candida aspergillus* | | ---- | ---- | ---- | ---- | ---- | |
| *Clostridium chauvoei* | 0.8 –2.0 | 180 | 20-30 | ---- | 4 | ---- | [6, 9] |
| *Clostridium tetani* | | | | | 6.8 | | |
| *Tricophyton verrucosum* | | | | | 3.7 | | |
| *Aspergillus flavus* | 60<br>11 | 4<br>60 | ---- | ---- | ---- | 78 % | [11] |

--- = Not registered

## 4. Spores

Some fungi and bacteria in adverse conditions generate a thick envelope and paralyze their metabolic activity, remaining in a latent state (spores). When conditions turn favorable, they develop normally and their metabolism recovers its activity. Said resistance forms are known as spores and are typical of bacteria such as the ones causing tetanus, gas gangrene, botulism and anthrax.

This resistance mechanism makes it very difficult to fight against them and generally useful treatments such as high temperatures and the use of antimicrobial agents become inefficient. Ozone at concentrations slightly higher than the ones used for the rest of bacteria can overcome spores resistance.

## 5. Bacteria

Since the beginning of the century ozone has been used in water treatment as can be seen in several works registered in Table 3. Its bactericidal and bacteriostatic effect is obvious at low concentrations ($\leq$ 0.01 ppm) and during very short exposition periods.

The following mechanisms are attributed to ozone disinfecting power: lethal oxidation of bacterial protoplasm, membrane oxidation followed by lysis, cell electron transfer or capture thus irreversibly altering the buffering mechanism and membrane alteration by ozonolysis of unsaturated fatty acids constituting the external membrane.

Low ozone concentrations and short contact times are sufficient for disinfecting mixed waters. However, it is impossible to extrapolate results, even less so when the weather condition favors the adaptation and development of a range of strains from the most diverse genera.

*Vibrio cholerae* causes cholera, an acute and severe bacterial intestinal disease that has been detected as the cause of important problems because of its environmental resistance. *V. cholerae 01* adapts to chlorine becoming more rugose

and so does *S. typhi* [12, 13]. *S. typhi* is the etiologic agent of typhoid fever. Said pathogen produces an endotoxin causing fever and diarrhea. An adequate water treatment helps handle said microorganism. As can be seen in Table 3, the international literature does not have registrations regarding ozone disinfectant evaluation on *Salmonella typhi and Vibrio cholerae 01* found in wastewaters.

**Table 3** Results of ozone application on bacteria in different types of water.

| Organisms | Dose $O_3$ (mg/L) | Time (min) | Temperature (°C) | pH | Log | Reduction (%) | References |
|---|---|---|---|---|---|---|---|
| **Gram-negative bacteria** | | | | | | | |
| *Escherichia coli* | 0.6-1 | | 12 | | 4 | 99.99 | [14] |
| | 0.01- 2.4 | 2 – 8.33 | 25 | 7.2 | | 90 | |
| *Staphylococcus sp.* | 0.6-10 | 0.5 –0.6 | | ---- | ---- | ---- | [14] |
| *Pseudomonas aeruginosa* | 2 | 0.18 – 4 | 25 | 7.2 | | 90 | [15] |
| *Pseudomonas flourescens* | | | | | | | [16] |
| *Streptococcus fecalis* | 0.6 | 1 | 25 – 30 | ---- | 99 | ---- | [4] |
| *Mycrobacterium tuberculosis* | 0.6 | 6 | ---- | ---- | ---- | ---- | [4] |
| *Fecal Coliforms* | | | ---- | ---- | | 1000 | |
| Tertiary effluent | 2 | | | | | (UFC/100 | |
| Secondary effluent | 6-17 | 0.6 | | | | mL) | [4] |
| Primary effluent | 20 | | | | | | [8] |
| Pretreated water | 25-30 | | | | | | |
| Pretreated wastewater | 30 | 2 | | | 2-3 | | |
| Municipal wastewater | 5-10 | 5-15 | | | 2 | | [17] |
| *Total Coliforms* (Municipal wastewater) | 5-10 | 5-15 | ---- | ---- | ---- | ---- | [18] |
| *Salmonella typhi* | 0.46-0.78 | 10 | ---- | ---- | ---- | ---- | |
| *Vibrio cholerae* | 0.48-0.84 | 15 | --- | ---- | ---- | 95 | |
| (Spring water) | 1.7 | 8 | | | | 100 | [13] |
| *Salmonella typhimurium* | 2.6 | 3.59 | 25 | 7.2 | | 89 | [15] |
| *Shigella sonnei* | | 1.35 | 25 | 7.2 | | 97 | |
| **Gram-positive bacteria** | | | | | | | |
| *Staphylococcus aureus* | 1.97 | 10 | 25 | 7.2 | | 99 | [10] |
| *Streptococcus faecalis* | 1.97 | 10 | 25 | 7.2 | | 99 | |
| *Brucella abortus* | 1.13 | 60 | 30 | ---- | ---- | ---- | [9] |
| *Pasteurella multocida* | | | | ---- | ---- | ---- | |
| *T. Coliform* | 300 | 20 | ---- | 7.2 | | 99 | [19] |
| *F. Coliform* | | | | | | | |
| *Streptococcus* | | | | | | | |
| *Salmonella* | | | | | | | |
| *E. coli* | | | | | | | |
| Heterotrophic bacteria | 0.1 | | 1 | 14.3 | 7.5 | 1.6 | 97.5 | |
| Coliform bacteria | 0.1 | 1 | | | 22 | 99.4 | [20] |
| *C. perfringens* | 23 | 5 | | 6.9 | 5.1 | | [21] |
| *Bacillus atrophaeus* | 20 | 2 | 25 | 4 | 5.25 | | [22] |

--- = Not registered

From previous experience, it is known that ozone can break down cell membranes and protoplasm, and that this process impedes cell reactivation in bacteria, coliform, virus, and protozoa [1, 5].

Ozone inactivates bacteria by means of oxidation reactions [10]. As can be seen in Figure 2, a) the cell membrane is the first site under attack; then b) the ozone attacks glycoproteins, glycolipids, or certain aminoacids, and also acts upon the sulfhydril groups of certain enzymes [5]; c) the effect of ozone on the cell wall begins to become apparent; d) the bacterial cell begins to break down after being in contact with ozone; e) the cell membrane is perforated during this process; and finally in f) the cell disintegrates or suffers cellular lysis (see Figure 1).

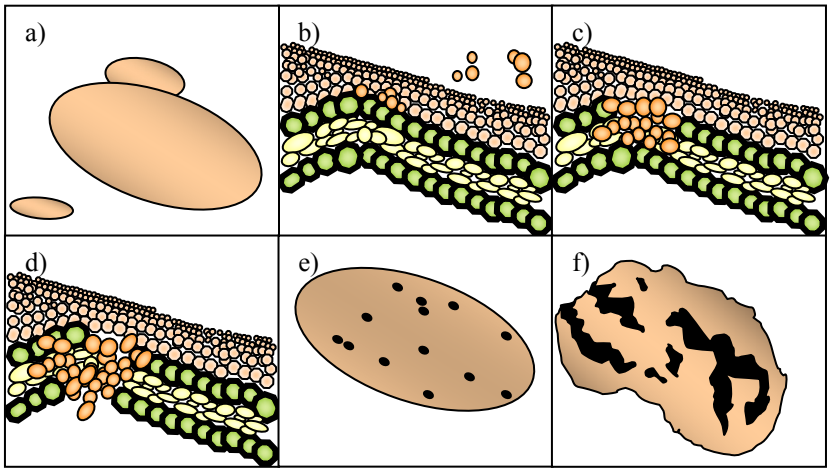



**Fig. 1**. Bacterias undergoing lysis during disinfection with ozone.

## 5. Protozoa

Protozoa present in their life cycle a vegetative fragile phase (trophozoites) and a more resistant phase (cysts). Cystic protozoa are much more resistant than viruses and bacteria vegetative forms. Cystic *Giardia lamblia* has an ozone sensitivity equivalent to the sensitivity of *Mycobacterium sp.* sporulated form [1]. Ozone is the most effective disinfectant for deactivating protozoa such as *Cryptosporidium sp.* [23, 24]. *Cryptosporidium sp.* is considered the most resistant protozoa and is ten times more resistant than *Giardia sp.* [5].

Ozone destroys protozoa cell membrane. This may be because it affects the wall, making it more permeable. Thus aqueous ozone enters the cyst and damages the cytoplasmic membrane. Nucleus, ribosome and other structural components [5] are penetrated. *Cryptosporidium sp.* concentrations are taken as the disinfection criterion proposed by the EPA for being one of the most difficult germs to remove [1]. Table 4 shows the results of ozone application on some protozoa and the time requested for *Giardia sp.* inactivation using different temperatures.

**Table 4.** Results of ozone application on some protozoa.

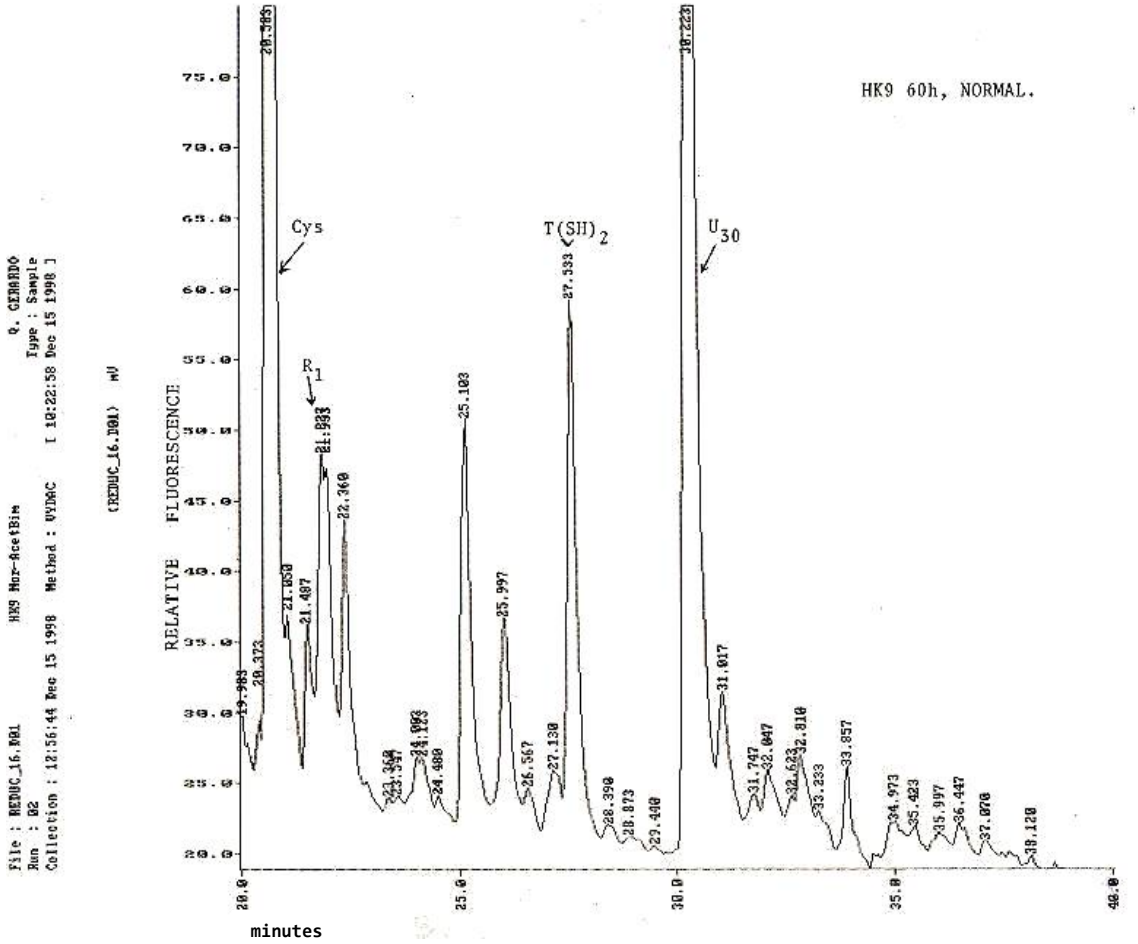
| Organisms | Dose $O_3$ (mg/L) | Time (min) | Temperature °C | | | | | pH | log | Reduction (%) | References |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | **5** | **10** | **15** | **20** | **25** | | | | |
| *Giardia lamblia* | 5-10 | 0.94- 5 | --- | ---- | ---- | ---- | ---- | 7 | 1.0 | 99 | [1] |
| *Giardia lamblia* | 10 | ---- | 0.32 | 0.23 | 0.16 | 0.12 | 0.08 | ---- | 0.5 | 99.9 | |
| | | | 0.63 | 0.48 | 0.32 | 0.24 | 0.16 | | 1.0 | | [7] |
| | | | 0.95 | 0.72 | 0.48 | 0.36 | 0.24 | | 1.5 | | [16] |
| | | | 1.3 | 0.95 | 0.63 | 0.48 | 0.32 | | 2.0 | | |
| | | | 1.6 | 1.2 | 0.79 | 0.60 | 0.40 | | 2.5 | | |
| | | | 1.9 | 1.4 | 0.95 | 0.72 | 0.48 | | 3.0 | | |
| *Giardia muris* | ---- | 2.8-12.9 | ---- | ---- | ---- | ---- | ---- | 7 | ---- | ---- | [1] |
| *Cryptosporidium* | 50/50 | ---- | ---- | ---- | ---- | ---- | ---- | 7 | ---- | 99 | [23] |
| *parvum* | 100/0 | | | | | | | | | | [24] |
| *Poliphaga sp.* | ---- | 4 | ---- | ---- | ---- | ---- | ---- | ---- | ---- | 95 | [23] |

--- = Not registered

Researchers working have found that cystic *Giardia lamblia* is as sensitive to ozone as the spore form of Mycobacteria [1]. Ozone has also been demonstrated as the most effective disinfectant for the inactivation of Cryptosporidium [23], and this is significant because Cryptosporidium is considered the most resistant of the protozoas, being as much as ten times more resistant than Giardia [5].

In addition to the above, results of experiments with *Acanthamoeba sp.* (see Figure 5) have demonstrated that these amoebas have mitochondrias and possess Glutation, as well as the gene for the glutation reductase, and that they possess Tiol compounds obtained through HPLC. This compound, Tiol, may be the target for the action of ozone against this parasite, because it has been reported that ozone enters into direct and swift action on the $R\text{-}HS^-$ sulfur compounds, with a velocity constant of $KO_3 = 1.1E10^6\ M^{-1}S^{-1}$ in an acid medium.

The survival percentage of each micro-organism resulting from the given CT with ozone, is reported in Table 1. In all cases, the micro-organisms were very susceptible to ozonation, and a marked reduction of bacterial concentration was observed. A linear correlation between the logarithm of bacterial concentration (N) and the contact time was found in all cases, the linear correlation coefficients (r) being significant ($\alpha = 0.05$) in all experiments (see Figures 2).




## 7. Algae

The ozonation of algae contaminated water oxides the algae and makes them emerge to the surface. Ozone oxidizes also the metabolic derivatives of the algae and eliminates undesirable tastes and odors (Richard and Dalga, 1993). Table 5 shows the results of ozone application on some algae using different temperatures and ozone doses, the applied doses ranging from 1.6 to 3.5 mg/L. Diatoms are the algae most difficult to eliminate obtaining only a 60 % reduction. No data are available on the temperatures and pH used.

**Table 5.** Results of ozone application on some genera of algae.

| Organisms | Dose $O_3$ (mg/L) | Time (min) | Temperature °C | pH | Log | Reduction (%) | References |
|---|---|---|---|---|---|---|---|
| Phytoplankton total | 1.6 | 8 | ---- | ---- | 1 | 99.5 | [25] |
| Diatoms | 1.6 | 8 | ---- | ---- | 1 | | |
| *Clorofita* | | | ---- | ---- | | | |
| *Cyanofita* | | | ---- | ---- | | | |
| Diatoms | 2.5-3.5 | ---- | ---- | ---- | ---- | 60 | [26] |
| **Blue green algae** | | | | | | | |
| *Aphanizomenon* | 2.5-3.5 | ---- | ---- | ---- | ---- | 90 | |
| *Anabaena* | | | | | | | |

--- = Not registered

## 8. Ozone destruction of helminths eggs

Despite their disinfection resistance, helminths eggs can be removed from water using physical-chemical treatments such as coagulation flocculation and filtration [27, 28], taking advantage of their size (20 to 100 μm) and their specific gravity (1 to 1.2). However, although this technique eliminates the water problem, it transfers it to muds, and thus other alternatives have been studied for destroying helminths eggs [29, 30].

Gadomska *et al.* (1991) [31] used 500 mg $O_3$/hour (8.3 mg $O_3$/L) and observed a helminths eggs reduction of 81.7, 94.6 and 95.7 % with contact times of 45, 90 and 180 minutes, respectively, without mentioning if the destruction was observed on viable or non-viable eggs.

Rojas and Orta (2000) [32] applied a concentration of 19 mg $O_3$/L (in 500 mL) in gas phase during 30 minutes. They observed that the disinfectant did not have effect on the structure and viability of *A. suum* fertile eggs at this dose and this contact time, because they found mobile larvae inside the egg, as well as in the witness. However, non-viable eggs were completely destroyed (see table 6).

**Table 6.** Results of ozone application on *Ascaris suum* eggs.

| Organisms | Dose $O_3$ (mg/L) | Time (min) | Temperature °C | pH | Reduction (%) | Observations |
|---|---|---|---|---|---|---|
| *Ascaris suum* | 8.3 | 180 | 19-22 | -- | 81-96 | ------ |
| *Ascaris suum* | 18 | 180 | 20 | -- | 94 | ------ |
| *Ascaris suum* | 19 | 30 | 20 | 7 | 90 | Non-viable eggs |

Source: [31, 32] --- = Not registered

All helminth eggs are morphologically similar. They are also of similar size and chemical constitution. The egg shell consists of three basic layers that are secreted by the egg itself, namely, a lipoidal inner layer, a chitinous middle layer, and outer layer of protein (see Figure 3). Their variation mainly depends on the number of amino acids incorporated into the layers, a result that agrees with the similarity of results obtained when ozone is applied under acid conditions.

During the oxidation process, ozone breaks the wall or shell of helminth eggs. The acid medium causes hydrolysis of proteins, with amino acids as terminal products. The biphenyls and the quinones are characterized by the presence of the OH donor group in aromatic nuclei. This donor group is strongly reactive to ozone. Doré (1989) reported velocity constants for ozonation of cysteine at pH 2, and cystine at pH 1.8, as 3 X $10^4$ and 5,5 X $10^2$ $M^{-1}s^{-1}$, respectively, concluding that sulphur amino acids are highly reactive to ozone. Such observations agree with the results obtained during this research, using pH 3. At pH 3, after the first hour, a removal rate of 96.7% of *Ascaris suum* eggs was achieved. This maximum removal confirms the influence of acid conditions on an increased reactivity of ozone. The double bond carbon-carbon, plus the sulphur and nitrogen atoms in the lateral chain of the amino acids, also constitute very selective attack centres for ozone [33].

Figure 3 The egg shell consists of three basic layers: a lipoidal inner layer, a chitinous middle layer, and outer layer of protein. Results obtained indicated that the degradation of the structure of each amino acid by ozone, depends on the applied ozone dose. The amino acids which contained a high reactive side chain, were preferentially attacked by ozone.

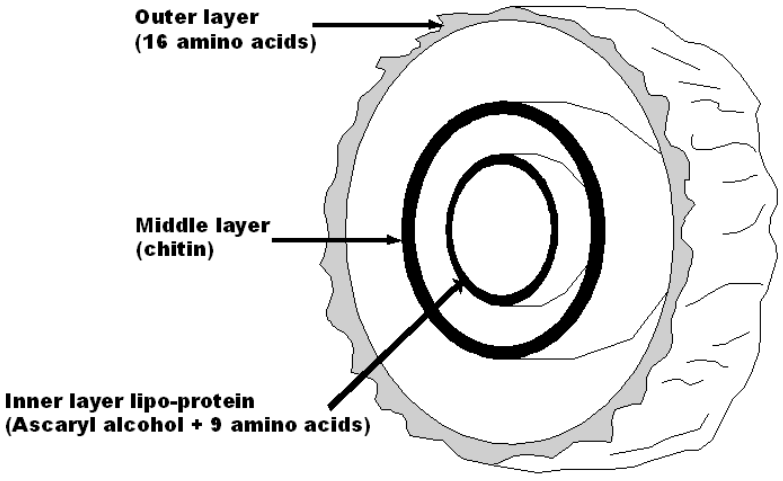



**Fig. 3.** The egg shell consists of three basic layers: a lipoidal inner layer, a chitinous middle layer, and outer layer of protein.

Results obtained indicated that the degradation of the structure of each amino acid by ozone, depends on the applied ozone dose. The amino acids which contained a high reactive side chain, were preferentially attacked by ozone.

It can be stated that ozone shows a significant reaction on the amino acids that form the shell of intestinal parasites, particularly in an acid pH medium, because the ozone acts upon the nitrogen atom, or upon the R group (alkyl sulphurated, or insaturated), or on both at the same time. Thus polypeptide or protein reactivity will depend on the nature of their constituent amino acids.

Using thin-layer chromatography, the following amino acids constituting the shell of *Ascaris lumbricoides* eggs have been identified: lysine, arginine, glutamic acid, serine, glycine, cystine, aspartic acid, threonine, alanine, valine,

tyrosine, leucine, isoleucine, tryptophane, phenylalanine and proline [33]. Results have been identified through liquid chromatography, show the components that form the external layers of helminth eggs (Figure 4).

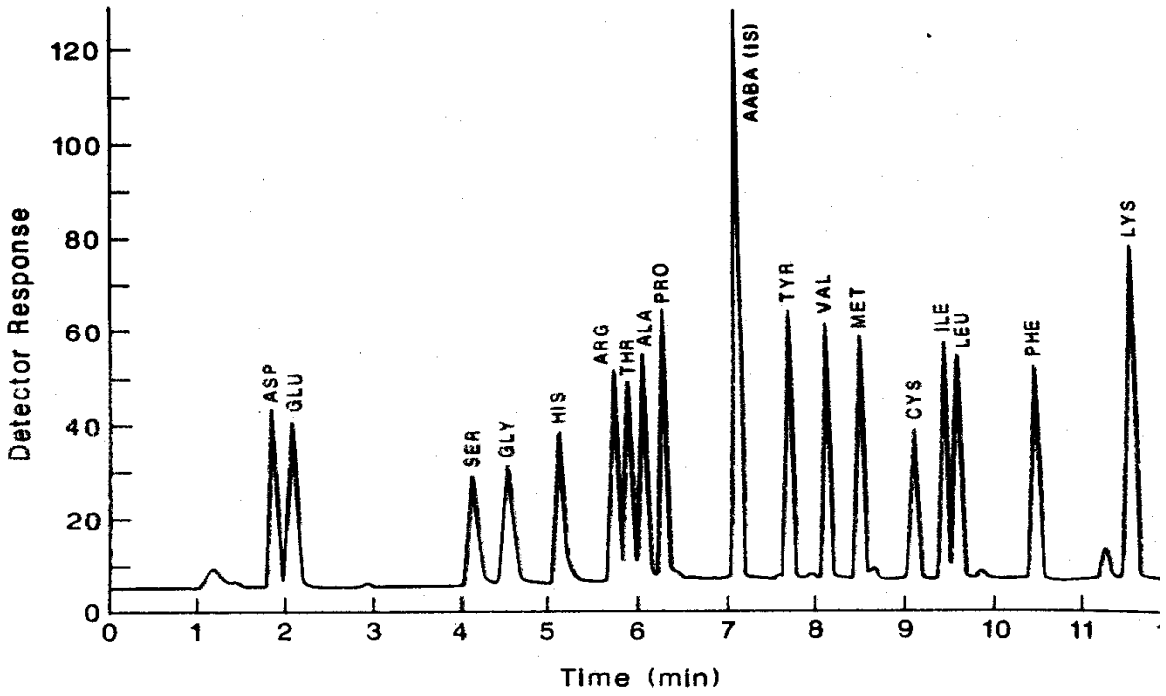



**Fig. 4.** Chromatogram of amino acids constituting the shell of *Ascaris lumbricoides* eggs. Separation of dansyl amino acids by HPLC, with UV detection at 250 nm.

Amino acids are symbolized by codes (according to the IUPAC-IUB Commission on Biochemical Nomenclature): ASP = aspartic acid; GLU= glutamic acid; SER= serine; THR= threonine; GLY= glycine; ALA= alanine; ARG = arginine; PRO= proline; VAL= valine, MET= methionine; ILE= isoleucine; LEU= leucine; W= tryptophan; PHE= phenylalanine; CYS= cystine; O= ornithine; LYS= Iysine; TYR= tyrosine.

Background information shows that the ozonation is a viable alternative showing interesting perspectives upon being applied to water treatment for various purposes.

In conclusion, it can be stated that ozone is a powerful oxidizing agent that can help destroy any type of pathogenic and non-pathogenic microorganisms.

## 9. Comparison of cost estimates for the two conventional disinfectants

Costs of chlorine disinfection systems depend on the manufacturer, location and capacity of the treatment plant, and on the characteristics of the wastewater to be treated.

Hypochlorite compounds, for example, tend to be more expensive than chlorine gas (see Table 5). Nevertheless, many large cities have chosen to use hypochlorite simply to avoid transporting chlorine gas through built-up areas. In addition to the costs of chlorination, in some cases it is also necessary to include the cost of dechlorination, because this can increase the total cost of disinfection by a further 30 to 50 per cent.

Annual costs for running and upkeep in a chlorine disinfection system also include electricity consumption, chemical compounds and cleaning materials, repair of equipment, and costs for employing personnel. The results of a 1995 study by the Water Environment Research Foundation, using secondary effluents from disinfection installations with flows of 0.04 to 7.4 $m^3/s$, revealed disinfection costs of \$28.14 USD/1000 $m^3$ or 0.02814 USD/$m^3$ for a dose of chlorine of 20 mg/L; and costs of \$40.55 USD/1000 $m^3$ or 0.04055 USD/$m^3$ for dechlorination [5].

To compare costs, contact times and the logarithmic reduction for each disinfectant, a dose has first to be established. The doses necessary for the inactivation of different micro-organisms vary substantially from one disinfectant to another, and also for the same disinfectant when applied to different micro-organisms (see Table 7).

**Table 7.** Comparative summary, results obtained and costs of the two disinfectants applied to tertiary effluents

| Disinfectant | Micro-organisms | Dose | Contact Time (min) | Log reduction | Method of disinfection | Costs USD/ $m^3$ | Reference |
|---|---|---|---|---|---|---|---|
| Chlorine | Fecal Coliforms | 5 to 20 (mg/L) | 15- 30 | 4 | Hypochlorite | 0.0547 | Costs calculated in Mexico |
| | | | | | Chlorine gas | 0.0292 | Costs calculated in Mexico |
| | | | | | Chlorine gas | 0.0405 | [5] |
| Ozone | Fecal Coliforms | 15 (mg/L) | 5 | 4 | Ozone | 0.043 | [16] |

At the present time, in terms of cost, chlorination is more efficient [\$0.028 USD/$m^3$] than disinfection with ozone [\$0.043 USD/$m^3$]. However, when de-chlorination is required, this elevates the cost to \$0.0427 USD/$m^3$, which effectively evens out the eventual cost.

From a bibliographic review, it can be said that ozone has the greatest germicidal power, followed by chlorine. Ozone is 25 times more effective than hypochloric acid (HOCl); 2,500 to 3,000 times more potent and swifter than hypochlorite (OCl); and 5,000 times better than chloramine ($NH_2Cl$). These results have been measured by comparing the constants of time against concentration (CT) needed to eliminate 99.99% of all micro-organisms [34].

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