THE EVALUATION OF ALTERNATIVE DISINFECTION PROCESSES FOR THE REMOVAL OF PROTOZOAN OOCYSTS AND CYSTS AND OTHER MICRO-ORGANISMS, IN THE TREATMENT OF FINAL WASTEWATER

Final Report

to the

Water Research Commission

by

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Executive Summary

Wastewater effluents are routinely chlorinated prior to discharge to river or sea, but low concentrations of chlorine residuals are toxic to aquatic life and some chlorination by-products have been shown to be mutagenic. The chlorine residual concentration limit imposed on discharge of wastewater effluents in this country by the Department of Water Affairs and Forestry (DWAF) is already relatively low (general standard of 0,1 mg/L) and is set to become even more stringent, although the limit for bacterial indicator organisms will not change (general standard of 0 faecal coliforms per 100 mL).

The tests carried out by Umgeni Water for the purposes of this investigation, determined *Eschericia coli* specifically, rather than total faecal coliforms. Essentially, there is little difference between *E. coli* and faecal coliform counts as *E. coli* are the main subgroup of faecal coliforms and therefore total faecal coliform counts will be generally slightly higher than *E. coli* counts, but the test specific for *E. coli* is more accurate than that for faecal coliforms. In practice, the *E. coli* results can be considered representative of faecal coliforms and for the purposes of this report are used interchangeably. Internationally much controversy surrounds the faecal coliform grouping, as not all of them are in fact faecal, leading to some confusion.

Achieving the general faecal coliform standard while not exceeding the chlorine residual limit of 0,1 mg/L is often not possible without abnormally large contact tanks and lowering the chlorine residual limit still further will only aggravate this situation. A number of pathogenic micro-organisms, especially viruses and parasitic cysts, such as *Cryptosporidium* and *Giardia*, are far more resistant to chlorine than faecal coliform organisms and can be found in the absence of normal bacterial indicator organisms such as *Eschericia coli*, faecal streptococci and coliform bacteria, so it can be assumed that in many cases pathogenic organisms are entering receiving waters via wastewater effluent discharges. This has serious implications in a water scarce country such as South Africa where a significant proportion of the population do not have access to treated water or adequate sanitation and rely on untreated river water as their only source of drinking water. Inadequate water supply and sanitation is largely responsible for more than 800 million estimated cases of diarrhoeal disease and 4,5 million associated deaths in developing countries every year. In South Africa alone 12 million people do not have access to an adequate potable water supply and 21 million lack basic sanitation.

Obviously more effective disinfection of wastewater effluents is required and a number of alternative disinfectants warrant investigation, including ultraviolet irradiation (UV) and ozone, since many pathogenic organisms are more susceptible to these than they are to chlorine. It is suspected that ozone and/or UV may inactivate these organisms without actually rendering them unviable, but infectivity tests using animals such as mice would be required in order to confirm this and these tests were beyond the scope of this investigation. Another option which should be assessed, is that of combined treatments including UV/chlorine and UV/ozone. Membranes could also possibly be employed in removing these pathogens on account of their size, as they are fairly large in microbiological terms (>4 nm).

1.1. Objectives

This project had a number of objectives. The objectives were to:

1. Adequately disinfect wastewater effluents in terms of parasitic (oo)cysts (*Giardia* and *Cryptosporidium*), viruses (coliphages) and bacterial indicator organisms (*E. coli*, faecal streptococci and coliforms) while not exceeding DWAF general standards for chlorine residual.

2. Adequately disinfect wastewater effluents in terms of the above without producing a chlorine residual.

3. Provide design and operational guidelines for simple and effective disinfection of wastewater effluents.

4. Provide a disinfectant protocol suitable for use under low technology applications.

5. Determine the potential for regeneration of micro-organisms, especially *Giardia*, *Cryptosporidium* and viruses after different methods of disinfection.

This project was expected to produce a number of research products, the most important being:

1. A tetramethyl red (TMR) - fluorescein diacetate (FDA) viability stain test for *Cryptosporidium* and *Giardia*. This test would allow for more comprehensive screening for protozoan cysts and would reduce the risk of outbreaks of disease

caused by these micro-organisms. This would be of benefit to all authorities involved in the treatment of potable water as well as to water and wastewater treatment consultants. This stain also has potential for use with flow cytometry.

2. The reduction or removal of chlorine residual from disinfected water, which would be ecologically beneficial.

3. A simple method for adequately disinfecting wastewater, which would be especially beneficial to rural communities where advanced technology is inappropriate.

4. Design and operational guidelines for effective wastewater disinfection, which would allow authorities involved in wastewater treatment to effectively disinfect wastewater without exceeding the general standards for chlorine residual concentration.

5. A research report detailing alternative disinfectants for adequate disinfection of wastewater effluents while not exceeding general standards for chlorine residual concentration, which would also benefit all authorities involved in the treatment of wastewater.

These objectives have, for the most part, been achieved. It has been shown that it is possible to adequately disinfect wastewater, while keeping within the DWAF standards for chlorine and in some cases without even producing a chlorine residual at all.

1.2. Results and Discussion

One of the primary aims of this project was to assess alternative disinfectants for use in wastewater applications, with special emphasis on minimising the chlorine residual produced while not compromising disinfection efficacy. Another important aspect considered was finding a disinfectant capable of inactivating parasitic organisms, such as *Cryptosporidium* and *Giardia*. For this reason a fairly wide range of disinfectants was investigated, both used independently and in conjunction with other disinfectants, in the hope that combinations might result in a synergistic effect capable of destroying these resistant organisms.

Both laboratory and pilot plant tests were conducted for this investigation. The wastewater effluent used in these tests came from the Darvill Wastewater Works (WWW) in Pietermaritzburg, which treats a predominantly domestic wastewater effluent and has a

nominal design capacity of 75 ML/day. The process consists of primary sedimentation, at times augmented using primary enhanced treatment with ferric salts, an activated sludge plant with aluminium sulphate and lime addition for phosphate removal, followed by secondary settling, chlorination and tertiary treatment through a series of oxidation ponds. The effluent used in both the laboratory and pilot plant tests was drawn out of the overflow channel from the secondary sedimentation tanks at a point well above the chlorination stage.

The effluent used in the laboratory tests was often, but not always, spiked with cultures of *Cryptosporidium* oocysts and *Giardia* cysts, although the effluent regularly contained these prior to spiking. The pilot plant tests were however conducted on unspiked effluent

A variety of disinfectants and combinations of disinfectants were assessed both in the laboratory and at pilot plant scale, these being:

1 Chlorine
2 Ozone
3 Low pressure UV irradiation
4 Medium pressure UV irradiation
5 Multiwave UV irradiation
6 Peracetic acid
7 Hydrogen peroxide
8 Peracetic acid and hydrogen peroxide
9 Bromine
10 Klor-Free: a potassium persulphate and copper sulphate based disinfectant (laboratory testing only)
11 Mixed oxidant generator (only laboratory testing)
12 Ozone combinations:
    - ozone / chlorine
    - ozone / low pressure UV
    - ozone / medium pressure UV
    - ozone / peracetic acid
    - ozone / bromine
13 UV combinations:
    - UV / chlorine
    - UV / peracetic acid
    - UV / bromine
    - UV (multiwave only) / hydrogen peroxide

A number of tests were conducted on the secondary effluent before and after being exposed to these various disinfectants. The microbial testing protocol included all or some of the following:

1 *E. coli*
2 Coliform organisms
3 Faecal streptococci
4 Total colony counts at 37 and 22°C
5 Coliphages
6 *Cryptosporidium*
7 *Giardia*

The following analyses were also conducted on many of the samples:

1 Total Organic Carbon (TOC)
2 Dissolved Organi Carbon (DOC)
3 Biodegradable Dissolved Organic Carbon (BDOC)
4 Trihalomethane formation potential (THMFP)
5 Ultraviolet (UV) extinction (254 nm).
6 pH
7 Temperature
8 Turbidity
9 Conductivity
10 Alkalinity
11 Calcium, magnesium and total hardness
12 Iron
13 Manganese
14 Total dissolved solids
15 Suspended solids
16 Chemical Oxygen Demand (COD)

In all cases the chlorine demand of the water was determined and whenever ozone was used, the ozone requirement was also measured.

1.3. Summary of the Results

A number of conclusions were made from this investigation. These are:

1 The most promising disinfectants for use in wastewater, which show potential for meeting the DWAF standard for effluent discharge in terms of both chlorine residual and faecal coliforms are:
ozone
medium pressure UV irradiation
bromine
peracetic acid.

2. Combinations of ozone and UV with other disinfectants are difficult to justify in terms of the improvements obtained in disinfection. It is possible to achieve similar disinfection at lower doses of the combined disinfectants than can be achieved using each disinfectant alone, but the additional costs and operational difficulties that would be incurred by using disinfectant combinations is not warranted by the improvement in disinfection.

3. The mixed oxidant generator for the most part produces hypochlorite and so does not offer any significant benefits over chlorine gas disinfection and Klor-Free did not prove to be a suitable alternative to chlorine for wastewater disinfection.

4 Work conducted by Umgeni Water on membranes for the treatment of potable water sludges made it clear that the nature of the sludge is vitally important and that serious operational problems, such as membrane fouling, could be expected when using membranes for these applications,. Therefore membranes were not investigated for the purposes of this investigation.

1.4. Cost Assessment

Based on the results of this investigation, ozone, medium pressure UV irradiation, peracetic acid and bromine were selected as potential candidates for the disinfection of wastewater treatment. A cost assessment was conducted and appears below. Chlorine is included in the cost assessment in order to provide a benchmark. The cost assessment has taken into account the dosages that will be required in order to satisfy the DWAF relaxation standard for wastewater discharge of 500 faecal coliforms/100 mL (<500 *E. coli* CFU/100 mL has been used) as well as the higher doses that will be needed if the general effluent standard of 0 faecal coliform CFU/100 mL is to be met (<10 *E. coli* CFU/100 mL has been used).

Cost assessment for most promising alternative disinfectants for doses required to meet both the relaxed (<500 CFU/100 mL) and general (0 CFU/100 mL) DWAF effluent discharge standards.

	Relaxed Standard		General Standard	
Disinfectant	**Dose for <500 *E. coli***	**c/ML Costs for <500 *E. coli***	**Dose for <10 *E. coli***	**c/ML Costs for <10 *E. coli***
Ozone	2 mg/L	3,45	8 mg/L	9,45
UV	40 mW.s/cm^2	3,72	150 mW.s/cm^2	11,2
Peracetic acid	5 mg/L	40,0	9,5 mg/L	76,0
Bromine	6 mg/L	50,0	9,5 mg/L	79,2
Chlorine	6 mg/L	3,00	9,5 mg/L	4,75

Ozone satisfied the DWAF relaxed effluent discharge standard of faecal coliforms less than 500 CFU/100 mL at doses generally less than 2 mg/L, but in satisfying the general standard of 0 faecal coliforms the ozone dose needed to be increased significantly to between 6 and 8 mg/L. Although the capital costs of ozone are expensive, this is offset in some degree by relatively low running costs. However, ozone requires skilled personnel in order to run and maintain the ozone generation plant, so this will most probably pose a problem at most wastewater works.

Using medium pressure UV irradiation it was possible to achieve 98% compliance of the relaxed discharge standard, even using doses as low as 30 mW.s/cm^2. However, to ensure better compliance of the relaxed discharge standard a slightly higher dose, between 40 and 50 mW.s/cm^2 would be recommended. In order to meet the general discharge standard it would be necessary to use higher UV doses and according to this investigation, doses of around 150 mW.s/cm^2 would be required to do this on a regular basis. Medium pressure UV has fairly high capital costs, but the running and maintenance costs are relatively low and in fact it compares very favourably with chlorine costs in achieving the special effluent discharge standard of 500 faecal coliform CFU/100 mL. It needs to be stressed that achievement of the general discharge standard of 0 faecal coliforms per 100 mL will be possible most of the time using doses much lower than 150 mW.s/cm^2. Another advantage to using UV is that it is extremely simple to maintain and operate.

Peracetic acid was found to have 100% compliance with the special effluent discharge standard when used at a concentration of 5 mg/L or more. However, in meeting the general

standard of 0 faceal coliforms it was necessary to almost double this dose. The current high price of peracetic acid would result in running costs which would be too high for this method of disinfection to be financially viable.

Bromine was found to be similar to chlorine for the disinfection of wastewater and in fact the bromine concentrations required to meet the special and general discharge standards were also similar. Bromine can be obtained in solution form as an 18 to 19% solution under the trade name of Aquatreat. However, at present the costs relative to chlorine are very high.

1.5. Recommendations for Future Research

Both ozone and UV have been researched fairly intensively and apart from work on high intensity, low pressure systems, no further work appears to be necessary at this stage.

The tests conducted in this investigation revealed that bromine is an effective disinfectant for wastewater effluent. However, the bromine solution used in the pilot plant tests described in this report, was found to be unstable and as such, not suitable for these applications. Different methods of bromination, such as ammonium bromide used in conjunction with chlorine warrant further investigation.

Peracetic acid was produced in relatively small quantities for the experimental tests carried out for this investigation, but should full scale plant applications be considered, commercially available solutions would be required before this would be a feasible option. Peracetic acid is commercially available in Europe and this would need to be tested before implementation locally, but the cost would not be attractive under present conditions.

1.6. Conclusions

The conclusions in terms of the objectives of this project are discussed:

1. Adequately disinfect wastewater effluents in terms of parasitic (oo)cysts (*Giardia* and *Cryptosporidium*), viruses (coliphages) and bacterial indicator organisms (*E. coli*, faecal streptococci and coliforms) while not exceeding DWAF general standards for chlorine residual.

   It was shown that there are a number of disinfectants capable of achieving this, such as ozone, UV, peracetic acid, bromine and combinations of chlorine with ozone or UV.

2. Adequately disinfect wastewater effluents in terms of the above without producing a chlorine residual.
   This was also achieved by using disinfectants such as ozone, UV, peracetic acid and bromine.

3. Provide design and operational guidelines for simple and effective disinfection of wastewater effluents.
   The basic operational parameters required when using the suitable options are listed in this report.

4. Provide a disinfectant protocol suitable for use under low technology applications.
   UV is a possibility for rural applications, provided that adequate instrumentation is installed to detect failure of lamps or poor transmissivity. Obviously a pre-requisite for UV disinfection would be an adequate power supply. Peracetic acid could also provide a method of wastewater disinfection suitable for rural areas. It is comparable to hypochlorite solutions in terms of required handling precautions, it is more stable than hypochlorite solutions, doesn't produce a chlorine residual and only requires a simple dosing pump. However, the cost would be prohibitive at this stage, and will remain so until peracetic acid is imported in bulk or is manufactured locally.

5. Determine the potential for regeneration of micro-organisms, especially *Giardia, Cryptosporidium* and viruses after different methods of disinfection.
   For normal bacteriological indicator organisms and parasitic (oo)cysts, regrowth does not appear to be a problem within the limits of the indications of the tests.

1.7. Status in Terms of *Cryptosporidium* and *Giardia*

One of the problems experienced in carrying out the research for this project, was difficulty in establishing the viability of (oo)cysts once they had been detected. Future work into tests demonstrating the viability or infectivity of (oo)cysts is needed before definitive results on *Cryptosporidium* and *Giardia* inactivation can be reported for investigations of this nature. Measuring *Cryptosporidium* and *Giardia* recoveries tends to be generally low internationally (typically 30%) and improvement in detection and recovery techniques would be beneficial as regards accuracy of results.

The data generated during this project will be archived by Umgeni Water.

Acknowledgements

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The evaluation of alternative disinfection processes for the removal of protozoan oocysts and cysts and other micro-organisms, in the treatment of final wastewater effluents.

The Steering Committee responsible for this project consisted of the following persons:

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Prof. C A Buckley.....……	University of Natal
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Abbreviations and Glossary

BDOC	Biodegradable dissolved organic carbon
CFU	Colony forming units
COD	Chemical oxygen demand
C_t	Product of the disinfectant residual concentration in mg/L and the contact time in minutes.
Cysts	Environmentally resistant stage resulting from an asexual reproductive cycle.
DBP	Disinfection by-product
DIC	Differential interference contrast
DPD	Diethyl phenylenediamine
DOC	Dissolved organic carbon
DWAF	Department of Water Affairs and Forestry
E. coli	*Escherichia coli*, part of the faecal coliform group, but almost definitely of faecal origin; require more precise identification than other faecal coliforms.
EPA	Environmental Protection Agency
Excystation	Emergence of sporozites from oocysts (*Cryptosporidium*), or trophozoites from cysts (*Giardia*) to form the next stage in the lifecycle.
Faecal coliforms	Artificial grouping of coliforms capable of growth at elevated temperatures and an indicator of probable faecal pollution (not all are of faecal origin)
FDA	Fluorescein diacetate
FITC	Fluorescein isothiocyanate
ICP-AES	Inductively coupled plasma-atomic emission spectroscopy
In-vitro excystation	Artificially induced excystation in culture.
In-vivo infectivity	Deliberate attempted infection of a live animal with (oo)cysts (usually mouse by ingestion)
NOM	Natural organic matter
Oocysts	Environmentally resistant stage resulting from a sexual reproductive cycle.
Org	Organisms
PFU	Plaque forming units

SABS	South African Bureau of Standards
SANAS	South Africa National Standards
SEM	Scanning Electronic Microscope
SWRT	Surface Water Treatment Rule
THM	Trihalomethane
THMFP	Trihalomethane formation potential
TMR	Tetramethyl Red
TOC	Total organic carbon
UV	Ultraviolet irradiation
WWW	Wastewater Works

1. Introduction

Wastewater effluents are routinely chlorinated prior to discharge to river or sea. However, low concentrations of chlorine residuals are toxic to aquatic life (Ward and DeGraeve, 1978) and some chlorination by-products have been shown to be mutagenic (National Research Council, 1980). The chlorine residual concentration limit imposed on discharge of wastewater effluents in this country by the Department of Water Affairs and Forestry (DWAF) is already relatively low (general standard of 0,1 mg/l) and is set to become even more stringent, although the limit for bacterial indicator organisms will not change (general standard of 0 faecal coliforms per 100 ml) (Compilation of Water Quality Guidelines and Standards, 1995).

Achieving the general faecal coliform standard while not exceeding the chlorine residual limit of 0,1 mg/l is often not possible without abnormally large contact tanks and lowering the chlorine residual limit still further will only aggravate this situation. A number of pathogenic micro-organisms, especially viruses and parasitic cysts, such as *Cryptosporidium* and *Giardia*, are far more resistant to chlorine than are faecal coliforms (National Research Council, 1980) and can be found in the absence of normal bacterial indicator organisms such as *E. coli*, faecal streptococci and coliform bacteria (Slade et al, 1985). It can therefore be assumed that in many cases pathogenic organisms are entering receiving waters via wastewater effluent discharges. This has serious implications in a water scarce country such as South Africa where a significant proportion of the population do not have access to treated water or adequate sanitation and rely on untreated river water as their only source of drinking water. In fact, inadequate water supply and sanitation is largely responsible for more than 800 million estimated cases of diarrhoeal disease and 4,5 million associated deaths in developing countries every year (Esrey, 1996). In South Africa alone 12 million people do not have access to an adequate potable water supply and 21 million lack basic sanitation according to DWAF statistics from 1994 (DWAF, 2001).

Certain pathogenic micro-organisms, such as *Giardia* and *Cryptosporidium* and some viruses can pass through normal potable water treatment processes intact (Smith et al, 1995); a fairly recent epidemiological study estimated that a significant proportion of unreported diarrhoea which occurred in a population in suburban Montreal, Canada, was associated with the consumption of municipal tap water obtained from a river contaminated with human sewage, although this water had been conventionally treated and filtered and had met all the current

water quality regulations of that country (Payment et al, 1991a; Payment et al, 1991b). The implications of such studies are that contamination of source waters by human sewage not only poses a threat of disease to rural populations who use this water untreated, but could also lead to contamination of conventionally treated and disinfected potable water.

Obviously more effective disinfection of wastewater effluents is required than can be achieved using chlorine under the present DWAF residual chlorine standards and monitoring of the microbiological quality of such effluents needs to be extended beyond the usual bacterial indicator organisms such as *E. coli*, faecal streptococci and coliform organisms. A number of alternative disinfectants warrant investigation, including ultraviolet irradiation (UV) and ozone, since many pathogenic organisms are more susceptible to these than they are to chlorine (Sobsey, 1989; Kaur et al, 1994). Another option which should be assessed is that of combined treatments including UV/chlorine and UV/ozone and membrane filtration prior to disinfection by any of the above. It will also be necessary to monitor the extent of disinfection by measuring concentrations of parasitic cysts (*Giardia* and *Cryptosporidium*) and bacteriophages in addition to the normal bacterial indicator organisms (*E. coli*, faecal streptococci and coliforms).

Meaningful results from a study such as this would allow South African wastewater utilities to design wastewater disinfection systems which would provide effective disinfection of wastewater effluents while not exceeding the chlorine residual limits. This would be of benefit not only to the ecology, but also to the downstream populations.

1.1. Project Objectives

This project had a number of objectives. The objectives were to:

1. Adequately disinfect wastewater effluents in terms of parasitic (oo)cysts (*Giardia* and *Cryptosporidium*), viruses (coliphages) and bacterial indicator organisms (*E. coli*, faecal streptococci and coliforms) while not exceeding DWAF general standards for chlorine residual.

2. Adequately disinfect wastewater effluents in terms of the above without producing a chlorine residual.

3. Provide design and operational guidelines for simple and effective disinfection of wastewater effluents.

4. Provide a disinfectant protocol suitable for use under low technology applications.

5. Determine the potential for regeneration of micro-organisms, especially *Giardia*, *Cryptosporidium* and viruses after different methods of disinfection.

This project was expected to produce a number of research products, the most important being:

1. A tetramethyl red (TMR) - fluorescein diacetate (FDA) viability stain test for *Cryptosporidium* and *Giardia*. This test would allow for more comprehensive screening for protozoan cysts and would reduce the risk of outbreaks of disease caused by these micro-organisms. This would be of benefit to all authorities involved in the treatment of potable water as well as to water and wastewater treatment consultants. This stain also has potential for use with flow cytometry

2. The reduction or removal of chlorine residual from disinfected wastewater, which would be ecologically beneficial.

3. A simple method for adequately disinfecting wastewater, which would be especially beneficial to rural communities where advanced technology is inappropriate.

4. Design and operational guidelines for effective wastewater disinfection, which would allow authorities involved in wastewater treatment to effectively disinfect wastewater without exceeding the General Standards for chlorine residual concentration.

5. A research report detailing alternative disinfectants for adequate disinfection of wastewater effluents while not exceeding general standards for chlorine residual concentration, which would also benefit all authorities involved in the treatment of wastewater.

These objectives have, for the most part, been achieved. It has been shown that it is possible to adequately disinfect wastewater, while keeping within the DWAF standards for chlorine and in some cases without even producing a chlorine residual at all. Guidelines for the use of these

disinfectants are provided in this report. Some of these disinfectants would also be suitable for use in low technology applications, due to their ease of use. The regeneration of micro-organisms was investigated, although difficulties were encountered in assessing the regrowth potential of micro-organisms such a *Giardia* and *Cryptosporidium*, since infectivity tests would be required in order to assess this properly and such testing was beyond the capabilities of the project team. A more detailed account of this is provided in **Chapter 3** of this report.

2. Literature Review

2.1. Introduction

Wastewater effluents in this country are presently disinfected using chlorine prior to discharge to receiving rivers or the sea. Apart from the fact that chlorine is detrimental to aquatic organisms and that some of the chlorination by-products are carcinogenic, chlorine is not particularly effective against some pathogenic micro-organisms at the chlorine concentrations commonly employed in wastewater disinfection (National Research Council, 1980; Ransome et al, 1993). The literature suggests that chlorine residual concentrations in the region of 1 000 mg/*l* are required to reduce *Cryptosporidium parvum* viability by around 80% (Ransome et al, 1993; Smith et al, 1989) and these concentrations are obviously impractical for use in wastewater disinfection.

It has emerged over the past few years that controlling disinfection of water based on the removal of the indicator organisms traditionally used, such as *E. coli*, coliforms and faecal streptococci, can lead to outbreaks of waterborne diseases since a number of pathogens, particularly spore-bearing bacteria and micro-organisms with encapsulated structures such as *Cryptosporidium Spp.* and *Giardia Spp.*, are far more resistant to chlorine disinfection than these indicator organisms (Ransome et al, 1995). These organisms have also been shown to have an increased resistance to many other disinfectants that have been used in water and wastewater treatment, such as chloramines, chlorine dioxide and ultraviolet irradiation (Ransome et al, 1993).

The resistance of many of these organisms to traditional methods of disinfection is of concern as large sectors of the population do not have access to treated drinking water and rely on river water as a supply, often drawing water at points below a sewage works discharge. Increasing the chlorine residual during disinfection of wastewater effluents to levels that would effectively eliminate all pathogens is not only impractical, but hazardous to the ecology and can result in the formation of harmful chlorination by-products and taste and odour problems. Furthermore, the limits imposed on wastewater effluents by the DWAF do not permit high chlorine residuals and present indications are that these are set to become even more stringent.

It is obvious then that an alternative method of disinfection is required for the treatment of wastewater if effective disinfection is to be achieved under the present standards. A number of options are available, some consisting of combinations of various disinfectants and these are discussed below.

2.2. Ozone

Ozone is a very strong oxidising agent. In fact, if the standard oxidation potential of the chemicals generally used in water and wastewater disinfection is considered as the only factor controlling the effectiveness of disinfection, then ozone would be far more effective than any of the other chemicals. However, the disinfectant properties of a chemical are also dependent on a number of other factors related to water quality, such as suspended solids, oxidisable organic and inorganic matter, pH and temperature (Acher et al, 1997). Ozone is none the less a potent disinfectant and has been found to be far more effective for inactivation of parasitic cysts than chlorine (Peeters et al, 1989; Parker et al, 1993).

Oxidation reactions of ozone in water occur via two pathways, namely direct oxidation by molecular ozone and indirect oxidation by free radical species. These radical species can be formed by decomposition of ozone and from reactions of ozone with some inorganic and organic compounds (Hoigne and Bader, 1976; Hoigne, 1982, Forni et al, 1982). Ozone decomposition in water is initiated by hydroxyl ions and accelerated by a number of free radicals and anions which are formed as intermediates of the reaction (Staehelin and Hoigne, 1982; Tomiyasu et al, 1985). At low pH and in the presence of radical scavengers, such as carbonate ions, the indirect, radical reaction is inhibited and direct oxidation is the predominant reaction pathway (Staehelin and Hoigne, 1982; Hoigne et al, 1985; Singer, 1990). Direct oxidation by molecular oxidation is generally slower than the radical reaction and is also more selective (Hoigne, 1982). However, there is no clarity on which of these two reactions is responsible for the inactivation of micro-organisms by ozone, and while some researchers are of the opinion that direct oxidation is the predominant mechanism (Hoigne and Bader, 1978; Finch et al, 1992; Labatiuk et al, 1994), others believe that indirect oxidation by radicals is responsible for inactivation (Dahi, 1976; Bancroft et al, 1984).

Chlorination at levels used in water and wastewater treatment is ineffective in preventing excystation in oocysts (Korich et al, 1990), with concentrations as high as 16 000 mg/*l* being quoted as necessary for inactivation of these micro-organisms. Ozone however appears far more promising. Peeters et al, (1989) in tests conducted on neonatal mice, found that for water

containing a *Cryptosporidium parvum* concentration of 10^4 oocysts/mL, an ozone residual concentration of 1,11 mg/L for 6 minutes totally eliminated oocyst infectivity in the mice. An ozone residual of 2,27 mg/L for 8 minutes was sufficient to completely remove oocyst infectivity from water containing 5 x 10^5 oocysts/m*l*. Korich et al (1990), using infectivity to assess oocyst viability, measured more than a 90% reduction when oocysts were exposed to 1 mg/L ozone. They also demonstrated that fairly good agreement was obtained between *in vitro* excystation and *in vivo* infectivity as indicators of oocyst viability. Parker et al (1993) found that ozone residual concentrations of 3 to 5 mg/L maintained for between 2 and 6 minutes at 20°C were needed to completely inactivate oocysts when present in water at a concentration of 10^6 oocysts/L (i.e. 10^3 oocysts/m*l*). Lowering the temperature from 20°C to 5°C resulted in a drop in the effectiveness of ozone. Ransome et al (1993) achieved more than a 96% reduction in excystation of *Cryptosporidium parvum* oocysts when an ozone residual of a water containing 10^6 oocysts/*l* was maintained at 1 mg/L for 10 minutes It is obvious from these figures that the effectiveness of ozone as a disinfectant, especially for more resistant micro-organisms, such as parasitic cysts, is dependent on a number of factors, such as micro-organism concentration, temperature, pH, alkalinity, the concentration of natural organic matter in the water and the method of ozone application.

Apart from the improved disinfection efficiency of ozone over a number of other potential disinfectants, it also offers a number of other advantages in terms of wastewater disinfection. Ozone dissipates rapidly and leaves no residual, which is an advantage for wastewater treatment where long-lasting disinfectant residuals can be environmentally harmful to receiving water courses (Isaac, 1996). Ozone, being a powerful oxidant, provides added benefits in reducing odours, improving suspended solids removal, conditioning sludge, reducing colour, oxidising pesticides and herbicides and improving biodegrability of the wastewater (Masten and Davies, 1994).

There are also a number of disadvantages to using ozone. Ozone is fairly expensive to produce and must be generated on-site (Masten and Davies, 1994; Isaac, 1996). Furthermore, because of the lack of selectivity of the hydroxyl radical reaction, ozonation for wastewater treatment is usually confined to effluents which contain low concentrations of organic matter or other hydroxyl radical scavengers, such as carbonate and bicarbonate ions (Masten and Davies, 1994). Although a great deal of interest in ozone as a disinfectant was generated from concern over the formation of disinfection-by-products (DBP) produced when using chlorine, ozone also results in the formation of a number of DBP's, especially non-halogenated DBP, which include aldehydes, ketones and carboxylic acids (Flessinger et al, 1980; Miltner et al, 1992;

Krasner et al, 1993a; Meijers et al, 1993; Goel et al, 1995). Toxicological data on some of these ozone DBP, including formaldehyde, acetaldehyde, glyoxal and methylglyoxal, indicate that they are mutagenic or carcinogenic (Andrews and Huck, 1994) and are therefore a health concern.

Another disadvantage of using ozone that has come to light over the last decade is the formation of bromates when bromide is present in the water (Haag and Hoigne, 1983; Miltner et al, 1992; von Gunten and Hoigne, 1992; Glaze et al, 1993; Krasner et al, 1993b; Kruithof et al, 1993, Siddiqui and Amy, 1993; von Gunten and Hoigne, 1994). Bromate formation is proportional to the bromide concentration in the water and the ozone dose (Shukiary et al, 1994; Najm and Krasner, 1995) and is affected by pH and DOC concentrations. The formation of bromate decreases as the pH is lowered, because bromate is predominantly formed by reaction of ozone, hydroxyl radicals and hypobromite and as the pH decreases, so the amount of hypobromite declines (Kruithof et al, 1993; Symons et al, 1994; von Gunten and Hoigne, 1994). Najm and Krasner (1995) found that bromate concentration increased with increasing DOC concentration and this has implications for wastewaters containing bromide.

2.3. Advanced Oxidation Processes

As explained previously, ozone reacts either by direct oxidation by molecular ozone or by indirect oxidation by free radical species. The use of advanced oxidation techniques in which ozone is used in conjunction with ultraviolet (UV) irradiation or hydrogen peroxide (H_2O_2) enhances radical formation and can increase the removal of atrazine, NOM and other organic pollutants (McNight et al, 1993; Beltran et al, 1994a, 1994b; Koga et al, 1992; Meijers et al, 1993; Hulsey et al, 1993; von Gunten and Hoigne, 1994). The radical reaction is preferable for the destruction of organic molecules and it has been found that removal of pollutants such as the herbicide atrazine, which is fairly resistant to attack by ozone, increases when the radical reaction pathway is encouraged (Masten and Davies, 1994; Koga et al, 1992; Meijers et al, 1993; Hulsey et al, 1993). One would therefore expect an overall improvement in the effect of ozone or UV disinfection when used together or in conjunction with other oxidants such as hydrogen peroxide.

2.4. UV Irradiation

UV radiation forms part of the electromagnetic spectrum and in the wavelengths ranging from 240 to 280 nm is effective for the inactivation of micro-organisms by causing irreparable

damage to their nucleic acid (Lykins and Griese, 1986; Wolfe, 1991). The most severe damage to the nucleic acid of micro-organisms occurs at a wavelength of 260 nm and the damage incurred is proportional to the UV dose ($W.s/cm^2$), which is the product of the intensity of the UV radiation (W/cm^2) and the exposure time (seconds). The actual UV dose received by an organism depends on a number of factors, such as the flow rate of the water through the UV system, the transmission efficiency of the water being treated and the shape and design of the UV radiation chamber.

Two types of commercial UV systems are available, those that use low pressure mercury vapour lamps and those that use medium pressure mercury vapour lamps (Combs and McGuire, 1989). The low pressure lamps produce a narrow band of radiation almost exclusively at a wavelength of 253,7 nm, which is close to the maximum biocidal wavelength of 260 nm, but they only emit approximately 40% of the power input at that wavelength (Wolfe, 1991). The medium pressure lamps emit a much broader band of UV light, but their overall energy output is significantly higher than that of the low pressure lamps. The low pressure lamps are however considered by some to be the most efficient source of UV radiation for disinfection purposes (Wolfe, 1991).

UV disinfection works on the principle that at radiation 254 nm UV alters the nitrogenous heterocyclic components of DNA and RNA, causing the molecules to form new bonds resulting in dimers, which can disable the micro-organisms from replicating (Parrotta and Bekdash, 1998). The majority of viruses and bacteria require fairly low UV doses for inactivation, these being in the range of 2 to 6 $mW.s/cm^2$. Slade et al (1986) in their studies of UV disinfection of a ground water contaminated with chlorine-resistant enteroviruses found that a UV dose of 25 $mW.s/cm^2$ effectively removed these viruses from the water. Parasitic cysts on the other hand appear to be much more resistant to UV radiation than other micro-organisms (Rice and Hoff, 1981). In a study conducted by Ransome et al (1993) a UV dose of 80 $mW.s/cm^2$ was needed to reduce excystation of *Cryptosporidium parvum* by 90% and 120 $mW.s/cm^2$ was required for a 99% reduction in excystation. Rice and Hoff (1981) found that a UV dose of 63 $mW.s/cm^2$ reduced excystation of *Giardia lamblia* by 90%. In both cases, the UV doses required to reduce these parasites by only 90% far exceeds the minimum dose of 16 $mW.s/cm^2$ recommended by the US Public Health Service and the 30 $mW.s/cm^2$ generally used in practice (Ransome et al, 1993).

However, many factors affect UV disinfection efficiency, including the lamp sleeve, fouling of the sleeve, ageing of the lamp, the concentration of suspended solids and of micro-

organisms, the depth of the water being irradiated and the transmissivity and turbidity of the water (Qualls et al, 1985) and this needs to be taken into account when quoting UV doses. For example Moreno et al (1997) found in their studies of UV irradiation of wastewater effluent that the UV dose required to reduce the faecal coliforms in the water to the target concentration of 1 000 /100 m*l* increased from 27 mW.s/cm^2 when the effluent contained a low bacterial load to 32 mW.s/cm^2 at higher bacterial concentrations.

UV treatment processes are well documented and a number of references describing UV disinfection of secondary wastewater effluents prior to discharge to rivers or sea can be found (Chrtek and Popp, 1991; Morena et al, 1997; Kunzel, 1998a; Kunzel, 1998b). However, although UV has been found to be an effective disinfectant, an effluent of a reasonably good quality in terms of UV absorbance or transmissivity is required in order for this method of disinfection to be feasible, both economically and technically. Furthermore, despite the fact that there is no technical limitation on the size of a UV plant, more units being added in for larger plants, the costs tend to become prohibitive at larger facilities because of the high operating costs (Rudd and Hopkinson, 1989). In terms of by-product formation, UV irradiation would appear to be one of the least harmful of the disinfectants, although it may have the potential to cause an increase in mutagenic activity (Rudd and Hopkinson, 1989).

A disadvantage of using UV disinfection lies in the ability of micro-organisms to repair damage caused to their DNA by UV irradiation if sub-lethal doses are administered (Harm, 1980; Moseley, 1984). Reactivation mechanisms include the enzymatic processes of photoreactivation and dark repair, with the former being the faster process (Lindenauer and Darby, 1994; Shaban et al, 1997). Photoreactivation results in the uncoupling of DNA dimers produced by the initial exposure to UV irradiation and takes place in the presence of light in the wavelength range of 310 to 480 nm. Dark repair obviously occurs in the absence of light and results in the removal of the DNA dimers produced during UV irradiation (Shaban et al, 1997). Both mechanisms can result in the regrowth of bacteria if the UV irradiated water is stored for any length of time. UV disinfection also leaves no disinfectant residual and although this may be viewed as a disadvantage in potable water disinfection, it is in fact beneficial for wastewater disinfection where a residual could be harmful to the ecology of the receiving water.

2.5. Peracetic Acid

Peracetic acid disinfection has been used for some years now in various fields, but has only fairly recently been employed for wastewater disinfection (Baldry and French, 1989a) and consequently data regarding DBP formation and toxicological effects are rather scarce (Veschetti et al, 1998). The literature presently available does not however indicate any adverse effects to the environment due to peracetic acid (Baldry and French, 1989a and 1989b; Morris, 1993).

Studies in which the disinfection efficacy of sodium hypochlorite and peracetic acid were compared (Veschetti et al, 1998) indicated that there was little difference between these two disinfectants as far as total and faecal coliforms are concerned, but that peracetic acid was less effective than sodium hypochlorite in removing faecal streptococci. The peracetic acid dose required to reduce faecal streptococci to below the Italian legal limits was in the region of three times that of the hypochlorite concentration required using the same contact time.

No information regarding the effect of peracetic acid on parasitic cysts has been found, but one would assume from the data available that it is no better than chlorine disinfection in this regard. However it may be possible to use peracetic acid as a replacement for chlorine disinfection of wastewater effluents or to use it in conjunction with another method of wastewater disinfection.

2.6. Membrane Filtration

Recent concern over the presence of parasitic cysts such as *Cryptosporidium* and *Giardia* in potable water, even after conventional treatment including coagulation, flocculation, filtration and disinfection, has renewed interest in membrane filtration. Membrane filtration could offer an effective means of removing these small parasitic cysts from wastewater effluent, provided that suitable pre-treatment of the water is carried out prior to membrane filtration.

Pressure-driven membrane separation technology removes substances ranging in size from ionic to molecular and membranes have been developed with mass transfer properties and pore sizes that enable ionic, molecular and organic substances ranging from 1 to 1000 Å (molecular weight of between 100 and 500 000 Daltons) to be removed from water (Bisconer, 1998). The membranes are in fact classified based on the physical size or molecular mass of the substances which they filter out. Four basic membrane systems exist, namely

microfiltration (MF), ultrafiltration (UF), nanofiltration (NF) and reverse osmosis (RO) (Marcus and Pastrick, 1988; Maravich et al, 1989; Harfst, 1992; Harfst, 1995; McClellan, 1995). The mechanism of particle removal is dependent upon the characteristics of the membrane, including the pore size and charge, as well as the properties of the solution being filtered. For low pressure membranes separation mechanisms include both sieving (i.e. size exclusion) and adsorption to the membrane surface (Jacangelo et al, 1995). In general, RO removes most contaminants, including ions, humic acids, proteins, viruses, bacteria and parasitic cysts; NF allows single charged anions to pass through, but rejects those with more than one charge; UF removes only those particles with high molecular mass as well as viruses, bacteria and parasitic cysts; while MF rejects some viruses, most bacteria and parasitic cysts (Bisconer, 1998).

The performance of a membrane filtration system deteriorates due to deposition of the rejected material on the membrane surface, known as fouling, which increases the membrane resistance. According to Flemming and Schaule (1988) fouling is caused by:

1. Deposition of metal hydroxides or inorganic material which becomes concentrated above its solubility product (i.e. scaling).
2. Organic fouling caused by adsorption of organic material.
3. Biofouling caused by adhesion and growth of micro-organisms.

Fouling gradually reduces both the permeate flow and the lifetime of the membrane and to overcome this, the membrane needs to be regularly backwashed. In spite of this, the permeate flux continues to decrease due to irreversible fouling which cannot be removed by normal backwashing and eventually the membrane requires chemical cleaning. The cleaning interval can vary from weeks to months depending on the water quality and the operation procedure (Amjad, 1989; Amjad et al, 1991; Durham, 1991).

A schematic depicting the filtration spectrum appears in **Figure 2.1** (Membratek) and indicates that for the purposes of sewage, MF would be the most appropriate option. MF is the oldest of the pressure-driven membrane technologies and has a pore size ranging from 0,5 to 5 µm. On account of this relatively large pore size it can be operated under ultra-low pressure conditions (Jacangelo and Buckley, 1996). Filtration can be operated in two modes, namely direct filtration (i.e. dead-end) and cross-flow, with the choice of mode depending on the quality of the raw water. The dead-end mode is generally used for water with an average turbidity of less than 2 NTU and an average TOC lower than 2 mg/*l*. Operated in this mode there are only two defined flow streams present, the feed and the permeate. In the cross-flow

mode there are 3 streams, the feed, the permeate and the concentrate and the flow is parallel to the membrane. In cross-flow filtration the turbulence created during operation removes material which was carried to the membrane surface by convection. This process allows for continuous removal of material, thus limiting deposition at the membrane surface. Therefore the cross-flow filtration mode minimises fouling resulting in longer filtration runs than can be obtained using direct filtration (Maravich et al, 1989) and makes the cross-flow mode preferable for wastewater effluents.

Membrane filtration has been used in South Africa for wastewater reclamation. A 400 m^3/d plant was installed in Port Elizabeth to treat secondary wastewater effluent (Odendaal, 1991). The water produced by this plant was found to be of a high quality chemically, but a number of problems were encountered, including the fact that although there was removal of bacteria, post-disinfection was still necessary.

Other disadvantages to using membranes for wastewater disinfection include the excessive fouling that can occur when filtering wastewater effluents, which require a stringent cleaning regimen. Costs can also be high, especially due to power consumption used in running booster pumps, air compressors, mechanical strainers, washwater pumps etc. (Gere, 1997).

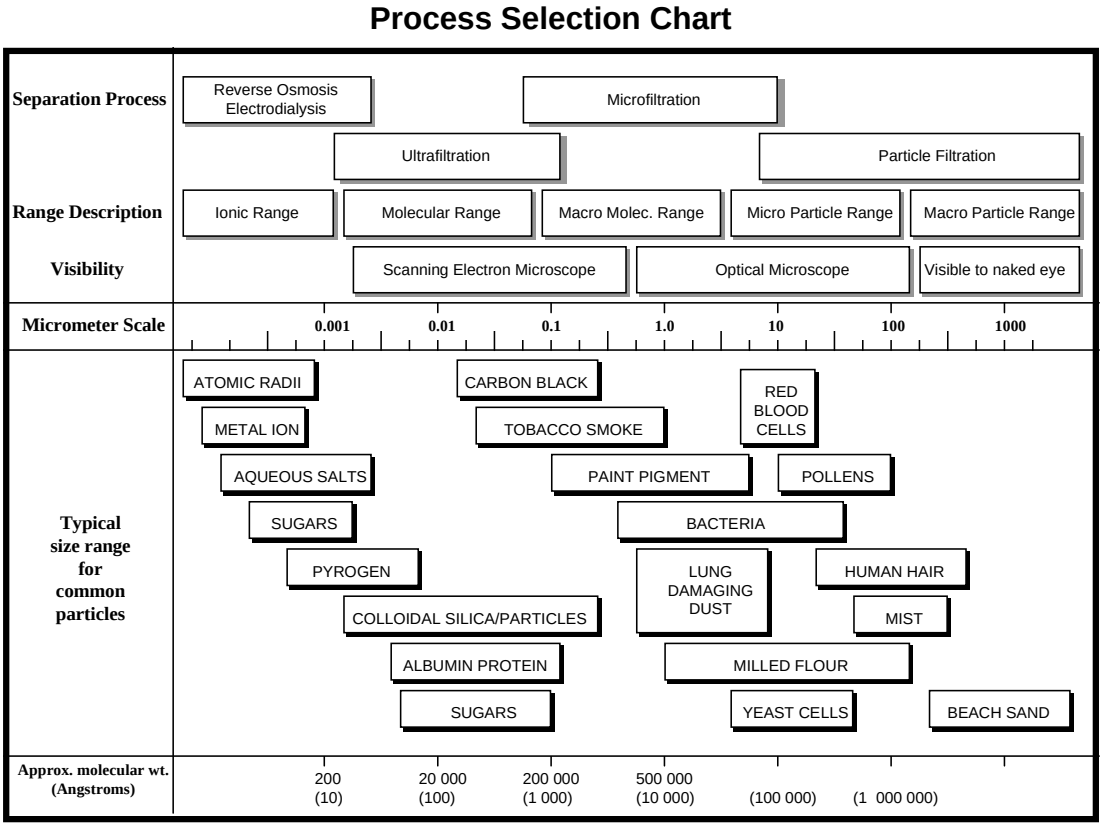

Figure 2.1: The filtration spectrum.

2.7. Bromine

Bromine disinfection has been used mainly for swimming pool treatment in the past, although there are a number of cases in which it has been used for wastewater disinfection. A bromine "*stick*" consisting of bromochlorodimethylhydantoin (DIHALO) was introduced in 1958 for swimming pool disinfection. Over the years a number of bromine based disinfectants have been developed, including polybromine anion exchange resins (White, 1992), but these were all directed at disinfection of small treatment units for household use and swimming pools. More recently some large utilities in the USA have started using bromine for disinfection (Brits et al, 1994), but this is still fairly rare. Some wastewater utilities use bromide together with chlorine. The chlorine oxidises the bromide to the active bromine.

The literature indicates some confusion regarding the disinfectant efficiency of bromine. Most of the investigations have in fact been carried out with sewage, where bromines were shown to be almost equal in efficiency to chlorine (measured as HOCl) (White, 1992). The disparity in the relative efficiency of bromine and chlorine was mainly due to the difference in consumption of the two disinfectants in order to satisfy the disinfectant "demand" of the water

(in equivalent mass concentration); since bromine is more reactive than chlorine, more bromine is consumed by the water resulting in a lower efficiency. In general chlorine is more reactive than bromine, but references to exceptions appear in the literature (Liptrot, 1987). The greater reactivity of bromine with natural organic matter in water can be attributed to the greater solubility of bromine in water when compared to chlorine. The fact that bromine reacts more quickly than chlorine does however allow for shorter contact times when using bromine. Keswick et al (1978) also demonstrated that bromine chloride was more effective in the inactivation of poliovirus than chlorine.

Bromine has also been found to form trihalomethanes more readily than chlorine, resulting in a shift from chloroform to bromoform (Lange and Kwaczynski, 1978). The implications for trihalomethane production when using bromine were amongst a number of other negative factors which have led to very little progress in the field of bromination for potable water disinfection. These include:

1. potential negative health aspects; although no data have been produced on the mutagenicity or carcinogenicity of bromine (Tate and Arnold, 1990), trihalomethanes are suspected carcinogens and bromine increases the potential for trihalomethane formation.

2. bromine imparts tastes and odours to most waters; this is attributed to its high reactivity with natural organic matter in the water.

3. the high reactivity of bromine also makes it very difficult to maintain disinfectant residuals when using bromine, although this can be a positive factor in terms of wastewater disinfection.

4. bromine is highly corrosive and in some cases it would be necessary to replace the existing chemical feed equipment (bromine chloride, although less corrosive than bromine, cannot be handled in PVC or ABS plastics, as can chlorine) (White, 1992).

5. bromine is hazardous to handle, although it is less hazardous than chlorine gas.

Despite some of the negative factors in using bromine for potable water disinfection, it has shown some promise for wastewater disinfection. Bromine species are better oxidising agents than their analagous chlorine species and organic compounds such as organic alcohols,

aldehydes, amines and mercaptans are completely oxidised by bromine chloride to form inorganic chloride and bromide salts as the main by-products (White, 1992). Also, the fact that bromine results in much lower residuals than equivalent doses of chlorine is a positive factor in trying to keep the disinfectant residual of effluent discharges low.

3. Laboratory and Pilot Plant Investigations

3.1. Introduction

The primary aim of this project was to assess alternative disinfectants for use in wastewater applications, with special emphasis on reducing the chlorine residual produced while not compromising disinfection efficacy. Another important aspect considered was finding a disinfectant capable of inactivating parasitic organisms, such as *Cryptosporidium* and *Giardia*. For this reason a fairly wide range of disinfectants was investigated, both used independently and in conjunction with other disinfectants, in the hope that combinations might result in a synergistic effect capable of destroying these resistant organisms. It was also considered essential that a disinfectant or disinfectants be identified that would be suitable for use in low-technology applications, since there is a great need for such a product in this country.

Both laboratory and pilot plant tests were conducted for this investigation. Because an extensive range of parameters can be tested under laboratory conditions, these tests allow for more comprehensive testing of a system than is usually possible at full scale or on pilot plants. However, scaling up from laboratory tests to full scale can result in fairly large margins of error and for this reason it was considered important to conduct pilot plant tests, since these allow for a more accurate simulation of the full scale plant conditions.

The wastewater effluent used in these tests came from the Darvill Wastewater Works (WWW) in Pietermaritzburg, a small city situated approximately 80 km inland of Durban on the Kwa-Zulu Natal coast of South Africa. The Darvill WWW treats a predominantly domestic wastewater effluent and has a nominal design capacity of 75 ML/day. The process consists of primary sedimentation, at times augmented using primary enhanced treatment with ferric salts, an activated sludge plant with aluminium sulphate and lime addition for phosphate removal, followed by secondary settling, chlorination and tertiary treatment through a series of maturation ponds. The effluent used in both the laboratory and pilot plant tests was drawn out of the overflow channel from the secondary sedimentation tanks at a point well above the chlorination stage. Water quality parameters of a typical secondary wastewater effluent sample appear in **Table 3.1**.

Table 3.1: Typical water quality data for a sample of secondary wastewater effluent from the Darvill Wastewater Works.

Determinand	**Units**	**Secondary wastewater effluent**
pH	-	7,44
Turbidity	NTU	1,83
Conductivity	mS/m	48,0
COD	mg/L O_2	27
Colour	° Hazen	8,53
UV abs. 254 nm	-	0,094
Soluble Reactive Phosphorus	µg/L	1310
Ammonia	mg/L N	2,15
Nitrates	mg/L N	3,8
Alkalinity	mg/L $CaCO_3$	90,3
Suspended Solids	mg/L	9,6
E. coli	CFU/100 mL	17 700
Coliforms	CFU/100 mL	49 500
Faecal streptococci	CFU/100 mL	3 650
Total colony counts at 37°C	CFU/ mL	11 575
Total colony counts at 22°C	CFU/ mL	13 075
Coliphages	PFU/ 10 mL	793
Cryptosporidium oocysts	org./10 L	4
Giardia cysts	org./10 L	235

The effluent used in the laboratory tests was often, but not always, spiked with cultures of *Cryptosporidium* oocysts and *Giardia* cysts, although the effluent sometimes contained these prior to spiking. Spiking ensured that oocyst and cyst counts were sufficiently high to ensure easy interpretation of the results. The pilot plant tests were however conducted on unspiked effluent, since spiking into such large volumes of wastewater would have been impractical and costly.

A variety of disinfectants and combinations of disinfectants were assessed both in the laboratory and at pilot plant scale, these being:

1. Chlorine
2. Ozone
3. Low pressure UV irradiation
4. Medium pressure UV irradiation
5. Multiwave UV irradiation
6. Peracetic acid
7. Hydrogen peroxide
8. Peracetic acid and hydrogen peroxide
9. Bromine
10. Klor-Free (only laboratory testing)
11. Mixed oxidant generator (only laboratory testing)
12. Ozone combinations:
    - ozone / chlorine
    - ozone / low pressure UV
    - ozone / medium pressure UV
    - ozone / peracetic acid
    - ozone / bromine
13. UV combinations:
    - UV / chlorine
    - UV / peracetic acid
    - UV / bromine
    - UV (Multiwave only) / hydrogen peroxide

A number of tests were conducted on the secondary effluent before and after being exposed to these various disinfectants. The microbial testing protocol included all or some of the following:

1. *E. coli*
2. Coliform organisms
3. Faecal streptococci
4. Total colony counts at 37 and 22°C
5. Coliphages
6. *Cryptosporidium*
7. *Giardia*

The following analyses were also conducted on many of the samples:

1. TOC
2. DOC
3. BDOC
4. THM formation potential (THMFP)
5. UV extinction (254 nm)
6. pH
7. Temperature
8. Turbidity
9. Conductivity
10. Alkalinity
11. Calcium, magnesium and total hardness
12. Iron
13. Manganese
14. Total dissolved solids
15. Suspended solids
16. COD

In all cases the chlorine demand of the water was determined and whenever ozone was used, the ozone requirement was also measured.

The tests carried out by Umgeni Water for the purposes of this investigation, determined *Eschericia coli* and not faecal coliforms. Essentially, there is little difference between *E. coli* and faecal coliform counts; *E. coli* are a subgroup of faecal coliforms and therefore feacal coliform counts are generally slightly higher than *E. coli* counts, but the test specific for *E. coli* is more accurate than that for faecal coliforms. In practice, the *E. coli* results can be considered representative of faecal coliforms and for the purposes of this report are used interchangeably.

3.2. Results and Discussion

3.2.1. Chlorine

Chlorine was used throughout this investigation to provide a baseline measurement against which the performance of the other disinfectants could be compared. The reason for this is that chlorine has been used extensively for wastewater effluent disinfection for the best part of the last century and as a result, a great deal of literature is available on the effectiveness of this disinfectant.

The chlorine demand tests which were carried out on the Darvill Wastewater secondary effluent for the purposes of this study, indicated that the chlorine demand could vary over a fairly wide range, from as low as 4 mg/L to over 20 mg/L, the average being 9 mg/L. The chlorine demand of the effluent was used to determine optimum disinfectant dosing rates of chlorine and some of the other disinfectants. Typical chlorine demand curves obtained during this project appear in **Figures 3.1** and **3.2.**

It was observed that chlorine, when dosed at the chlorine demand concentration, generally gave between a 2 and 3 log reduction in most of the bacterial indicator organisms after the 30 minute contact period, although it was less effective in removing coliphages. Disinfection using chlorine (sodium hypochlorite) was used as a standard against which the other disinfectants have been compared and therefore the results of the chlorine tests are not discussed under a separate section, but under the other sections where relevant.

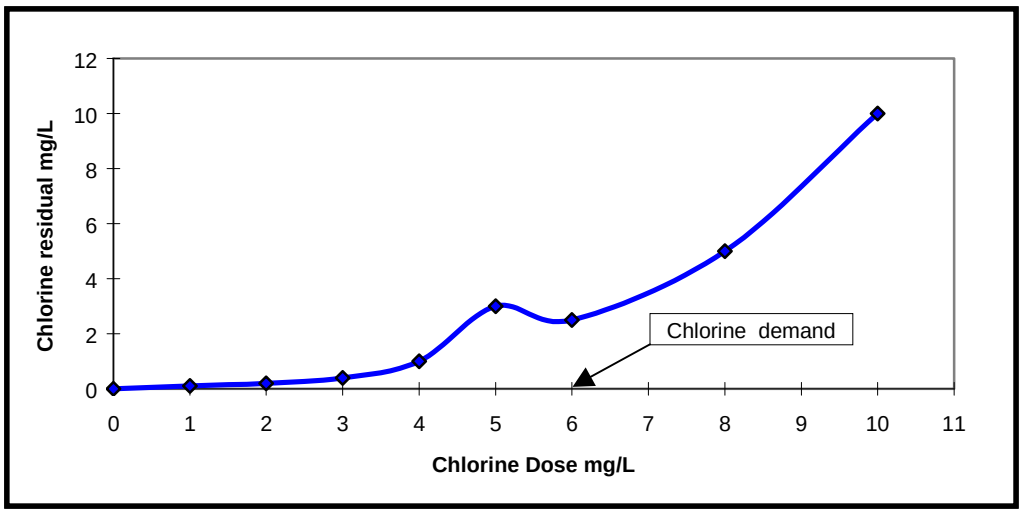

Figure 3.1: Typical chlorine demand curve of Darvill WWW secondary effluent (chlorine demand approximately 6 mg/L).

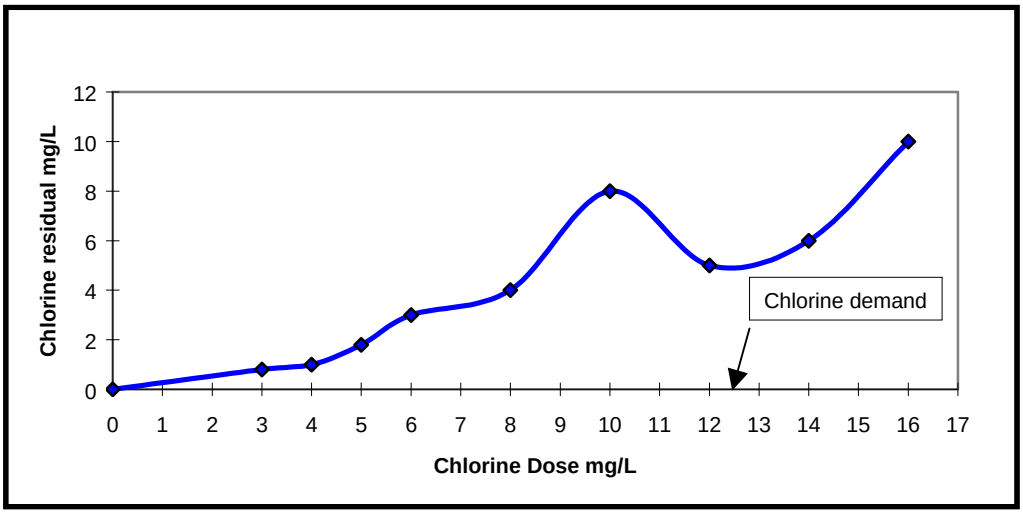

Figure 3.2: Typical chlorine demand curve of Darvill WWW secondary effluent (chlorine demand approximately 12,5 mg/L).

3.2.2. Ozone

Ozone, when used at the ozone requirement dose (see **Section 6.5.4**) gave a 2 to 3 log reduction in coliforms, *E. coli* and total colony counts at 37 and 22 °C, very similar in fact to that obtained using chlorine at its demand value. However, on average, the faecal streptococci organisms only underwent a 1 log reduction when ozone was dosed at the demand value, in contrast to an average reduction of 3 log in these organisms when dosing chlorine at its demand concentration. Another interesting factor that was observed in this study is that ozone is more effective in removing coliphages than chlorine when these disinfectants are dosed at their respective demand doses. Ozone often achieved complete removal of these organisms at the ozone demand dose of the effluent, whereas chlorine, when used at the chlorine demand concentration, usually did not result in complete removal.

Despite the similarities in the performance of ozone and chlorine when used at their respective demand concentrations, it was evident from the results that ozone is a more effective disinfectant than chlorine, achieving better removals of the indicator organisms at lower concentrations as is clearly demonstrated in **Figure 3.3**, which shows the effectiveness of ozone and chlorine in removing coliforms.

The average ozone demand or requirement determined from more than 24 samples of wastewater effluent, was 5 mg/L, and this was in the region of 55 to 60 of the chlorine demand. **Figure 3.4** graphically represents the chlorine and ozone demand results for a

number of effluent samples and clearly indicates that the ozone demand of the effluent is always significantly lower than the chlorine demand.

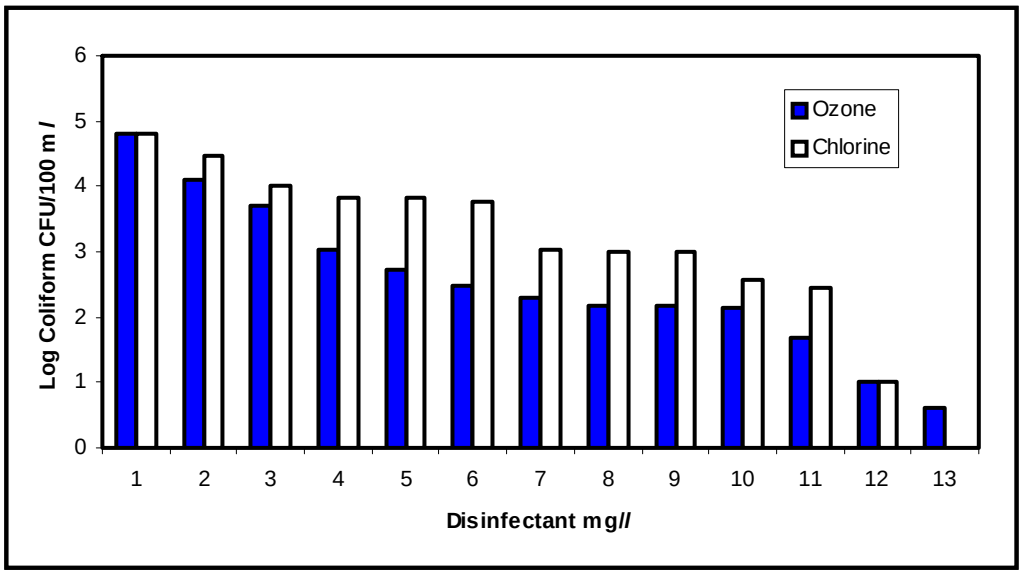

Figure 3.3: Viable coliform organisms (log coliform concentration CFU/100 mL) after dosing secondary clarified wastewater effluent with ozone and chlorine.

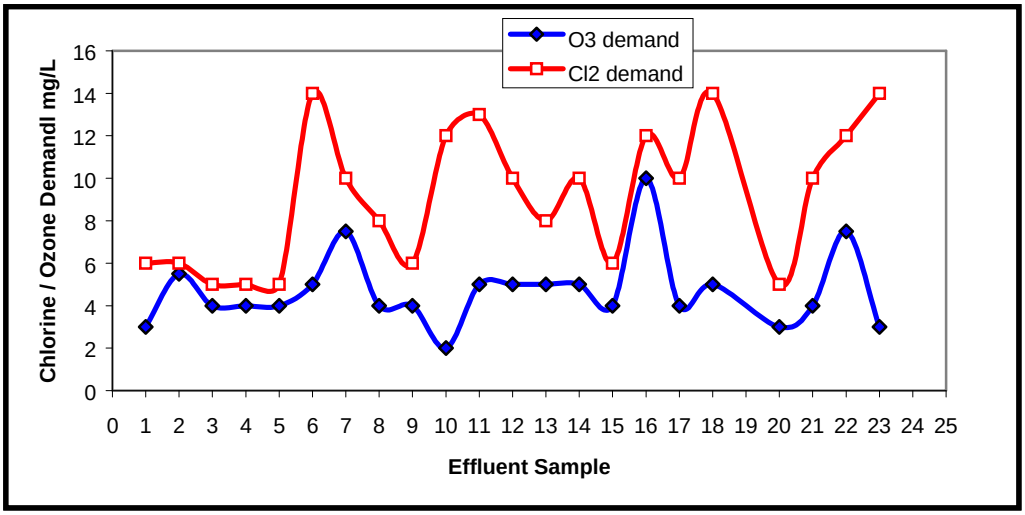

Figure 3.4: Chlorine and ozone demand values for different wastewater effluent samples.

An important variable for determining the disinfection efficiency of any disinfectant is the C_t factor and this is particularly widely used for ozone disinfection. The C_t factor was originally formulated by the United States Environmental Protection Agency (EPA), as part of the Surface Water Treatment Rule (SWRT) to establish the desired efficiency of the disinfection process (White, 1992). The C_t is the product of the disinfectant residual C in mg/L at the end

of the contact time, and the exposure time t in minutes, to this residual. The EPA recommended C_t for adequate disinfection using ozone is 1,6.

Calculating C_t for the laboratory tests was easier than for the pilot plant tests, since the laboratory tests were conducted in a semi-batch configuration. However, this did mean that as the applied ozone concentration was increased, so the contact time also increased. Obviously at the ozone requirement or demand concentration, the ozone residual was always fairly low, meaning that at this dose, the C_t was small, generally less than 0,2 (the average C_t for more than 25 effluent samples was 0,17 at the demand concentration). **Table 3.2** shows typical values for a number of effluent samples, giving the C_t at the demand concentration as well as the applied dose required to produce a C_t of approximately 1,6. In **Table 3.3** the log removals achieved for the various indicator organisms at the demand concentration as well as at a C_t of approximately 1,6 are given.

Table 3.2: Ozone demand/requirement value, residual and C_t at demand concentration, applied ozone concentration required for a C_t of approximately 1,6, residual at this applied ozone concentration and actual C_t at this concentration.

Sample	O_3 demand mg/L	Residual at demand mg/L	C_t at demand	Applied O_3 for C_t of 1,6	Residual at C_t of 1,6	Actual C_t
1	4	0	0	6	0,278	1,58
2	3	0,095	0,34	4	0,323	1,53
3	2	0	0	5	0,479	2,12
4	5	0,058	0,24	7,5	0,333	2,04
5	5	0	0	7,5	0,425	2,45
6	5	0,024	0,09	15,75	0,132	1,58
7	5	0,021	0,08	7,5	0,262	1,49
8	4	0,034	0,05	7,5	0,558	1,46

What is of interest is the fact that the improvement in disinfection efficiency at the higher C_t value, in terms of coliforms, *E. coli* and faecal streptococci, is not significantly different from that at the demand dose, although the concentration at the higher C_t value is generally much higher than the ozone demand. This implies that for inactivation of these organisms, the reactions of ozone are very fast and that increasing the dose beyond the demand concentration does not significantly increase the disinfection efficiency. It must also be remembered that the

reductions in indicator organisms are measured in terms of log values, and therefore a reduction of at least 10 fold is required before a significant difference is observed. With coliphages, the higher ozone dose did appear to have an additional effect.

Table 3.3: Log reductions in bacterial indicator organisms at the ozone demand value and at a C_t of approximately 1,6.

	Coliforms CFU/100 mL		***E. coli* CFU/100 mL**		**F. Streptococci CFU/100 mL**		**Coliphages CFU/100 mL**	
No.	**Demand**	**C_t = 1,6**	**Demand**	**C_t = 1,6**	**Demand**	**C_t = 1,6**	**Demand**	**C_t = 1,6**
1	2	2	1	1	1	1	1	>2
2	1	1	0	0	1	1	1	>2
3	1	1	1	2	1	2	1	1
4	2	2	2	2	0	1	2	>2
5	3	3	3	3	>3	3	>2	>2
6	4	4	5	5	-	-	>2	>2
7	1	1	2	2	1	1	>2	>2
8	3	2	3	2	1	2	-	-

Tracer tests had to be conducted on the pilot plant in order to determine the C_t. The time taken for the disinfectant to pass through the contact column was obviously dependent on the flow rate and the volume of effluent maintained in the ozone contact column, but under the conditions that the plant was typically operated, the contact time in the column varied between 20 and 40 seconds. A typical tracer test for the ozone contact column of the pilot plant is displayed in **Figure 3.5**. Unfortunately, due to problems experienced in achieving the required applied ozone concentrations, the C_t values obtained using the pilot plant ozone generator were in most cases well below the required C_t of 1,6.

Table 3.4 shows typical results for disinfection of a wastewater effluent with both ozone and chlorine. As the ozone concentration increases, a rapid decrease in the bacterial indicator organisms is observed until the ozone demand value is reached. At concentrations greater than the demand value, not much extra reduction occurs in the coliform, *E. coli* and faecal streptococci, although a reduction in coliphages does usually occur (usually by about 1 log).

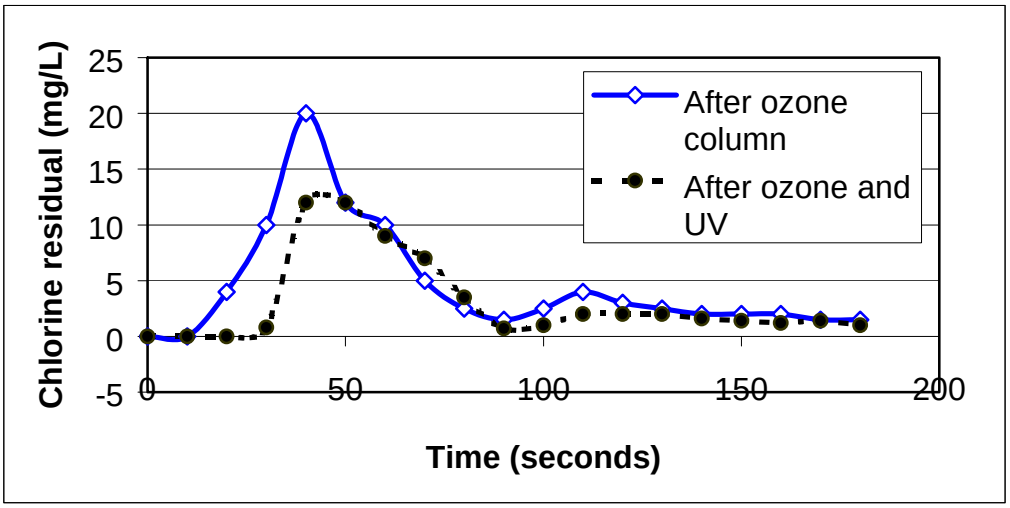

Figure 3.5: Results of a typical tracer test conducted on the ozone and medium pressure UV pilot plant using a chlorine spike. Spike added for 10 seconds, effluent flow rate 1,7 L/s and column volume 250 L.

It was found that in general an ozone concentration of between 1 and 2 mg/L was sufficient to reduce the *E. coli* count of the effluent to below the DWAF relaxed discharge limit of 500 CFU/100 mL. However, for removal of *E. coli* to less than 10 CFU/100 mL, much higher concentrations were required, usually between 6 and 8 mg/L and sometimes higher. This is obviously dependent on the ozone requirement of the effluent. In **Table 3.4** the ozone demand of the secondary wastewater effluent was 7,5 mg/L, which was higher than the average value and accounts for the fact the ozone doses required in order to reduce the *E. coli* to below the 500 CFU/100 mL and 10 CFU/100 mL levels were between 3 and 4 mg/L and >10 mg/L respectively.

A number of laboratory and pilot plant samples were analysed for *Cryptosporidium* and *Giardia*. The overall viability of these organisms was found to be approximately 77% and 60% for *Cryptosporidium* and *Giardia* respectively after disinfection compared to 88% and 75% in the effluent before, showing little reduction through the process. However, the disinfection process did appear to have a significant effect on reducing the number of parasites by some mechanism. The removal of *Cryptosporidium* that was obtained after ozonation was often good when the oocysts concentration was low (less than 50 oocysts/10 L) and the ozone concentration was at or near the ozone demand value of the effluent, but removals were less effective at higher oocyst concentrations (300 and more oocysts/10 L), not exceeding a 1 log reduction even at the ozone demand dose (see **Table 3.5**). The reduction in *Giardia* cysts after ozonation was even more erratic. In some cases complete removals were obtained even

when the cyst concentration was fairly high (>100 cysts/10 L), but in other instances little or no reduction was obtained, even at ozone doses close to the demand value. In comparison to chlorine when used at the chlorine demand dose of the effluent, there was in fact no significant difference in the reduction of parasitic cysts and oocysts achieved at the ozone demand dose. Using chlorine at the demand value on wastewater effluent would be impractical, but since the ozone requirement was usually around 5 mg/L, it would be feasible to dose at this concentration. One could therefore expect to obtain better removal of these parasitic cysts and oocysts when using ozone.

Table 3.4: Typical disinfection of wastewater effluent using chlorine and ozone at concentrations of up to 20 mg/L. The chlorine demand of the effluent sample used for this test was 10,5 mg/L and the ozone requirement was 7,5 mg/L.

Dose	*E. coli* CFU/100 mL		Coliforms CFU/100 mL		F. Streptococci CFU/100 mL		Coliphages PFU/10 mL	
mg/L	Ozone	Chlorine	Ozone	Chlorine	Ozone	Chlorine	Ozone	Chlorine
0	9 200	9 200	62 000	62 000	1 100	1 100	430	430
1	7 500		12 600		920		270	
2	2 700		4 900		760		30	
3	650	2 000	1 070	6 600	198	740	0	200
4	440	5 200	540	6 700	164	880	10	230
5	290	2 800	310	5 800	112	630	0	140
6		970		1 090		164		190
7,5	102		150		70		0	
8		720		990		178		160
10	90	26	140	360	40	58	0	30
12		16		290		46		130
14		0		10		40		20
15	0		4		2		0	
16				410		14		90
20	0		0		0		0	

Table 3.5: Effect of ozone and chlorine on *Cryptosporidium* oocysts and *Giardia* cysts.

***Cryptosporidium* Results**						
Effluent Oocysts/10 L	**O_3 Demand mg/L**	**O_3 Dose mg/L**	**Oocysts/10 L after O_3**	**Cl_2 demand mg/L**	**Cl_2 Dose mg/L**	**Oocysts/10 L after Cl_2**
10	10	10	0	12	12 10	0 0
45	4	4 2	0 0	10	10 8	0 0
10	4	5 3	0 0	14	14 12	5 0
30	3	3	0	12	5	7
325	7,5	7,5	65	12	12	105
390	3	3	330	14	14	10
330	3	3	440	14	-	-
50	2	2	84	9	9	144
***Giardia* Results**						
Effluent Cysts/10 L	**O_3 Demand mg/L**	**O_3 Dose mg/L**	**Cysts/10 L after O_3**	**Cl_2 demand mg/L**	**Cl_2 Dose mg/L**	**Cysts/10 L after Cl_2**
350	10	10 8	0 0	12	12 10	118 75
105	4	4 2	0 0	10	10 8	0 200
645	5	5 3	27 11	14	14 12	82 65
375	3	3 2	405 188	4	4 2	120 120
580	3	3	0	12	5	142
70	4	4	0	10	10	0
175	7,5	7,5	25	12	12	40
550	3	3	100	14	14	20
310	3	3	270	14	-	-
324	2	2	264	9	9	330
105	3	0,85	105	14	-	-
525	1	0,43	605	14	5	295

It is not clear whether this removal is due to physical means, disruption of the antigenic binding sites (although their presence was still apparent in the viability assays), or lysis of the (oo)cysts.

Pilot plant tests which were carried out to assess ozone, met with some difficulties, the most serious being the inability to apply ozone concentrations of much above 1 mg/L. The reason for this was partly due to the fact that the ozone generator, which had a design output of 30 g/h, when calibrated was found to have a maximum output of around 15 g/h under optimal conditions, although it was often not possible to obtain more than 12 or 13 g/h. Since it was also not possible to drop the flow rate of effluent through the pilot plant to much below 1,5 L*l*/s (i.e. 5,4 m^3/h), this limited the applied ozone dose to less than 2 mg/L in most cases. A typical calibration curve for the ozone generator appears in **Figure 3.6**.

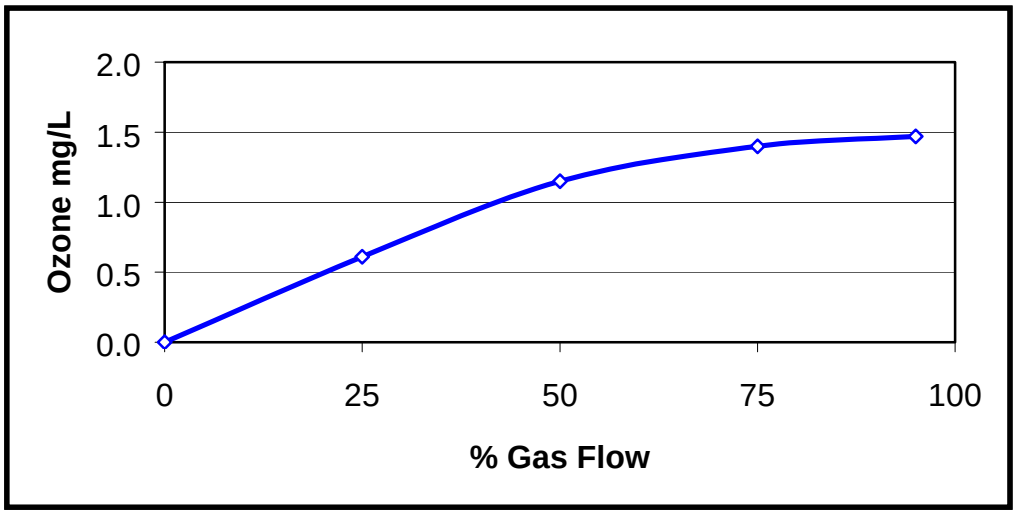

Figure 3.6: Calibration curve for pilot plant ozone generator operating on 2,1 A, an oxygen flow rate of 3 L/minute and an effluent flow rate of 2,38 L/s. Maximum ozone production approximately 1,5 mg/L.

Another factor that affected the pilot plant tests was variation in the quality of the effluent, which was not possible to control. Fluctuations in turbidity and organic contaminants would sometimes occur during experimental trials and despite rigorous analysis of the various water quality parameters, it was not possible to account for all of these variations. For example, the ozone demand was generally determined at the end of the trial, but in some cases this was an hour or more after the initial sampling for the trials had begun, but time did not permit the more frequent measurement of this parameter. In spite of this, results of tests conducted on the pilot plant confirmed those obtained in the laboratory, although it was never possible to

apply ozone doses equal to the ozone demand of the effluent when using the pilot plant. The ozone disinfection data obtained on the pilot plant for combinations of ozone with other disinfectants such as UV irradiation, chlorine and peracetic acid will be discussed more fully under the relevant sections.

From the chemical and physical analyses performed it was observed that ozonation sometimes resulted in a decrease in the turbidity, although no explanation has been found for this. It may be due to a reduction in the colour of the effluent after ozonation giving rise to an apparent reduction in turbidity.

The UV absorbance was found to decrease after ozonation, which is in agreement with the literature (Hoigne and Bader, 1983). Ozone breaks down the double bonds present in the organic matter content of the effluent and this in turn reduces the UV absorbance. The BDOC generally increased after ozonation, which is to be expected as ozone breaks down larger, more polar compounds into smaller, more biodegradable compounds (Janssens et al, 1984; Somiya et al, 1986; Sontheimer, 1979). The THMFP often increased slightly after ozonation, although not always and iron and manganese concentrations in the effluent generally decreased after ozonation, obviously due to the oxidation and subsequent precipitation of these metals. No other significant changes in the water quality were observed after ozonation. A typical set of water quality data for wastewater effluent both before and after ozonation at the demand concentration appears in **Table 3.6**.

Hypochlorite solution, which was used for chlorination of the wastewater, also impacted on some of these water quality parameters. The pH, alkalinity and the conductivity of the effluent generally increased slightly after sodium hypochlorite addition, obviously as a result of the high pH and salt content of the hypochlorite solution used. The only other significant effect of chlorine disinfection was the fairly significant increase in THMFP which occurred. These results indicated that ozone provides additional benefits in terms of effluent quality, other than those related purely to disinfection.

Table 3.6: Typical water quality data for a wastewater effluent before and after ozonation at the ozone demand concentration.

Determinant	Effluent before disinfection	Effluent after 7,5 mg/L O_3 (Ozone demand)
UV at 254 nm	0,106	0,042
pH	7,26	7,28
Temperature °C	19,7	19,6
Turbidity °Hazen	1,34	1,20
Coilforms CFU/100 mL	4 800	16
***E. coli* CFU/100 mL**	7 000	44
F. Strep. CFU/100 mL	2 100	26
Total counts 37°C CFU/ mL	1 230	350
Total counts 22°C CFU/ mL	6 700	3 600
***Cryptosporidium* oocysts/10 L**	325	65
Giardia cysts/10 L	175	25
TOC mg C/L	9,47	9,83
DOC mg C/L	9,57	9,85
BDOC mg/L	1,31	2,39
THMFP µg/L	388	152
Conductivity mS/m	48,2	47,5
Alkalinity mg/L $CaCO_3$	84,1	82,5
Total Hardness mg/L $CaCO_3$	90,0	92,9
Calcium mg/L	29	30
Magnesium mg/L	4,2	4,3
Iron mg/L	0,06	0,08
Manganese mg/L	0,08	0,08
Total Dissolved Solids mg/L	312	311
Suspended Solids mg/L	<4	<4
COD mg/L	<20	<20

3.2.3. Ultraviolet Irradiation

For the purposes of this investigation, both low pressure and medium pressure UV systems were used. Two low pressure UV systems were used; both were operated in a flow through mode on a small pilot plant and one was also used in batch mode in the laboratory. Two medium pressure systems were also tested, one a standard medium pressure unit and the other a Multiwave system. Both of these were tested on the pilot plant described in **Section 6.4.15**.

3.2.3.1. UV unit with lamp immersed in effluent

According to the literature, the majority of viruses and bacteria require fairly low UV doses for inactivation, these being in the range of 2 to 6 mW.s/cm^2, while parasitic cysts appear to be much more resistant to UV radiation than other micro-organisms (Rice and Hoff, 1981). Slade et al (1986) in their studies of UV disinfection of a ground water contaminated with chlorine-resistant enteroviruses found that a UV dose of 25 mW.s/cm^2 effectively removed these viruses from the water. In a study conducted by Ransome et al (1993) a UV dose of 80 mW.s/cm^2 was needed to reduce excystation of *Cryptosporidium parvum* by 90% and 120 mW.s/cm^2 was required for a 99% reduction in excystation. Rice and Hoff (1981) found that a UV dose of 63 mW.s/cm^2 reduced excystation of *Giardia lamblia* by 90%. In both cases, the UV doses required to reduce these parasites by only 90% far exceeds the minimum dose of 16 mW.s/cm^2 recommended by the US Public Health Service and the 30 mW.s/cm^2 generally used in practice (Ransome et al, 1993).

However, among the many factors affecting UV disinfection efficiency are the depth of the water being irradiated and the transmissivity and turbidity of the water (Qualls et al, 1985) and these need to be taken into account when quoting UV doses. For example Moreno et al (1997) found in their studies of UV irradiation of wastewater effluent that the UV dose required to reduce the faecal coliforms in the water to the target concentration of 1 000 /100 mL increased from 27 mW.s/cm^2 when the effluent contained a low bacterial load to 32 mW.s/cm^2 at higher bacterial concentrations. The UV doses used in the laboratory tests reported here were calculated based on an average transmittance of the effluent of approximately 70% (the transmittance was found to vary between 65 and 73%) and were between approximately 110 and 5 300 mW.s/cm^2 for UV irradiation exposure times of between 5 and 240 seconds, certainly much higher than the recommended minimum doses.

In calculating the UV doses applied to the effluent using this apparatus, some assumptions and approximations were necessary, since the unit did not possess a UV detector. The dose was

obtained by multiplying the power consumption of the lamp, according to the supplier's specifications (65 W), with the time that the effluent was exposed to the UV irradiation and then dividing this by the surface area of the UV contact chamber. For the laboratory batch tests, the exposure time was simply the period during which the effluent was subjected to UV irradiation. In the case of the flow through tests conducted at pilot plant scale, the exposure time was calculated from the chamber volume and flow rate. The dose was then corrected for the UV absorbance of the effluent since this is such an important parameter affecting the UV transmissivity.

The most common form of the combined Lambert-Beer law, often referred to as Beer's law is (Peters et al, 1974; Daniels and Alberty, 1975):

$$\log \frac{I_o}{I} = A = \varepsilon c l \qquad (1)$$

Where: ε is the molar adsorption coefficient
c is the concentration (mol)
l is the sample path length (cm)

The quantity $\log ({}^{I}_{0}/_{I})$ is referred to as the Absorbance, A, and as can be seen in Equation (1), A is proportional to l, the sample path length and since I_0 and I , the intensity of the incident and transmitted radiation appear as a ratio, and units of radiant intensity can be used.

The dimensions of the unit were measured and it was then assumed, based on the size of the contact chambers and the dimensions of the UV lamp, that average irradiation would occur at a point approximately two thirds of the distance between the surface of the lamp and the wall of the contact chamber. This distance was calculated to be close to 10 mm and so for the purposes of these calculations, a sample path length of 10 mm was used.

Also according to the Lambert-Beer:

$$A = \log \frac{100}{T}$$

Where: T is the transmittance (%).

Since the absorbance was measured in this study using a sample path length of 10 mm, one can assume that for an absorbance of 0 (i.e. a transmittance of 100%), the UV dose calculated

as described above would then be multiplied by 1, since theoretically all the UV irradiation passing through that samples would reach the sample which was 1 cm away from the UV source. In the same way, if the absorbance were for example 0,1, then the transmittance would be 80% and the calculated dose would have to be adjusted by a factor of 0,8 to account this.

In the laboratory tests conducted on this unit, most of the bacterial indicator organisms were found to be less susceptible to UV irradiation than to chlorine when considering the UV doses recommended in the literature. This was the case for *E. coli*, coliforms and faecal streptococci. UV doses of approximately 600 mW.s/cm^2 generally achieved only between a 1 and 2 log reduction in these organisms and UV doses of as high as 4 500 mW.s/cm^2 did not usually yield more than a 3 log reduction. This was in contrast to the effect that the UV irradiation had on coliphage organisms, where doses of less than 250 mW.s/cm^2 effected complete removal of these organisms. In comparison, chlorine concentrations significantly higher than the chlorine demand doses were not always effective in completely removing coliphages. A typical set of data obtained using this low pressure UV unit in batch mode is graphically represented in **Figure 3.7**.

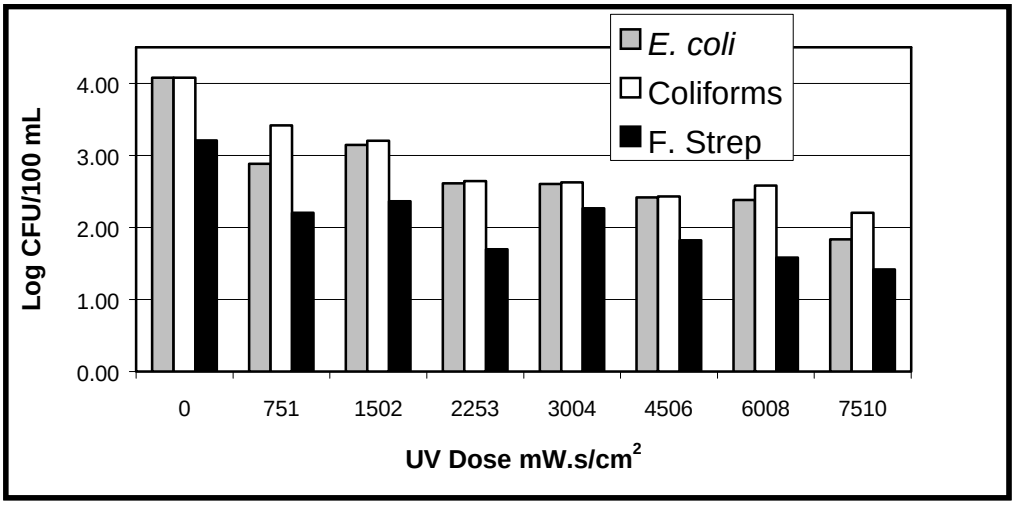

Figure 3.7: Typical set of data obtained with low pressure UV unit (lamp immersed in water) during laboratory batch tests.

In the flow through or pilot plant tests, this unit was operated at two flow rates, one at approximately 1 600 L/h and the other at 800 L/h and in an upflow mode and at a flow rate of 1 800 L/h in a downflow mode.

When operated in the upflow mode at a flow rate averaging 1 600 L/h over a test period of almost 2 months, reductions in bacterial indicator organisms were in the region of 2 to 3 log. The UV dose at this flow rate was calculated as being approximately 150 mW.s/cm^2 (average of 152 mW.s/cm^2, minimum of 127 mW.s/cm^2 and maximum of 186 mW.s/cm^2). Reducing the flow rate to 800 L/h effectively increased the UV dose to approximately 300 mW.s/cm^2 (average of 303 mW.s/cm^2, minimum of 253 mW.s/cm^2 and maximum of 370 mW.s/cm^2). At this reduced flow rate an improvement was noted in the coliform reduction which increased to over 3 log, but no significant difference in the removal of *E. coli* was observed. Unfortunately, due to analytical constraints it was not possible to analyse for all the other bacterial indicator organisms in the tests performed at 800 L/h, but since the removal of the other indicator organisms were similar to that of *E coli* in the previous tests (at 1 600 L/h), it one would expect their removal to have been much the same as that for *E coli* obtained in the tests at the lower flow rate. In all cases complete removal of coliphages was obtained.

The results imply that doubling the dose from 150 to 300 mW.s/cm^2 results in a relatively small improvement in disinfection. However, during the period that the higher flow rates were used, the turbidity of the effluent averaged at 2,56 NTU (the minimum value was 1,2 NTU and maximum value 4,75 NTU during this period), while the UV absorbance during the same period averaged 0,109 cm^{-1} (the minimum value being 0,025 cm^{-1} and the maximum 0,185 cm^{-1}). During the period that the lower flow rate was used the turbidity was far more variable, although the average was only 3,68 NTU (the minimum was 1,74 NTU and the maximum 7,91 NTU) and the UV absorbance was also fairly similar (average 0,092 cm^{-1}, minimum 0,049 cm^{-1} and maximum 0,128 cm^{-1}). It is therefore possible that the higher turbidity of the effluent during the lower flow tests may have caused a bias in the results.

The tests conducted with the unit operating in the downflow mode were very similar to those obtained in the upflow mode at the same flow rate and therefore at the same UV dose (i.e. 150 mW.s/cm^2). The turbidity and UV absorbance results during the two week period in which these tests were conducted were also similar to those for the tests performed in the upflow mode at a similar flow rate (i.e. an average turbidity of 1,89 NTU, a minimum value of 0,86 NTU and a maximum of 3,17 NTU and an average UV absorbance of 0,106 cm^{-1}, a minimum value of 0,03 cm^{-1} and a maximum of 0,153 cm^{-1}).

The presence of *Cryptosporidium* and *Giardia* were only detected on one occasion during these tests. The unit was being operated in the upflow mode at a flow rate of 1 350 L/h and the effluent prior to UV irradiation was found to have 40 *Cryptosporidium* per 6 L and

192 *Giardia* per 6 L. After irradiation no *Cryptosporidium* were detected and the *Giardia* concentration dropped to 80 *Giardia* per 6 L. The viability status of the oocysts and cysts was not determined.

It should be noted that better results were obtained with this unit when it was used in continuous mode as intended by the designers, rather than in the batch mode as was the case in these tests. The reason for this may be that due to the design of the UV contact chamber, pockets of effluent may have received less exposure to UV irradiation in batch mode.

3.2.3.2. UV unit with lamp positioned above effluent

This unit was operated at three different flow rates, approximately 800, 350 and 150 L/h. According to the manufacturer, the unit consumes 40 watts power and at a retention time of 12 seconds provides a UV dose of 80 mW.s/cm^2 for water with a transmissivity of 100%. The transmissivity of the effluent during this investigation was fairly constant at approximately 68%, which means that 800, 350 and 150 L/h correspond to UV doses of 61, 141 and 326 mW.s/cm^2 respectively.

Reducing the flow rate from around 800 to 350 L/h brought about a noticeable improvement in disinfection. At a flow rate in the region of 800 L/h, reductions of 2 to 3 log in coliforms, *E. coli* and faecal streptococci were obtained, while 2 log reductions in total counts occurred. By reducing the flow rate to a little under half this flow rate, i.e. to 350 L/h, the reductions in coliforms, *E. coli* and faecal streptococci were increased to between 3 and 4 log, while reductions in total count organisms were usually at least 3 log. However, when the flow rate was reduced still further to approximately 150 L/h, no noticeable improvement in bacterial cell removal was observed. This could be interpreted as indicating that above a certain UV dose no additional disinfection benefit occurs, but it was observed that at about the same time as the flow rate was reduced from around 350 L/h to 150 L/h, the turbidity of the effluent increased from an average of between 3 and 4 NTU, to an average of over 6 NTU. However, the UV absorbance or transmissivity of the effluent remained fairly constant through the entire investigation, averaging between 0,16 and 0,17 cm^{-1} at 254 nm for each period during which a different flow rate was used.

Coliphages proved to be very susceptible to UV and in all cases 100% removal of these organisms was obtained, even at the lowest UV dose used (i.e. 800 L/h or 61 mW.s/cm^2). Coliphage concentrations as high as 1 810 PFU/10 mL were completely removed at a flow rate of 846 L/h. Unfortunately there was no evidence of the presence of either

Cryptosporidium or *Giardia* during these tests, although analyses were conducted on a regular basis.

It is possible that a certain amount of shielding from the UV irradiation is provided by particles present in the effluent and that additional UV irradiation is then ineffectual or the effect greatly reduced. The unit accumulated particulate matter and had to be regularly cleaned in order to maintain effective operating conditions. These results indicate that some other method of disinfection may need to be used in conjunction with this low pressure UV unit in order to ensure adequate disinfection.

3.2.3.3. Medium Pressure UV

The medium pressure UV unit was only operated at pilot scale, since it was designed for flow rates of between 1,4 and 13 m^3/h, yielding applied UV doses of between 300 and 32 $mW.s/cm^2$ respectively. The unit contained a UV detector, so that once this had been connected to the flow meter measuring the effluent flow through the UV unit, the UV dose could be measured directly from the detector. The medium pressure UV unit was operated for a period of more than 3 months with measurements being taken twice daily in many cases. The UV dose administered was adjusted by altering the flow rate between 0,5 L/s (1,8 m^3/h) and 2,5 L/s (9 m^3/h) which correlated to UV doses of between approximately 280 and 30 $mW.s/cm^2$ respectively.

It was interesting to note that in terms of log removals of indicator organisms (see **Table 3.7**), the improvement between an average UV dose of 67 $mW.s/cm^2$ and 212 $mW.s/cm^2$ was not all that significant. However, the results did clearly show that at the higher UV doses complete removal of the indicator organisms was far more common than at the lower doses.

E. coli and faecal streptococci were more often removed completely than was the case for the total coliform organisms; in fact the coliform organisms were almost never completely removed. It needs to be pointed out though that the *E. coli* and faecal streptococci counts were generally lower than those of the coliform organisms. Also, in most cases where complete removal of *E. coli* and faecal streptococci were obtained, the counts present in the raw water were generally less than 10 000 CFU/100 mL, so that a 3 to 4 log reduction in cell counts was sufficient to reduce the counts to zero. UV doses of 150 to 200 $mW.s/cm^2$ were usually required in order to effect complete removal of *E. coli* from the effluent on a consistent basis, although in some cases UV doses of around 70 to 80 $mW.s/cm^2$ removed all of these

organisms, even though they had been in concentrations of more than 20 000 CFU/100 mL in the effluent prior to irradiation.

The faecal streptococci organisms were completely removed even more frequently than the *E. coli* organisms, which is interesting since these organisms are generally considered to be a little more resistant to disinfection than *E. coli* and coliform organisms. It is suspected that the reason for this may be due to the fact that the cell counts for the faecal streptococci organisms were in many cases lower than those for the *E. coli* organisms, sometimes by as much as a factor of ten, so that a 3 to 4 log removal could effect complete removal for the faecal streptococci, but not necessarily for the *E. coli* organisms.

Table 3.7: Average values for flow rate and UV dose of the medium pressure UV lamp, together with the average log removals obtained for the various indicator organisms (see Table 3.6 for typical bacterial indicator organism counts for the secondary effluent prior to UV disinfection).

Flow Rate L/s	Average UV Dose mWs/cm^2	Log Removal *E. coli*	Log Removal Coliforms	Log Removal F. Strep.	Log Removal TC 37 °C	Log Removal TC 22 °C	Log Removal Coliphage
0,5	212	3,1	3,2	3,3	1,6	1,5	>2
1,0	98	3,0	3,0	3,0	1,5	1,5	>2
1,5	67	2,8	2,8	2,7	1,2	1,4	>2
2,0	47	2,1	2,3	2,2	1,2	1,5	>2

Total count organisms account for a wide range of bacterial organisms and the removals of these when using medium pressure UV ranged between 1 and 3 log. Certainly the removals were lower than those for the other indicator organisms at equivalent doses.

Although not evident from **Table 3.7**, the coliphage organisms were, in almost every case, completely removed, even at UV doses of as low as 32 mW.s/cm^2. These organisms usually occurred in the wastewater effluent at concentrations of less than 1 000 PFU/10 mL, although on a number of occasions this value was exceeded, but even when present in fairly high concentrations, low UV doses were sufficient to completely eradicate these organisms. This is in agreement with the findings of the low pressure UV tests, although significantly higher UV doses had been used in the low pressure tests compared with these tests.

Despite regular analyses for *Cryptosporidium* oocysts and *Giardia* cysts during the 6 month period that the medium pressure UV tests were conducted, *Cryptosporidium* was never detected and *Giardia* was found in only a few of the secondary wastewater effluent samples. It was not possible to spike the effluent flowing through the pilot plant with parasitic cysts and it was also not possible to conduct laboratory scale tests using the medium pressure unit, and therefore it was necessary to rely upon the natural occurrence of these organisms in the secondary wastewater effluent. The *Giardia* cysts were only found in 5 of the effluent samples and then only in fairly low numbers (<30 cysts/10 L). After UV irradiation at doses of between 90 and 284 mW.s/cm^2, the cysts were either completely removed or reduced to below 5 cysts/10 L. There is presently no explanation for the removal of these organisms, but the viability of the cysts detected in the effluent prior to irradiation averaged 94%, while that of the cysts present after irradiation was 23%. **Table 3.8** shows a set of typical results obtained for wastewater effluent before and after irradiation at a dose of 212 mW.s/cm^2 using a medium pressure UV lamp.

In comparison to the disinfection efficiency that one could expect when using chlorine for wastewater treatment, the results obtained with the medium pressure UV lamp were very good. Even at the chlorine demand of the effluent, a reduction in the bacterial indicator organisms of 2 to 3 log was obtained, whereas the same reductions could be achieved at UV doses of less than 50 mW.s/cm^2 when using medium pressure UV. Increasing the UV dose to over 200 mW.s/cm^2 increased the reductions of these organisms to between 3 and 4 log and even as high as 5 log in some cases. Chlorine was also found to be far less effective against coliphage organisms than UV irradiation. It must be remembered that the chlorine demand of the wastewater effluent used in this study was fairly high, averaging at about 11 to 12 mg/L (ranging between 4 and 22 mg/L), so in most cases it would not be practical to dose the effluent at the demand value. However, these tests were conducted using a contact time of 30 minutes, but with wastewater a significant proportion of the chlorine added can be transformed into chloramines and these require a far longer contact times than free chlorine in order to achieve suitable disinfection. The chlorine dose on the wastewater plant where this investigation was conducted is generally between 3,5 and 4,5 mg/L and this dose fairly consistently reduces the *E. coli* levels of the effluent to below 500 CFU/100 mL (the DWAF relaxed standard for effluent discharge), but seldom results in complete removal.

Table 3.8: Typical results for secondary wastewater effluent before and after UV irradiation at a dose of 168 mW.s/cm^2.

Determinand	Effluent before UV	Effluent after UV
UV Abs 254 nm	0,119	0,109
pH	7,46	7,28
Temperature °C	20,5	22,6
Turbidity NTU	1,64	1,73
***E. coli* CFU/100 mL**	27 000	8
Coliforms CFU/100 mL	57 000	12
F. Streptococci CFU/100 mL	5 200	2
Total counts at 37°C CFU/ mL	2 400	70
Total counts at 22°C CFU/ mL	5 600	140
Coliphages PFU/10 mL	1 270	0

During the three month period that the medium pressure UV system was assessed, the *E. coli* counts of the secondary wastewater effluent varied between 900 and 174 000 CFU/100 mL, with the average concentration being 31 314 CFU/100 mL. The flow rate through the unit was varied between 0,5 and 2,5 L/s (1,8 to 9 m^3/h) to yield UV doses of between 30 and 280 mW.s/cm^2 and a total of 133 results were obtained. For 98% of these, the *E. coli* counts after UV treatment met the DWAF special standard of 500 CFU/100 mL and those that failed this standard were all obtained using UV doses of under 100 mW.s/cm^2. In 55% of these results, the *E. coli* counts were reduced below 20 CFU/100 mL and in 88% they were reduced below 100 CFU/100 mL after irradiation. More importantly, when using the lowest flow rate (0,5 L/s) at which the UV dose never dropped below 150 mW.s/cm^2, a 100% compliance with the DWAF 500 *E. coli* CFU/100 mL limit was achieved. In fact, of the effluent samples treated at this flow rate, in 84% of them the *E. coli* counts were reduced to below 20 CFU/100 mL.

One of the disadvantages of using UV irradiation for disinfection of wastewater effluents is that its success is dependent upon a good quality secondary effluent, which must be relatively low in both turbidity and UV absorbance. The Hanovia medium pressure unit used in these tests could be used, according to the supplier (and confirmed in our tests) at UV intensities of 60% and higher. When the UV intensity dropped below 60% it was usually due to fouling of the lamp or to a poor quality effluent and UV disinfection had to be discontinued until the UV intensity could be improved. During these pilot plant trials, some problems were experienced

on the full scale wastewater works, which resulted in significant carry over of activated sludge from the secondary clarifiers. During these periods it was impossible to conduct any test work, since the UV intensity sometimes dropped as low as 20%. Therefore if one is considering installing UV at wastewater works, one needs to ensure that a fairly good quality effluent is achievable. In this investigation UV disinfection was used successfully on effluent with turbidities as high as 4 NTU, but in all cases the UV intensity, as measured by the UV intensity meter on the instrument, was above 60%.

Regrowth tests were conducted on effluent which had been subjected to medium pressure UV irradiation at different doses. Three sets of tests were conducted at each of three different flow rates. The UV doses for the tests carried out at a flow rate of approximately 2 L/s, varied between 57 and 67 mW.s/cm^2 and the chlorine demand of the effluent prior to irradiation ranged between 8 and 22 mg/L. The UV doses administered in the tests conducted at flow rates of around 1,5 L/s were between 68 and 92 mW.s/cm^2 and the chlorine demand of the effluent during these tests varied between 12 and 16 mg/L. The regrowth tests using the UV doses were obtained at flow rates of approximately 0,5 L/s giving UV doses of between 173 and 285 mW.s/cm^2. These tests were conducted on effluent with a chlorine demand of between 10 and 18 mg/L. Assessment of the regrowth in effluent irradiated with UV under these conditions were carried out on treated effluent samples which had been stored for periods of up to three weeks. The effect of light was also determined by storing the irradiated effluent in both the light and in the dark.

The results of these regrowth tests were not really conclusive. No significant and consistent differences in the regrowth rate were observed to indicate that certain conditions should be avoided or encouraged to prevent regrowth after UV irradiation. There was in many cases better and more rapid growth in the irradiated samples, which had been exposed to light after UV disinfection, but this was not consistent enough to draw definite conclusions. Any regrowth which occurred usually ceased within 10 days and the total count organisms at 37 and 22°C indicated far more regrowth than for coliforms, *E. coli* and faecal streptococci. In fact very little regrowth of faecal streptococci organisms was observed and no coliphage regrowth was ever observed. Higher UV doses did appear to have a small positive effect in restricting the regrowth of *E. coli* and faecal streptococci, but again, the effect was not great. It was observed that in many of the tests two regrowth peaks occurred. This is possibly due to organisms metabolising different substrates, possibly the first peak being due to metabolism of an easily utilised substrate and the second for a substrate requiring induced enzymes before it can be metabolised. The results indicate that storage in the dark may be preferable to storage

in the light, although again, the results were not conclusive and in some tests more regrowth was actually found to occur in the samples which had been stored in the dark.

Medium pressure UV did not appear to impact on the water in terms of water quality parameters such as TOC, DOC, BDOC, THMFP, UV absorbance and turbidity.

3.2.3.4. Multiwave UV

The Berson Multiwave UV unit used in this investigation was fitted with a UV sensor, but it was not possible to link this signal to the flow meter in order to obtain a direct measurement of the UV dose from the instrument, as was the case with the Hanovia medium pressure UV unit. Instead, the suppliers provided a calibration sheet which allowed one to determine the flow rate required at various UV intensities in order to produce a UV dose of 25 mW.s/cm^2. For example, if the UV intensity was 94%, then according to the calibration table, a flow rate of 5 L/s would result in a UV dose of 25 mW.s/cm^2. If the actual flow rate was in fact only 1 L/s and not 5 L/s, then the UV dose would be five times greater (i.e. 5 x 25 = 125 mW.s/cm^2).

The results obtained with this unit were similar to those obtained with the Hanovia medium pressure unit. It was possible to achieve higher UV doses using this unit than had been the case with the Hanovia unit, but when comparing the disinfection efficiency achieved at corresponding UV doses, the log removals obtained were similar (see **Table 3.9**). The results do indicate that the Hanovia medium pressure unit was better than the Multiwave unit in complying with the *E. coli* limits of 500, 100 and 20 CFU/100 mL used in the table, but the Multiwave data is based on a far smaller sample number (generally 5 per UV dose) compared to the medium pressure lamp (30 per UV dose), so one non-compliance appeared to have a greater impact.

Tests were conducted at UV doses varying between approximately 150 and 475 mW.s/cm^2, on secondary clarified wastewater effluent in which the chlorine demand ranged between 12 and 22 mg/L (average 17 mg/L). In 93% of the irradiated samples, the *E. coli* counts were reduced to below the DWAF relaxation of the discharge standard of 500 CFU/100 mL, in 64% of the samples the *E. coli* levels dropped to below 100 CFU/100 mL after UV irradiation and in 29% this was below 20 CFU/100 mL. When using a UV dose of around 200 mW.s/cm^2 and higher, 100% compliance with the *E/ coli* limit of 500 CFU/100 mL was achieved.

The Multiwave UV tests, like the Hanovia medium pressure tests, could only be conducted at pilot plant scale, making spiking of the effluent with parasitic cysts and oocysts impractical.

Unfortunately, despite regular monitoring for these organisms, they were not found during the 5 week period in which these tests were carried out. One would expect the Multiwave to be very similar to the Hanovia medium pressure UV in terms of disinfection of parasitic organisms since both use medium pressure UV irradiation and were found to perform similarly in the disinfection of the bacterial indicator organisms.

In order to directly compare the disinfection efficiency of the Multiwave and medium pressure lamps, tests were conducted simultaneously on the pilot plant using both systems. The results appear in **Table 3.10** and again indicate that the medium pressure unit appears to give slightly better disinfection than the Multiwave unit at equivalent UV doses. This could be due to less accurate determination of UV dose when using the Multiwave unit, since the UV dose had to be calculated from a calibration curve when using the Multiwave system.

Table 3.9: Average values for flow rate and UV dose for both the Berson Multiwave and Hanovia medium pressure UV units together with average log removals of indicator organisms and the percentage compliance with *E. coli* limits of 500, 100 and 20 CFU/100 mL.

Unit	Flow L/s	Dose mW.s/cm^2	Log Reduction					Compliance with *E. coli* limits		
			E. coli	Colif.	F. Strep	TC 37°C	TC 22°C	% <500	% <100	% <20
Multi	0,5	475	3,4	3,3	3,2	2,2	2,2	100	100	80
Multi	1,1	216	3,2	2,8	3,0	2,0	2,0	100	80	20
Med.	0,5	212	3,1	3,2	3,3	1,6	1,5	100	97	93
Multi	1,5	158	2,0	2,2	2,2	1,6	1,8	80	20	0
Med	1,0	98	3,0	3,0	3,0	1,5	1,5	93	93	60

Table 3.10: Comparison of Multiwave and medium pressure UV units in treating wastewater effluent at different flow rates and UV doses (flow rate in L/s, UV dose in mW.s/cm^2 and bacterial indicator organisms in CFU/100 mL).

	Before UV	Multiwave UV				Medium Pressure UV			
Flow (L/s)	-	0,3	0,67	1,11	1,6	0,61	1,0	1,5	2,5
UV Dose	0	790	354	214	148	188	114	76	46
Trans.(%)	80,9	81,1	79,9	79,1	78,0	82,4	80,8	81,7	80,6
E. coli	14 000	4	0	4	84	0	0	2	52
Coliforms	33 000	8	20	36	370	0	50	26	340
F. Strep	5 000	4	8	10	32	0	2	6	8
CC 37°C	9 200	234	176	900	1 100	800	10	800	380
CC 22°C	14 300	376	496	1 000	<100	<100	8	<100	624

Regrowth tests were carried out on effluent treated with the Multiwave unit. The results of these tests were very similar to those obtained with the medium pressure system and also proved to be largely inconclusive. These tests were carried out at three different effluent flow rates or UV doses, namely 0,5 L/s (475 mW.s/cm^2), 1,0 L/s (237 mW.s/cm^2) and 1,5 L/s (158 mW.s/cm^2) on effluent samples which had chlorine demand values of between 14 and 20 mg/L. Regrowth of most of the indicator organisms occurred in almost all cases, except for faecal streptococci organisms. Regrowth of coliphage organisms was not tested since in all

cases the Multiwave unit completely removed these and the medium pressure regrowth tests had conclusively shown that no coliphage regrowth occurred.

As was found with the medium pressure regrowth tests, regrowth had usually stopped after about 10 days and often within 7 days of irradiation and two peaks were often evident. A typical regrowth curve appears in **Figure 3.8**. As was found in the medium pressure regrowth tests, no significant differences in regrowth were observed for effluent exposed to light or kept in the dark after irradiation.

Multiwave UV irradiation had no obvious effect on any of the other water quality parameters which were monitored in this investigation, such as turbidity, pH, TOC, DOC, BDOC, THMFP and UV absorbance.

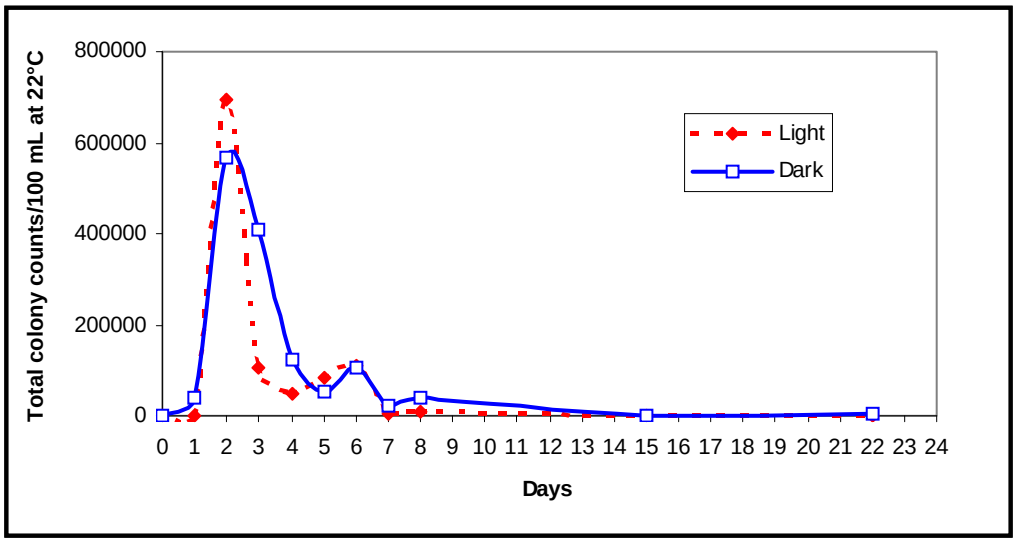

Figure 3.8: A typical regrowth curve of total colony counts at 22°C obtained after UV irradiation of secondary wastewater effluent (chlorine demand 18 mg/L) using the Multiwave UV system at a flow of 1,0 L/s (237 mW.s/cm^2).

3.2.4. Peracetic Acid and Peracetic Acid / Hydrogen Peroxide

Peracetic acid proved to be a particularly effective disinfectant. Tests were conducted at concentrations of up to 60 mg/L, but even at much lower concentrations, peracetic acid was found to effect good disinfection. In many cases the disinfection efficiency achieved using peracetic acid was comparable to that achieved using chlorine (sodium hypochlorite) at equivalent mass concentrations to chlorine.

During the period that these tests were conducted, the chlorine demand of the effluent varied between 2 and 18 mg/L, while the peracetic acid demand was found to vary similarly. Reductions in *E. coli* and coliforms of between 3 and 4 log and even complete removals were obtained using peracetic acid at 5 mg/L, which was as effective as the disinfection obtained using 5 mg/L chlorine. This is in agreement with the findings of Veschetti and co-workers (1998), who concluded from their studies that peracetic acid and sodium hypochlorite have similar bactericide efficiency against *faecal* and total coliforms. However, they also concluded that peracetic acid is less effective than hypochlorite for the removal of faecal streptococci, while in this study peracetic acid was found to be at least as effective as chlorine in removing these organisms and in many cases slightly better.

Table 3.11: Log removals in *E. coli*, coliforms, faecal streptococci and coliphages using 5 mg/L of either chlorine or peracetic acid (PAA) for disinfection of different secondary wastewater effluent samples.

Effluent		*E. coli*	Coliforms	F. Strep	Coliphages
1	**Chlorine**	4	3	2	>3
	PAA	3	2	3	>2
2	**Chlorine**	4	3	>2	>2
	PAA	3	2	>2	>2
3	**Chlorine**	4	3	>3	>2
	PAA	3	3	3	1
4	**Chlorine**	>3	3	>2	-
	PAA	3	4	1	-
5	**Chlorine**	>3	>3	>2	>1
	PAA	>3	3	>2	1
6	**Chlorine**	>4	4	>3	-
	PAA	>4	3	>3	-
7	**Chlorine**	4	3	>3	>2
	PAA	>4	>4	>3	>2
8	**Chlorine**	0	0	1	0
	PAA	3	3	2	1
9	**Chlorine**	1	1	>1	2
	PAA	2	2	3	2

However, peracetic acid was not quite as effective as chlorine in reducing coliphages and total plate count organisms. **Table 3.11** shows the log removals in various indicator organisms

obtained for chlorine and peracetic acid for various effluent samples. The slightly less effective reduction of coliphages when using peracetic acid is not really evident from **Table 3.11**, but when used at concentrations higher than 5 mg/L, the improved removal obtained with chlorine can be seen. More interesting is the fact that of the 9 trials conducted in which chlorine and peracetic acid were compared in terms of disinfection of wastewater effluent at a concentration of 5 mg/L, 100% compliance with the DWAF relaxation of the effluent discharge standard of 500 faecal coliforms CFU/100 mL was obtained using peracetic acid compared to 89% compliance with chlorine. In fact, in all cases the *E. coli* count in the effluent disinfected with peracetic acid was below 100 CFU/100 mL compared to 78% of the chlorine-disinfected effluents being below 100 *E. coli* CFU/100 mL. Reduction of the *E. coli* to below 20 CFU/100 mL occurred in 67% and 78% of the trials for peracetic acid and chlorine respectively. Based on these results, the disinfection efficacy of peracetic acid is certainly comparable to that of chlorine.

The disinfection efficiency of peracetic acid in terms of parasitic organisms was not quite as good as that achieved using chlorine at equivalent mass concentrations. Much of the test work in which *Cryptosporidium* and *Giardia* were determined was carried out using peracetic acid at equivalent concentrations to the chlorine demand value. In practice one would not use chlorine at the demand value for disinfection of wastewater, but since chlorine residual limits are not a factor when using peracetic acid, these limitations do not apply to peracetic acid. The *Cryptosporidium* concentration in these tests was in many cases fairly low (<50 oocysts/10 L) and so complete removal was generally achieved using both chlorine and peracetic acid at concentrations equivalent to the chlorine demand. Many of the results were obtained from pilot plant experiments and the effluent usually contained higher concentrations of *Giardia* than *Cryptosporidium*, resulting in there being more *Giardia* results than *Cryptosporidium* results for peracetic acid. The removal of *Giardia* cysts obtained at peracetic acid concentrations equivalent to the chlorine demand of the effluent averaged 43% compared to an average of 80% removal of these organisms when using chlorine at the demand value. Some of the results obtained for these organisms using peracetic acid appear in **Table 3.12**.

Table 3.12: Effect of peracetic acid and chlorine on *Cryptosporidium* oocysts and *Giardia* cysts.

***Cryptosporidium* Results**					
Effluent Oocysts/10 L	**Cl_2 demand mg/L**	**Peracetic acid dose mg/*k***	**Oocysts/10 L after peracetic**	**Cl_2 Dose mg/L**	**Oocysts/10 L after Cl_2**
10	12	12	0	12	0
		10	0	10	0
45	10	10	0	10	0
		8	0	8	0
325	12	12	180	12	105
390	14	14	140	14	10
***Giardia* Results**					
Effluent Cysts/10 L	**Cl_2 demand mg/L**	**Peracetic acid dose mg/*k***	**Oocysts/10 L after peracetic**	**Cl_2 Dose mg/L**	**Oocysts/10 L after Cl_2**
350	12	12	140	12	118
		10	290	10	75
105	10	10	5	10	0
		8	305	8	200
375	4	4	175	4	120
		2	150	2	120
175	12	12	190	12	40
550	14	14	310	14	20
600	7	5	580	5	210
525	14	5	415	5	295

Using hydrogen peroxide in conjunction with peracetic acid was expected to produce a synergistic effect. Peracetic acid is provided in solution form and the product used in these tests was supplied as approximately 39% peracetic acid in acetic acid, but it can also be obtained in hydrogen peroxide solution (Veschetti at al, 1998). These tests, in which a mixture of the two was produced using a solution of peracetic acid in acetic acid and a solution of hydrogen peroxide, clearly indicated that the hydrogen peroxide had a detrimental effect on the disinfection efficiency of peracetic acid. This is graphically represented in **Figure 3.9**, which shows the effect of peracetic acid and the mixture of peracetic acid and hydrogen peroxide on coliphage organisms.

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Tests were also conducted to assess the effect of hydrogen peroxide alone for wastewater disinfection, but it was found to be quite inadequate in comparison to chlorine. Reductions in the indicator organisms were low, even at high concentrations. For example, it was often not possible to reduce the *E. coli* counts to below 500 CFU/100 mL, even at applied hydrogen peroxide doses of more than 100 mg/L. The hydrogen peroxide demand was of the secondary wastewater effluent was generally between about 60 and 300 mg/L for the wastewater effluent.

In these tests it was found that where chlorine at a concentration of 5 mg/L produced a 75% compliance with the relaxation of the effluent discharge standard of 500 *E. coli* CFU/100 mL, hydrogen peroxide dosed at concentrations of 40 mg/L would have resulted in only about 25% compliance with the standard. Based on these results, hydrogen peroxide was not considered as a contender for replacing chlorine for wastewater disinfection.

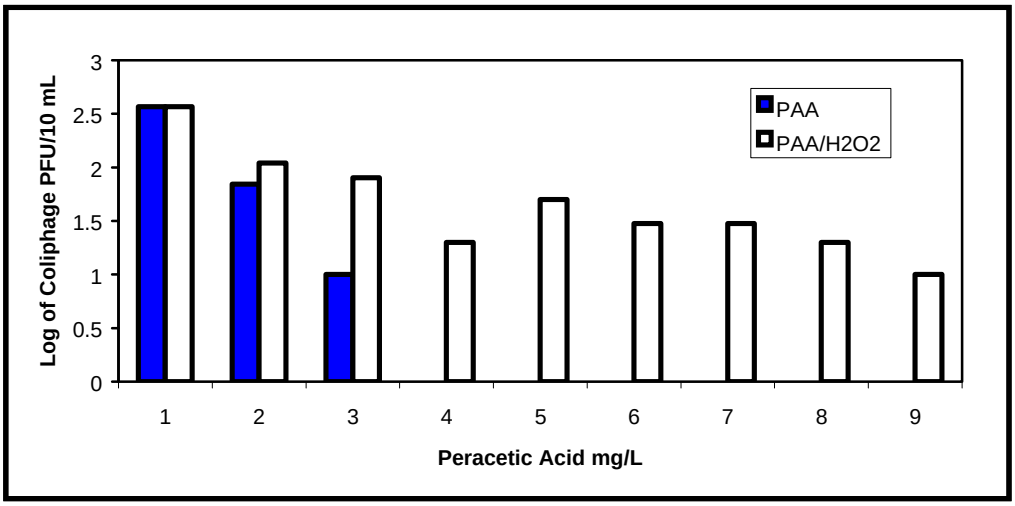

Figure 3.9: Effect of peracetic acid and a mixture of peracetic acid and hydrogen peroxide on coliphage organisms.

3.2.5. Bromine

Bromine was only included in this investigation at a late stage and as a result it was not possible to obtain as much data for this disinfectant as was the case with the other disinfection options. The tests conducted indicate that bromine may be a suitable replacement for chlorine. In tests conducted on the pilot plant, the reduction in microbiological indicator organisms obtained when using bromine at concentrations of approximately 5 mg/L were generally as good as, if not better than those obtained at similar chlorine concentrations, although initial indications are that bromine is not quite as effective as chlorine in removing faecal streptococci organisms. In two separate trials, both chlorine and bromine were successful in reducing the *E. coli* count of the effluent to below 20 CFU/100 mL at applied concentrations of approximately 5 mg/L.

Very little data were obtained for bromine disinfection of parasitic organisms. All the tests were conducted at pilot plant scale and no *Cryptosporidium* oocysts were found in the effluent during the period when these tests were conducted, although a few of the samples were found to contain *Giardia* cysts. The results obtained with bromine were not as good as those obtained using chlorine at an equivalent concentration. However, more comprehensive data will be required before any definite conclusions can be reached in this regard.

The bromine solution used for these tests is manufactured under the tradename of Aquatreat and contained 18-19% m/v Br_2. However, it was found that this solution was unstable and rapidly transformed into bromates. Within the space of a few days the solution could lose

more than half of its strength, due to the formation of bromates. This does not preclude bromine from use for wastewater disinfection, since different methods of bromination are available, which would prevent the formation of bromates. For example ammonium bromide can be used in conjunction with chlorine; the chlorine oxidises the bromide to bromine at the point of additional, so storage of bromine solution is not necessary.

Reports in the literature on the bactericidal efficiency of bromine are sometimes conflicting, but this is probably due to the fact that it is not always clear which species of bromine was being investigated (White, 1992). In wastewater effluents containing ammonia nitrogen and at a pH of between 7 and 8, the dominant species of bromine will be dibromamine. The ammonia concentration of the effluent when these tests were conducted was in fact relatively low (approximately 0,5 mg/L), so one would not have expected the formation of dibromamine to have been particularly high. In any case the disinfection efficiency of dibromamine has been found to be almost equal to that of hypobromous acid, but less effective than that of free bromine, which only exists below pH 7. Since it has been shown that both hypobromous acid and hypochlorous acid are similar in terms of disinfection ability, this means that dibromamine is also comparable to free chlorine for disinfection (White, 1992). This is important in the disinfection of wastewater, since the dibromamine formed is a far stronger disinfectant than the chloramine compounds, giving bromine an advantage over chlorine in this aspect.

One of the reasons that some researchers have reported disappointing results with bromine could be due to the fact that because bromine is such a strong oxidising agent, a significant portion can be lost in side reactions with organic matter present in the wastewater. This is obviously a factor that needs to be investigated further. Other researchers have reported differences in the effectiveness of bromine and chlorine depending on the indicator organism being tested. For example, Koski et al (1966) found that when using *E. coli* as the test organism, 1,0 mg/L bromine was required to give the same reduction as 0,6 mg/L chlorine, but when using another indicator organism, *S. faecalis*, up to 2,0 mg/L was required to elicit the same effect as the 0,6 mg/L chlorine control. It is factors such as this that will require further research.

One disadvantage of bromine which is stated in the literature, is that bromine is more reactive in forming halogenated organics. This means that bromine could potentially have problems when used for wastewater disinfection, although the possibility of using chlorine to first satisfy the halogen demand, followed by bromine and Kott (1969) has demonstrated the

superior disinfection efficiency of a combination of chlorine and bromine. A positive factor for bromine is the fact that because dibromamine has a much greater reactivity than chloramines, the residual when using bromine for wastewater disinfection is generally much lower than when chlorine is used. This could make achievement of the disinfectant residual limit of the DWAF discharge standard easier. Certainly bromine warrants a more in depth study in terms of wastewater disinfection applications.

3.2.6. Mixed Oxidant Generator

The mixed oxidant unit used in this investigation was found to be quite difficult to operate. Fluctuations occurred in the current, which affected the concentration of the anolyte, as well the pH and the oxidation-reduction potential of this stream. The salt tank also needed to be replenished regularly and the depth sensor in the salt tank, which consisted of two electrodes, scaled rapidly, necessitating regular stoppages in order to descale the electrodes. Leaking at the various joints was also a problem and the unit would require some modifications to its design before it could be used for reliable disinfection.

The anolyte emitted a strong chlorine odour when the unit was supplied with 5% sodium chloride. Effluent samples were dosed with the anolyte at concentrations equivalent to chlorine concentrations based on the iodometric titration and under these conditions, there were no significant differences in disinfection efficiency observed between the anolyte and sodium hypochlorite. When using a 5% sodium sulphate solution to supply the unit, the chlorine equivalent concentration of the anolyte as determined from the iodometric titration was too low to allow dosing of the effluent at concentrations of more than 5 mg/L. At 5 mg/L chlorine equivalent concentration, 3 to 4 log reductions in *E. coli* and coliforms were obtained using the anolyte stream produced from sodium sulphate, which was comparable to the disinfection efficiency of chlorine at the same concentration. However, the effect on faecal streptococci and coliphages under the same conditions was significantly worse than that of chlorine. The effect of the mixed oxidant stream on parasitic cysts and oocysts was not assessed, since based on the initial results obtained, the decision was taken not to pursue this method of disinfection further for the purposes of this investigation.

The lack of any obvious benefits in using this mixed oxidant generator as opposed to an ordinary hypochlorite generator, together with the operational problems experienced in operating the unit, do not make this an attractive technique for use on wastewater effluent. In addition, using this unit would not limit the chlorine residual of the treated effluent, and therefore not reduce the harmful disinfection-by-products that would be formed during disinfection.

3.2.7. Klor-Free

Klor-Free was found to be virtually ineffectual for wastewater effluent disinfection. Initially a contact time of 30 minutes was used, but when this failed to produce any disinfection effect at concentrations of up to 50 mg/L, the contact time was increased to 4 hours and concentrations of up to 100 mg/L were used. Even under these conditions, Klor-Free failed to achieve even a 1 log reduction in any of the bacterial indicator organisms.

3.2.8. Ozone Combinations

3.2.8.1. Ozone and UV

The disinfection efficiency of ozone and UV combinations was hindered by problems experienced with the ozone plant towards the end of this investigation when these tests were carried out and so unfortunately, the amount of data obtained for ozone and UV used in combination was limited. Also, the ozone generator problems affected the ozone doses that could be applied, so that it was not possible to apply doses of more than about 0,5 mg/L. The results obtained indicated that a small ozone dose used in conjunction with medium pressure UV irradiation could result in a small improvement in the disinfection in terms of reductions in *E. coli* and coliform organisms, but that it was not significant enough to warrant the additional capital and running costs that ozone would incur. For example when using an ozone dose of as low as 0,1 mg/L it was possible to get complete removal of *E. coli* using doses of around 100 to 150 mW.s/cm^2. The same UV doses used without ozone did not always give complete removal of *E. coli*, but increasing the UV dose (i.e. reducing the flow rate) could in many cases achieve the same result.

The improvement in disinfection in terms of parasitic cyst and oocyst removal was not conclusive. During the period that these tests were conducted, parasitic oocysts (*Cryptosporidium*) were only found to be present during one trial and no *Giardia* cysts were ever found. Unfortunately spiking effluent with parasitic cysts and oocysts for the pilot plant trials was not a feasible option. In the one pilot plant trial in which *Cryptosporidium* was found in the effluent, it was relatively low (11 org/10 L) and was reduced to 1 org/10 L after ozonation and to 0 after ozonation and UV irradiation.

The combination of ozone and UV had very little impact on the normal water quality parameters of the effluent. The BDOC, as expected increased as the ozone dose increased, but apart from that, no obvious effect was observed.

The cost of installing additional lamps to accommodate a lower flow rate through each system would far outweigh the expense of installing ozone. Since the improvement obtained in disinfection using this combination was not significant, this does not appear to be a viable option.

3.2.8.2. Ozone and Chlorine

Ozone, as was demonstrated in **Table 3.4**, is a stronger disinfectant compared to chlorine. However it was observed that the disinfection obtained at the ozone requirement dose was often not as good as that obtained at the chlorine demand dose. The chlorine demand dose was always significantly higher than the ozone demand dose, but the reason for this is probably due to the fact that ozone has a very short residual, while chlorine, especially when used for wastewater disinfection, has a much longer lasting residual. The chlorine demand tests were conducted using a 30 minute contact time, but because of the very short residual obtained with ozone, it is measured immediately after ozone addition, although bacterial indicator tests were only carried out after a 30 minute contact time had been allowed for each disinfectant. This means that the chlorine losses that naturally occur during the 30 minute contact time are accounted for in the chlorine demand determination, but this is not the case for ozone.

It was possible using a combination of ozone and chlorine to effect similar disinfection at much lower disinfectant doses. It was found that when using only half the ozone demand value together with 0,5 mg/L chlorine it was possible to obtain similar disinfection to that obtained at the ozone demand dose. For example, when treating a wastewater effluent with an ozone requirement of 3 mg/L and a chlorine demand of 12 mg/L, it was possible to achieve a 2 log removal of coliform organisms and 3 log removal of *E. coli* at the ozone demand dose of 3 mg/L. A chlorine dose of 3 mg/L was unable to achieve even a 1 log removal of these indicator organisms. However, using half the ozone demand value (1,5 mg/L ozone) together with 0,5 mg/L chlorine effected a 3 log removal of coliforms and complete removal (>3 log) of *E. coli.* **Table 3.13** shows some of the results obtained and compares the disinfection obtained at different ozone and chlorine doses as well as with combinations of ozone and chlorine.

Table 3.13: Log removals in *E. coli* and coliforms obtained using ozone, chlorine and combinations of ozone and chlorine in laboratory tests.

Disinfectant	*E. coli*		Coliforms		Demand	
	Log rem.	Dose	Log rem.	Dose	Chlorine	Ozone
Ozone	3	3	2	3	12	3
Chlorine	0	3	0	3		
O_3 / Cl_2	>3*	1,5 O_3 0,5 Cl_2	3	1,5 O_3 0,5 Cl_2		
Ozone	2	7,5	2	7,5	12	7,5
Chlorine	>3*	12	>3*	12		
O_3 / Cl_2	2	3,75 O_3 1,0 Cl_2	2	3,75 O_3 1,0 Cl_2		
Ozone	>3*	4	3	4	10	4
Chlorine	>3*	10	>3*	10		
O_3 / Cl_2	3	2,0 O_3 0,5 Cl_2	2	2,0 O_3 0,5 Cl_2		
Ozone	2	3	2	3	14	3
Chlorine	3	14	3	14		
O_3 / Cl_2	2	1,5 O_3 1,0 Cl_2	2	1,5 O_3 1,0 Cl_2		
Ozone	1	2	1	2	2	9
Chlorine	>4*	9	>4*	9		
O_3 / Cl_2	3	0,66 O_3 1,0 Cl_2	2	0,66 O_3 1,0 Cl_2		

*: complete removals of these organisms were obtained at these concentrations.

Modifying a wastewater works to include ozonation would obviously be an expensive option, although it may offer a means of achieving effective disinfection without exceeding the chlorine residual limit of 0,1 mg/L. In the tests conducted in this investigation, it was possible to achieve 100% compliance with the relaxation of the DWAF effluent discharge standard of 500 faecal coliforms CFU/100 mL when using half the ozone demand together with 0,5 mg/L chlorine. This compared favourably with using ozone at the demand concentration to disinfect the same effluent. In 50% of the cases the *E. coli* counts obtained using half the ozone demand dose and 0,5 mg/L chlorine were less than 100 CFU/100 mL and in about one third this was below 20 CFU/100 mL. These results were comparable with those obtained using ozone at the

demand value, where about 80% of the disinfected effluent samples had *E. coli* counts of less than 100 CFU/100 mL and in about 50% of the cases this was below 20 CFU/100 mL.

In terms of *Cryptosporidium* and *Giardia*, the only benefit observed when using ozone and chlorine rather than ozone alone, was that, as was found with the other indicator organisms, equivalent disinfection to that obtained at the ozone demand dose could be achieved at about half this dose when used together with 0,5 to 1 mg/L chlorine.

3.2.8.3. Ozone and Peracetic Acid

Peracetic acid was found to be comparable to chlorine in terms of disinfection efficiency. It was therefore not surprising to find that ozone and peracetic acid used together gave results that were similar to those obtained using ozone with chlorine. As was found with ozone and chlorine, when using ozone together with peracetic acid, it was possible at half the ozone demand dose and between 0,5 and 1,0 mg/L peracetic acid to achieve similar disinfection to that obtained using ozone alone at the demand concentration. In a total of four laboratory trials and one pilot plant trial, it was possible to achieve an *E. coli* count of less than 500 CFU/100 mL in each case using an ozone dose of half the demand concentration together with 0,5 mg/L peracetic acid. However, without increasing the peracetic acid concentration to 1 mg/L, it was not possible under these conditions to get the *E. coli* count to below 100 CFU/100 mL. The removal of *Cryptosporidium* and *Giardia* when using half the ozone demand dose and 0,5 to 1,0 mg/L peracetic acid was also comparable to that achieved at the ozone demand concentration.

A benefit that ozone and peracetic acid have over ozone and chlorine is that with the former, it is possible to limit the chlorine demand to zero, but since ozone and ozone/peracetic acid did not display an improvement in parasitic cyst and oocysts inactivation over peracetic acid used alone at higher concentrations, there would be no incentive for installing a disinfectant such as ozone.

3.2.8.4. Ozone and Bromine

Bromine has been found to form bromates in the presence of ozone and therefore only preliminary testing was conducted on this combination of disinfectants as it was suspected that it would be unsuitable to combine ozone and bromine. Bromate levels of over 1 mg/L were found in the wastewater effluent treated with bromine at 5 mg/L even though no ozone had been added. Although there are no bromate limits stated for wastewater effluents, this figure is high and caution will need to be exercised when using this source of bromine for disinfection.

The results of these preliminary tests suggested that ozone actually impairs the disinfection efficiency of bromine and this is most likely due to ozone converting bromine to bromates, which have no disinfection activity. The bromate concentration was found to increase after ozonation. When using lower ozone and bromine concentrations (between one third and one half the ozone demand concentration and 0,5 mg/L bromine), the ozone/bromine combination was generally found to be more effective than the ozone/chlorine combination at equivalent concentrations, but as the ozone dose increased, so the disinfection efficiency of ozone/bromine deteriorated. Only preliminary testing was carried out to assess the effect of an ozone/bromine combination on parasitic organisms. Initial results indicate that there may be some improvement in the disinfection of *Cryptosporidium* oocysts relative to that obtained using ozone alone, although no improvement was evident for *Giardia* cysts. However, a more in depth investigation would be required before reaching any definite conclusions. This combination is however, not considered a practical method of wastewater disinfection.

3.2.9. Ultraviolet Irradiation Combinations

3.2.9.1. UV and Chlorine

All the investigation work on this combination of disinfectants was carried out on the pilot plant. The reason for this was that only medium pressure UV irradiation and chlorine were studied and it was not possible to conduct laboratory tests using the medium pressure UV unit. It was therefore not possible to assess as wide a range of UV doses and chlorine doses as would have been the case in laboratory testing. In spite of this, UV doses from around 50 and to above 200 mW.s/cm^2 were investigated with chlorine doses from as low as 1 mg/L to 2 mg/L above the chlorine demand value. The chlorine demand of the effluent varied between 7 and 18 mg/L during these trials.

Chlorine could offer some benefits in terms of disinfection efficiency when used in conjunction with UV. For example, when using UV irradiation at a dose of approximately 60 mW.s/cm^2 it was possible to achieve a 2 to 3 log removal in the coliforms, *E. coli* and faecal streptococci organisms. Using the same UV dose together with 1 mg/L chlorine (the chlorine demand of the effluent in this case was 7 mg/L) brought about a complete reduction in *E. coli* and faecal streptococci and a 4 log reduction in coliforms. No improvement in the removal of total plate count organisms was obtained. In most cases though, the chlorine doses which needed to be added in order to improve the disinfection efficiency relative to that obtained using UV alone, were fairly high, often close to the chlorine demand of the effluent.

There was also no evidence to suggest that this combination of disinfectants would increase inactivation of parasitic cysts and oocysts and since UV disinfection was so effective in removing coliphage organisms, the addition of chlorine was found to be superfluous. No data on the disinfection of *Cryptosporidium* oocysts were obtained for a UV/chlorine combination, since none of these organisms were found to be present in the effluent when these tests were carried out. *Giardia* cysts were only found in the effluent on a few occasions, but no obvious benefits for using UV and chlorine as opposed to UV alone, were evident.

Assessment of the compliance obtained with the DWAF relaxed discharge standard for faecal coliforms highlighted the fact that chlorine used together with UV does not improve disinfection significantly enough to warrant its use. In these tests it was found that chlorine, when used at the chlorine demand concentration, which is in fact far higher than the doses that would be used in practice, achieved 100% compliance with the DWAF relaxation standard of 500 faecal coliforms CFU /100 mL and in as many as 80% of the cases, the *E. coli* counts were below 100 CFU/100 mL. At these chlorine concentrations it was possible to achieve *E. coli* counts of less than 20 CFU/100 mL about 40% of the time. This compared favourably with the results obtained using medium pressure UV (see **Table 3.14**). When using chlorine together with UV irradiation, there was a small improvement. It was possible to reduce the *E. coli* counts to below 100 CFU/100 mL 100% of the time and to below 20 CFU/100 mL about 70% of the time, but this was only when using chlorine doses close to the demand dose. Apart from the fact that this would not be an economically viable option for wastewater disinfection, the high chlorine doses generally required for improved disinfection would exceed the discharge residual limit of 0,1 mg/L.

Table 3.14: Compliance achieved with *E. coli* counts of <500, <100 and <20 CFU/100 mL using chlorine (within 2 mg/L of chlorine demand dose), medium pressure UV irradiation (48 to 219 mW.s/cm^2) and UV/chlorine (UV 48 to 219 mW.s/cm^2 and chlorine at between 1 and 20 mg/L).

	Chlorine Compliance (%)	UV Compliance (%)	UV and Chlorine Compliance (%)
<500 CFU/100 mL	100	100	100
<100 CFU/100 mL	80	80	100
<20 CFU/100 mL	40	40	70

3.2.9.2. UV and Peracetic Acid

As with the UV/chlorine tests, all the UV/peracetic acid tests were conducted on the pilot plant, since laboratory testing using the medium pressure unit were not possible. The chlorine demand of the effluent during the period in which these tests were conducted, varied between 7 and 20 mg/L and UV doses of between 60 and 135 mW.s/cm^2 were used with peracetic acid concentrations of between 1 and 20 mg/L.

It has already been found that the disinfection efficiency of peracetic acid is similar to that of chlorine at equivalent concentrations, so it was not surprising to find that UV/peracetic acid combinations gave similar results to those obtained for UV/chlorine at corresponding doses. It was possible to improve the disinfection efficiency of UV using peracetic acid, but usually only at peracetic acid doses close to the chlorine demand of the effluent. Since only pilot plant tests were carried, it was not feasible to spike the effluent with *Cryptosporidium* and *Giardia* prior to disinfection. Unfortunately *Cryptosporidium* was not found in the effluent during these tests, only *Giardia*, but since there was very little reduction in these cysts after treatment, it can be assumed that the reduction in *Cryptosporidium* oocysts that could be achieved would also be low.

As with the UV/chlorine combination, the small improvement achieved using UV/peracetic acid, hardly warrants the high peracetic acid doses required. It would be far better to consider using higher UV doses before incurring the additional inconvenience and cost of using peracetic acid in conjunction with UV irradiation.

3.2.9.3. UV and Bromine

Only preliminary testing was carried out on bromine and UV irradiation, since bromine was only included in the investigation at a very late stage. As with the other UV combinations, testing was conducted on the pilot plant only. These initial tests indicate, that as was found with UV/chlorine and UV/peracetic acid, bromine can cause an improvement in the disinfection obtained using UV, but that this is not significant enough to justify using this combination of disinfectants. Tests on parasitic cysts and oocysts were dependent on a natural supply in the wastewater effluent and once again this did not contain any *Cryptosporidium* oocysts, although relatively high concentrations of *Giardia* cysts were present. The removal of these organisms was not improved by using bromine in conjunction with UV irradiation.

3.2.10. Membranes

During this project, alternative disinfectants for the removal of *Cryptosporidium* oocysts and *Giardia* cysts and other bacterial indicator organisms in wastewater effluent were investigated, but another approach would be to use filtration and physically remove these organisms from the effluent. Parasitic cysts and oocysts, being larger than most other micro-organisms, are especially suited to this type of treatment. However, despite initial intentions to investigate the possibility of using membranes for wastewater disinfection, work being conducted by Umgeni Water on tubular filter press for the treatment of potable water sludges, made it evident that the nature of the sludge is very important. The tubular filter press is a dynamic microfiltration process using woven fabric tubes as the support (Richards et al, 2000). However, discussions with the process engineer involved in this project made it clear that the viscous nature of wastewater sludges, together with the propensity of wastewater to develop biofilms, would create serious operation problems and that membranes would not be a suitable candidate for these applications, especially when cheaper and simpler alternatives are available.

3.2.11. *Cryptosporidium* and *Giardia* Results

In the first half of the project, after extensive trials and modifications of the methods, very few viable oocysts had been recovered. Further investigations proved that there could have been a number of causes contributing to this. Owing to the degradation of the microscope lens and filters giving very poor fluorescence and the possible malfunction of the flow cytometer, it was never possible to check the viability of the stock seed oocysts. The stock seed was very difficult and expensive to obtain from the USA, requiring a special import permit and transport conditions. Also, the nature of the secondary effluent gave rise to extremely difficult microscopy owing to the concentration of suspended solids, which obscured the view of the

oocysts, even with sucrose flotation clarification procedures. *In vitro* excystation performed was also impossible to view successfully for the same reason.

In the second half of the project and after changes in equipment, source of seed and reagents and after various trials one viability assay was found to give reliable results. Propidium iodide (PI) and fluorescein isothiocyanate (FITC) staining (Waterborne Inc.2000) were chosen and gave positive results for the viable (oo)cysts. The ratio of viable and non-viable counts was used to calculate the viability and to compare to the effluent before disinfection treatment.

Overall, little decrease in viability was found for *Cryptosporidium* over all treatments and there appeared to be little significant difference between them. The viability of the raw averaged 88% and after treatment averaged 77%.

Similarly, little decrease in viability was found for *Giardia* over all treatments and there appeared to be little significant difference between them. The viability of the raw averaged 75% and after treatment averaged 60%.

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However, the disinfection processes did appear to have a significant effect on reducing the actual number of parasites by some mechanism. It is not clear whether this removal is owing to physical means, disruption of the antigenic binding sites (although their presence was still apparent in the viability assays), or lysis of the (oo)cysts.

Clancy et al (2000) showed that all four of the surrogate viability assays used (DAPI/PI, SYTO®-9, SYTO®-59, and *in vitro* excystation) significantly underestimated oocyst inactivation when compared to that measured by mouse infectivity. It appeared that doses as low as 40 mW.s/cm^2 (fresh oocysts) and 14 mW.s/cm^2 (aged oocysts) provided over 2 log inactivation by mouse infectivity, while the surrogates showed less than 0.5 log inactivation. Further work has shown that this phenomenon (lack of correlation between the *in vitro* assays and animal infectivity) extends to oocysts exposed to ozone as well. Therefore, although our viability assays apparently show little inactivation of the oocysts and cysts, they may still nevertheless be non-infective. Unfortunately, animal infectivity studies were beyond the scope of the project.

3.3. Summary

A number of conclusions were made from this investigation. These are:

1. The most promising disinfectants for use in wastewater, which show potential for meeting the DWAF standard for effluent discharge in terms of both chlorine residual and faecal coliforms are:
   - ozone
   - medium pressure UV irradiation
   - bromine
   - peracetic acid.

2. Combinations of ozone and UV with other disinfectants are difficult to justify in terms of the improvements obtained in disinfection. It is possible to achieve similar disinfection at lower doses of the combined the disinfectants than can be achieved using each disinfectant alone, but the additional costs and operational difficulties that would be incurred by using disinfectant combinations is not warranted by the improvement in disinfection.

3. The mixed oxidant generator for the most part produces hypochlorite and so does not offer any significant benefits over chlorine gas disinfection and Klor Free did not prove to be a suitable alternative to chlorine for wastewater disinfection.

4. Membranes were not investigated, but work conducted by Umgeni Water on membranes for the treatment of potable water sludges made it clear that the nature of the sludge is vitally important and that serious operational problems could be expected when using membranes for these applications. Therefore membranes were not investigated for the purposes of this investigation.

4. Considerations for Wastewater Disinfection

4.1. Cost Assessment

Based on the results of this investigation, ozone, medium pressure UV irradiation, peracetic acid and bromine were selected as potential candidates for the disinfection of wastewater treatment. Details of the equipment and products used are given in Chapter 6 on Analytical Methodology in the Appendix, and references pertaining to these can be found in Chapter 5, References. The advantages and disadvantages of these disinfectants are discussed and a cost assessment has been carried out in order to compare these alternatives. Chlorine is included in the cost assessment in order to provide a benchmark. The cost assessment has taken into account the dosages that will be required in order to satisfy the DWAF relaxed standard for wastewater discharge of 500 faecal coliforms CFU/100 mL as well as the higher doses that will be needed if the general effluent standard of 0 faecal coliforms CFU/100 mL is to be met. The problem of diminishing returns occurs with some of the disinfectants in trying to meet the 0 faecal coliforms standard. For example with chlorine, a dose of around 6 mg/*l* was generally needed to meet a standard of 500 *E. coli*, while a dose of about 50% more than this was required to satisfy a 0 *E. coli* standard. With ozone, an applied concentration of between 1 and 2 mg/*l* generally satisfied a 500 *E. coli* standard, but in order to reach a goal of 0 *E. coli*, three to four times this dose needed to be applied. The cost assessment appears in **Table 4.1**.

TABLE 4.1: Cost assessment for most promising alternative disinfectants for doses required to meet both the relaxed (<500 CFU/100 mL) and general (0 CFU/100 mL) DWAF effluent discharge standards.

Disinfectant	**Dose for <500 *E. coli***	**c/kL Costs for <500 *E. coli***	**Dose for <10 *E. coli***	**c/kL Costs for <10 *E. coli***
Ozone	2	3,45	8	9,45
UV	40 mW.s/cm^2	3,72	150 mW.s/cm^2	11,2
Peracetic acid	5	40,0	9,5	76,0
Bromine	6	50,0	9,5	79,2
Chlorine	6	3,00	9,5	4,75

4.2. Ozone

It was determined from the experimental data, that an applied ozone concentration of almost 2 mg/L is needed to meet the relaxed effluent discharge standard. In satisfying the general standard of 0 faecal coliforms the ozone dose needed to be increased significantly to between 6 and 8 mg/L. The capital costs for ozone were based on a 10 kg/h plant to achieve the relaxed standard. This allows for a 50% error of margin on a dose of 2 mg/L for an 80 ML/d wastewater works. The 50% margin of error was used since the dosage is so low (50% equates to only 1 mg/L). The costs for achievement of the general standard were based on a 30 kg/h plant, which allowed for a 20% margin of error at an applied concentration of 8 mg/L. The capital costs were calculated as being R4,8 million and R15 million for the 10 and 30 kg/h plants respectively and these costs were amortised over a period of 10 years at 12% interest per annum.

Although the capital costs of ozone are expensive, this is offset in some degree by relatively low running costs. However, ozone requires skilled personnel in order to run and maintain the ozone generation plant, so this will most probably pose a problem at most wastewater works. In terms of meeting the relaxed discharge standard of <500 faecal coliforms CFU/100 mL, ozone costs are comparable to chlorine. However, this limit is only applied under special circumstances and when using ozone to meet the general standard, the costs rise steeply. In addition it will in most cases be necessary to hire additional, highly skilled staff to run the ozone plant. Based on this, ozone does not appear to be a suitable alternative to chlorine for the disinfection of wastewater effluent.

4.3. Medium Pressure UV

It was possible to achieve 98% compliance of the relaxed discharge effluent with the medium pressure UV system, even using doses as low as 30 mW.s/cm^2. However, to ensure better compliance a slightly higher dose, closer to 50 mW.s/cm^2 would be recommended. In order to meet the general discharge standard it would be necessary to use higher UV doses and according to this investigation, doses of around 150 mW.s/cm^2 would be required to do this consistently.

The capital costs for medium pressure UV were based on both open channel and closed systems, but in both cases to treat to a dose of around 40 to 50 mW.s/cm^2, which would allow compliance with at least the relaxed discharge limit. This would require in the region of thirty 7 kW medium pressure lamps and gives capital costs for both open channel and closed systems of approximately R3 million. This figure was amortised over a period of 10 years using an annual interest rate of 12%. Lamp replacement and maintenance would cost in the region of R40 000 to R50 000 per

annum, while electricity costs, taken as 27 c/kW hour, would add on approximately 1,7 c/kL. This gives a total cost in the region of 3,715 c/kL. Increasing the dose to 150 mW.s/cm^2 to ensure compliance with the general effluent discharge standard of 0 faceal coliforms would obviously increase the costs significantly. In this case, capital costs would be around R9 million, and including lamp replacement and maintenance costs as well as electricity costs, this would come to a figure of around 11,16 c/kL. It needs to be stressed though that it will be possible to operate the plant at a much lower dose than 150 mW.s/cm^2 and still achieve the general standard for much of the time.

Apart from the fact that the running and maintenance costs of UV are relatively low and that UV produces no disinfectant residual, another advantage to using UV is that it is extremely simple to maintain and operate. UV sensors and alarms can be fitted to the units to warn of low UV intensity, lamp fouling etc. It is also relatively simple to retrofit a plant for UV disinfection, or to upgrade a plant. A disadvantage that was discovered during the pilot plant tests conducted in this investigation is that the wastewater effluent being irradiated needs to be of a fairly high standard. Whenever problems were experienced on the full scale wastewater works which affected the quality of the secondary wastewater effluent, the transmittance of the effluent would drop to an unacceptable level, so compromising UV disinfection. It needs to be added though that during these periods, chlorine disinfection was also inadequate.

4.4. Peracetic acid

In this investigation, peracetic acid was found to have 100% compliance with the relaxed effluent discharge standard when used at a concentration of 5 mg/L or more. However, in meeting the general standard of 0 faceal coliforms it was necessary to almost double this dose. At present peracetic acid could be purchased in solution form consisting of 15% peracetic acid and 23% hydrogen peroxide at a cost of around R12-00/kg, if purchased in bulk. However, at this price peracetic acid is not an economically viable option and despite the promising results obtained, the price would need to drop before this could be considered for disinfection of wastewater.

4.5. Bromine

Bromine was found to be similar to chlorine for the disinfection of wastewater and in fact the bromine concentrations required to meet the relaxed and general discharge standards were also similar. Bromine was obtained in solution form as an 18-19% solution under the trade name of Aquatreat, however this solution was found to be unstable and rapidly reacted to form bromates. In a period of only three days the bromine solution was found to have lost about half its strength, the balance having transformed to bromates. The instability of the solution together with the high costs relative to chlorine, do not make this solution a suitable alternative for wastewater disinfection. Different methods of bromination, such as ammonium bromide used in conjunction with chlorine, may however offer cost effective options for wastewater disinfection.

4.6. Recommendations for Future Research

Both ozone and UV have been researched fairly intensively and apart from work on high intensity low pressure systems, no further work appears to be necessary at this stage.

The tests conducted in this investigation revealed that bromine is an effective disinfectant for wastewater effluent. However, the bromine solution used in the pilot plant tests described in this report, was found to be unstable and as such, not suitable for these applications. Different methods of bromination, such as ammonium bromide used in conjunction with chlorine warrant further investigation.

Peracetic acid was produced in relatively small quantities for the experimental tests carried out for this investigation, but should full scale plant applications be considered, commercially available solutions would be required before this would be a feasible option. Peracetic acid is commercially available in Europe and this would need to be tested before implementation locally, but the cost would not be attractive under present conditions.

4.7. Status in Terms of *Cryptosporidium* and *Giardia*

One of the problems experienced in carrying out the research for this project, was difficulty in establishing the viability of (oo)cysts once they had been detected. Future work into tests demonstrating the viability or infectivity of (oo)cysts is needed before definitive results on *Cryptosporidium* and *Giardia* removal can be reported for investigations of this nature. Measuring *Cryptosporidium* and *Giardia* recoveries tends to be low (typically 30%) and

improvement in detection and recovery techniques would be beneficial as regards accuracy of results.

4.8. Conclusions

The conclusions are discussed in terms of the objectives of this project:

1. Adequately disinfect wastewater effluents in terms of parasitic (oo)cysts (*Giardia* and *Cryptosporidium*), viruses (coliphages) and bacterial indicator organisms (*E. coli*, faecal streptococci and coliforms) while not exceeding DWAF general standards for chlorine residual.
   It was shown that there are a number of disinfectants capable of achieving this, such as ozone, UV, peracetic acid, bromine and combinations of chlorine with ozone or UV.

2. Adequately disinfect wastewater effluents in terms of the above without producing a chlorine residual.
   This was also achieved by using disinfectants such as ozone, UV, peracetic acid and bromine.

3. Provide design and operational guidelines for simple and effective disinfection of wastewater effluents.
   The basic operational parameters required when using the suitable options are listed in this report.

4. Provide a disinfectant protocol suitable for use under low technology applications.
   UV is a possibility for rural applications, provided that adequate instrumentation is installed to detect failure of lamps or poor transmissivity. Obviously a pre-requisite for UV disinfection would be an adequate power supply. Peracetic acid could also provide a method of wastewater disinfection suitable for rural areas. It is comparable to hypochlorite solutions in terms of required handling precautions, it is more stable than hypochlorite solutions, doesn't produce a chlorine residual and only requires a simple dosing pump. However, the cost would be prohibitive at this stage, and will remain so until peracetic acid is imported in bulk or is manufactured locally.

5. Determine the potential for regeneration of micro-organisms, especially *Giardia*, *Cryptosporidium* and viruses after different methods of disinfection.
For normal bacteriological indicator organisms and parasitic (oo)cysts, regrowth does not appear to be a problem within the limits of the indications of the tests.

5. References

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APPENDIX

6. Analytical Methodology

6.1. Microbiological Analyses

6.1.1. Coliforms and *E. coli*

Coliforms and *E. coli* were analysed by a membrane filtration method using membrane lauryl sulphate broth, according to a South African National Accreditation Services (SANAS) accredited method equivalent to SABS method 221.

6.1.2. Faecal Streptococci

Faecal Streptococci were analysed by a membrane filtration method using an agar medium containing sodium azide according to a SANAS accredited method equivalent to a method in Standard Methods for the Examination of Water and Wastewater.

6.1.3. Coliphages

Coliphages were analysed using a double agar layer plaque assay for somatic coliphages, according to a SANAS accredited method equivalent to ISO method10705-2 1997.

6.1.4. Colony counts

Total colony counts at 37°C (24 hour), were analysed by a pour-plate method with yeast-extract agar according to a SANAS accredited method equivalent to SABS method 221. Counts at 22°C were conducted similarly, but for 3 d.

6.1.5. *Cryptosporidium* and *Giardia*

Cryptosporidium and *Giardia* were analysed by an immuno-fluorescence assay using fluorescein isothiocyanate (FITC) labelled antibodies specific to *Cryptosporidium* and *Giardia* (Waterborne Inc 2000). Concentration was either by direct centrifugation or flocculation (Vesey et al, 1993).

Viability assays included all or some of the following:

1. *In vitro* excystation (Upton et al.1994 and Current and Box, modified by Waterborne Inc. 2000)

2. Fluorescein diacetate (FDA) and tetramethyl red (TMR) staining (Jarmey-Swan et al. 2000)
3. Propidium iodide (PI) and fluorescein isothiocyanate (FITC) staining (Dowd and Pillai 1997, and Waterborne Inc. 2000)
4. Differential interference contrast (DIC) microscopy (to confirm results of fluorogenic dyes).

6.2. Chemical Analyses

6.2.1. pH and temperature

pH was measured on a Radiometer PHM 95 pH/ion meter with a temperature compensation probe and thermometer, which was also used to measure the temperature.

6.2.2. Turbidity

Turbidity was determined using a Hach Ratio/XR model 43900 turbidity meter.

6.2.3. Conductivity

Conductivity was measured in mS/m on a conductivity meter using a potassium chloride regference solution (0,0100 M) according to a SANAS accredited method.

6.2.4. Alkalinity

Alkalinity analyses were performed on a Mettler DL25 Autotitrator using 0,02 N hydrochloric acid and titrating to the m-value (approximately pH 4,6) to allow for determination of the total alkalinity according to a SANAS accredited method.

6.2.5. Calcium, Magnesium and Hardness

Calcium and magnesium were determined using ICP-AES on a Varian Radial ICP according to a SANAS accredited method. Hardness (as mg/L $CaCO_3$) was calculated from the calcium and magnesium analyses.

6.2.6. Iron

Iron was analysed by ICP-AES on a Varian Radial ICP according to a SANAS accredited method.

6.2.7. Manganese

Manganese was analysed by Inductively Coupled Plasma - Atomic Emission Spectroscopy (ICP-AES) on a Varian Radial ICP according to an SANAS accredited method.

6.2.8. Total Dissolved Solids

Total dissolved solids were determined by accurately weighing the residue obtained after drying 250 mL sample overnight in an oven at 105 ± 5 °C according to a SANAS accredited method.

6.2.9. Suspended Solids

Suspended solids were determined by measuring the residue trapped on a glass microfibre filter paper after filtering a measured volume of sample through the filter paper under vacuum and then drying the filter paper to constant weight at 105 ± 5°C according to a SANAS accredited method.

6.2.10. Chemical Oxygen Demand

Chemical oxygen demand was determined using a standard potassium dichromate digestion procedure conducted in a microwave, according to a SANAS accredited procedure.

6.3. Organic Analyses

6.3.1. Total Organic Carbon, Dissolved Organic Carbon and Biodegradable Dissolved Organic Carbon Analysis

Total organic carbon (TOC) and dissolved organic carbon (DOC) concentrations were determined using the persulphate-ultraviolet oxidation method (method 5310C in Standard Methods for the Examination of Water and Wastewater, 1998) utilising an Aquadoc Total Organic Carbon Analyser. Prior to analysis of DOC, samples were filtered through 0,45 µm membrane filters (Millex, Millipore). All analyses were performed in at least duplicate.

Biodegradable dissolved organic carbon (BDOC) is defined as the fraction of DOC that is removed by heterotrophic microorganisms over a period of 28 days and analyses were performed according to the method described by Servais et al (1989). 200 mL of sample was sterilised by filtration through 0,2 µm membrane filters (Sartorius cellulose acetate membrane filters), carefully rinsed first with ultrapure water (Millipore Milli-Q) and then with water

sample. An inoculum was prepared by filtering a raw water obtained from the same environment as the sample through a 1,2 µm membrane filter (Sartorius cellulose acetate membrane filter). The method described by Servais et al (1989) called for a 2,0 µm filter for filtration of the inoculum, but despite repeated efforts to obtain these filters, it became necessary to use the 1,2 µm filters instead. 2 mL of inoculum were added to 200 mL of sterilised sample, part of which was then placed in a 100 mL glass stoppered reagent bottle and water sealed. The sample was incubated in the dark at between 20 and 22 °C for 28 d. Analysis of the DOC was carried out on a subsample of the water collected prior to incubation and on the sample at the end of the incubation period. In this case filtration was obviously through a 0,2 µm membrane filter and not a 0,45 µm membrane filter as described above for DOC analysis The BDOC value was calculated as the difference between the initial and final DOC results.

6.3.2. Trihalomethane Formation Potential Analysis

Trihalomethane formation potential (THMFP) was determined using the THMFP test described in section 5710B of Standard Methods for the Examination of Water and Wastewater (1998), although the test was carried out at a pH of 9,2 ± 0,2 as recommended in section 5710C of the 18th Edition of Standard Methods for the Examination of Water and Wastewater (1992) for the basic THMFP test. This test simulates the conditions experienced in high pH waters and accelerates THM formation. A measured amount of the water sample was placed in a glass stoppered bottle and sufficient chlorine added to the water sample to ensure that a chlorine residual of at least 3 mg/*l*, but not more than 5 mg/*l*, remained at the end of the 7 day incubation period. The pH of the chlorinated water sample was raised to 9,2 ± 0,2 and the bottle was water sealed and incubated in the dark at 25 ± 2°C for 7 d. The THM concentration of the water sample prior to chlorination and at the end of the 7 day incubation period was measured and the THMFP calculated from the difference between these THM concentrations. THMs were determined on a Varian 3600 gas chromatograph using direct aqueous injection with a suitable thermal programme and an internal 1,2-dibromomethane standard.

6.3.3. Ultraviolet Absorbance

Ultraviolet (UV) absorbance of water samples, after filtration through 0,45 µm membrane filters (Millex, Millipore), was measured at 254 nm using a Pharmacia Ultraspec III spectrophotometer and Autofill III autosampler with a 10 mm quartz cell. The UV light source was provided by a deuterium lamp. The procedure followed is described in section 5910B of Standard Methods for the Examination of Water and Wastewater (1998).

6.4. Determination of Chlorine, Bromine, Ozone, Hydrogen Peroxide and Peracetic Acid

6.4.1. Iodometric Titration for the Determination of Chlorine

The iodometric method involves the liberation of iodine from potassium iodide by an oxidant, in this case chlorine, followed by titration of the liberated iodine with sodium thiosulphate. The titration is carried out at a pH of between 3 and 4 since the reaction is not stoichiometric at neutral pH as a result of partial oxidation of the thiosulphate to sulphate. This method is suitable for measuring concentrations of chlorine of around 1 mg/*l* or higher, but is subject to interference from strong oxidising agents, such as ozone and hydrogen peroxide. The method used is described in section 4500-Cl B of Standard Methods for the Examination of Water and Wastewater (1998).

6.4.2. Iodometric Titration for the Determination of Bromine

Bromine concentration was determined using the same method as that used for the chlorine species, taking into account the equivalent mass of bromine.

6.4.3. Iodometric Titration for the Determination of Ozone

An iodometric titration method was also used for measuring ozone concentration in the reactor column of the laboratory apparatus during calibration and for determination of the ozone concentration in the off-gas from this reactor column. When determining the ozone concentration in the reactor column, the column was filled with a solution of potassium iodide and then a measured amount of ozone containing gas was passed through the column after which the concentration of liberated iodine in a measured amount of the potassium iodide solution was determined by titration. The off-gas was passed through a potassium iodide trap and then the concentration of liberated iodine in the trap determined.

The same procedure was used to measure the concentration of ozone in the gas being injected into the pilot plant as well as the concentration of ozone in the off-gases from the contact/separator column of the pilot plant. In both cases, measured amounts of gas (measured on a Alexander Wright Model Number DM3 B gas flow meter at ambient temperature and pressure) were passed through potassium iodide traps and then the amount of liberated iodine

determined titrimetrically as described in **section 6.4.1** taking into account the equivalent mass of ozone.

6.4.4. Indigo Colorimetric Method for the Determination of Ozone

The indigo colorimetric method was used for measurement of dissolved ozone in water samples. This method is based on the principle that under acidic conditions, ozone rapidly decolorises indigo dye and the decrease in absorption is linearly proportional to the ozone concentration. The method used is described in section 4500-O_3 B of Standard Methods for the Examination of Water and Wastewater (1998).

10 mL of indigo reagent were placed in a 100 mL volumetric flask and the flask was filled to the mark with the ozone-containing sample or a dilution of the sample, ensuring that complete decoloration of the indigo did not occur. A sample blank was prepared by adding ozonated sample from which the ozone was first removed by the addition of sodium thiosulphate, to a flask containing 10 mL indigo reagent and filling the flask to the mark. The absorbance of the samples and sample blank were measured at 600 nm on a Pharmacia Ultraspec III spectrophotometer and Autofill III autosampler using a 10 mm quartz cell. The ozone concentration in a sample could then be calculated from the difference between the blank and sample absorbance (Standard Methods for the Examination of Water and Wastewater (1998) (section 2350B(g)), using 0,01N sodium thiosulphate).

6.4.5. Iodometric Titration for the Determination of Hydrogen Peroxide

An iodometric titration was used for standardisation of the hydrogen peroxide solution used in the disinfection tests. The principle of the method is similar to that described in **section 6.4.1** in that under acidic conditions hydrogen peroxide reacts with iodide to release iodine, which can then be measured by titration against sodium thiosulphate. The method used is described in section X, 123 of Vogel's Textbook of Quantitative Inorganic Analysis (1993). The hydrogen peroxide solution to be standardised was diluted to approximately 0,3%. A 25 mL sample of diluted solution was added gradually with constant stirring to a solution of 1 g potassium iodide in 100 mL of sulphuric acid (1:20) contained in a stoppered bottle. The mixture was allowed to stand for 15 minutes after which the liberated iodine was titrated with standard 0,1 N sodium thiosulphate with the addition of 2 mL starch solution when the colour of the iodine was almost completely discharged.

6.4.6. Iodometric Titration for the Determination of Peracetic Acid

The concentration of peracetic acid was determined using an iodometric titration procedure described in Vogel's Practical Organic Chemistry, in which iodine liberated by peracetic acid under acidic conditions is titrated against a standardised sodium thiosulphate solution.

6.5. Materials and Procedures

6.5.1. Seeding and Viability of the Protozoa

In the first half of the project viable samples of *Cryptosporidium* oocysts and *Giardia* cysts were obtained from Sterling Parasitology Laboratory for seeding experiments. 1000 to 4000 *Giardia* cysts and *Cryptosporidium* oocysts were sorted using the Becton Dickinson FACSCalibur flow cytometer and seeded per litre of effluent.

In the second half of the project, new viable (oo)cysts were sourced from Waterborne Inc., USA. Different levels of seeding were applied and optimised at 2000 oocysts and cysts per 10 liters (in theory, by calculation) into the effluent, which was found to give good recoveries for enumeration and adequate numbers for the viability assays. Empirical enumeration with the flow cytometer was not carried out on the seed, however and reliance was placed on the ratio of counts before and after disinfection treatment.

6.5.1.1. Viability Assays

Cryptosporidium and *Giardia* (oo)cyst viability has been evaluated by methods such as *in vivo* infectivity (Hoff et al., 1985), *in vitro* excystation (Bingham et al., 1979; Hoff et al., 1985; Smith and Smith, 1985), the presence or absence of specific internal morphological features (Schupp and Erlandsen, 1987b), fluorogenic dye staining methods (Jones and Senft, 1985; Schupp and Erlandsen, 1978a; Hudson et al., 1988; Ongerth et al., 1989), molecular techniques (Mahbubani et al., 1991; Abbaszadegan et al., 1997) and biophysical techniques (Smith, 1995). *In vitro* excystation and *in vivo* animal infectivity models are impractical due to the low concentrations of (oo)cysts present in water (Sauch et al., 1991). Animal infectivity models provide a direct measure of (oo)cyst viability, but are very time consuming and unsuited to routine laboratory analysis. Subsequently, good correlation between animal infectivity models and *in vitro* excystation have favoured the use of the latter (Labatiuk et al., 1991).

However, the use of fluorogenic dye staining techniques and differential interference contrast (DIC) microscopy are techniques that have shown the most promising results for assessment of viability of (oo)cysts in water (Campbell et al., 1992). In the former technique, when incident UV-illumination is used, microorganisms tagged with the specific antibody and fluorescent dye can be located by their bright fluorescence in contrast to a dark background (Sauch, 1985). The combination of fluorescein diacetate (FDA) and tetramethyl red (TMR), as well as propidium iodide (PI) and fluorescein isothiocyanate (FITC) are examples of fluorogenic staining techniques that have been used for viability assessment of *Cryptosporidium* and *Giardia* (Jarmey-Swan et al., 2000; Thiriat et al., 1997).

6.5.1.2. Materials and Procedures

Viability assays included all or some of the following:

1. *In vitro* excystation (Upton et al.1994 and Current and Box, modified by Waterborne Inc.2000)
2. Fluorescein diacetate (FDA) and Tetramethyl red (TMR) staining (Jarmey-Swan et al, 2000)
3. Propidium iodide (PI) and Fluorescein isothiocyanate (FITC) staining (Dowd and Pillai, 1997, and Waterborne Inc. 2000)
4. Differential interference contrast (DIC) microscopy (to confirm results of fluorogenic dyes)

In the first half of the project viable samples of *Cryptosporidium* oocysts and *Giardia* cysts were obtained from Sterling Parasitology Laboratory. Viability assays were performed before and after disinfection treatment and microscopic examination was carried out using the Zeiss Axioplan fluorescence microscope. Owing to problems encountered as described below, in the second half of the project, a new Olympus fluorescence BX51 microscope was used and new viable oocysts sourced from Waterborne Inc., USA. All new immuno-fluorescence labels and flurogenic dye reagents were sourced and prepared from scratch.

6.5.2. Assessment of Disinfection Efficacy

In all cases, whether assessing disinfectants used alone or in conjunction with other disinfectants, a contact time of 30 minutes was used, unless otherwise stated, after which any disinfectant residual present was removed by the addition of a small amount of sodium thiosulphate. When using UV irradiation only, thiosulphate was not added to the water after the half hour contact time, since no measurable disinfectant residual remained, only in cases where UV irradiation was used in conjunction with other disinfectants with a measurable residual, was thiosulphate added at the end of the 30 minute contact time. The samples were then poured into sterilised bottles and submitted for *E. coli*, coliform, faecal streptococci and coliphage analysis. In some cases samples of the effluent prior to treatment and also after treatment were also analysed for pH, turbidity, temperature, conductivity, alkalinity, calcium, magnesium, total hardness, iron, manganese, total dissolved solids, suspended solids, chemical oxygen demand (COD), TOC, DOC, BDOC, THMFP and UV absorbance at 254 nm. A number of samples were also analysed for *Cryptosporidium* oocysts and *Giardia* cysts both before and after disinfection.

6.5.3. Chlorine Demand Tests and Assessment of Chlorine Disinfection

The disinfection efficiency of chlorine (hypochlorite solution) was used for comparative purposes, so in all cases chlorine demand tests and assessment of chlorine disinfection were carried out in conjunction with the other disinfectants used. Chlorine demand tests were conducted using available chlorine concentrations of between 0 and 20 mg/*l*. A commercial sodium hypochlorite solution (BDH 10-14% m/v available chlorine) was used for chlorine dosing. This solution was standardised according to the method in Standard Methods for the Examination of Water and Wastewater (1998) (section 2350B(g)), using 0,01 N sodium thiosulphate. The chlorine demand tests were carried out according to section 2350B of Standard Methods for the Examination of Water and Wastewater (1998) using the DPD test for free and total chlorine concentration and a Lovibond comparator, and measuring the free and total chlorine concentrations after a 30 minute contact period. The chlorine demand was determined by plotting the chlorine residual (total chlorine concentration) against the chlorine added and finding the point at which either the total chlorine concentration approaches zero, or the total chlorine residual starts increasing pro rata with the chlorine addition indicating the chlorine breakpoint and chlorine demand value.

6.5.4. Ozone Requirement/Demand Tests and Assessment of Ozone Disinfection

A schematic figure of the apparatus used for the ozone demand appears in **Figure 6.1**. Ozonation was carried out in a glass contact column with a capacity of approximately 10 L and 1,57 m in height, with an internal diameter of approximately 90 mm. A Sorbios laboratory ozone generator model GSG 1.2 capable of producing 1 g/h ozone was used to generate ozone from oxygen (>99.5% oxygen, < 10 mg/*l* moisture) at a pressure of 0,5 bar and a flow rate of 15 *l*/h. The apparatus consisted of glass, stainless steel or teflon with silicon tubing.

Ozone was introduced into the column through a sintered glass diffuser (number 1 diffuser) positioned at the base of the column. Gas exiting the column was fed through a potassium iodide trap before passing through a gas flow meter (Alexander Wright Model Number DM3 B). The contact column was calibrated by filling it with a solution of potassium iodide and passing a measured volume of ozone-containing gas (at ambient temperature and pressure) through the column. During ozonation the solution was recirculated from the bottom of the contact column to the top using a peristaltic pump. Ozone liberates iodine from potassium iodide and the amount of liberated iodine after ozonation was measured using an iodometric titration (Standard Methods for the Examination of Water and Wastewater, 1998). The process was repeated until at least three calibrations varying by not more than 5% in concentration had been obtained. It was then possible to calculate the amount of ozone-containing gas that would have to be added to the sample for a particular applied ozone dose.

After calibration of the equipment, the reaction column was thoroughly cleaned and 10,6 L of secondary clarified effluent was placed into the column. Ozone-containing gas was passed through the sample and the potassium iodide trap at the exit of the column was replaced after the addition of ozone in incremental amounts and the ozone concentration in each trap then determined using the iodometric titration (Standard Methods for the Examination of Water and Wastewater, 1998). The ozone residual of the effluent was also measured after each incremental ozone addition using the indigo trisulphonate method (Bader and Hoigne, 1981; Standard Methods for the Examination of Water and Wastewater, 1998). This allowed calculation of the actual ozone concentration transferred to the effluent at each applied ozone dose. When using this procedure the contact time obviously increases with increasing ozone concentration and the volume of sample in the column reduces slightly after each ozone dose, although this was compensated for by calculation.

Ozone demand or requirement is determined by plotting ozone dose against the ozone residual. Once the ozone demand has been satisfied straight line correlation is observed between the ozone dose and the ozone residual. If this line is extrapolated it intersects the abscissa at the ozone demand value (Roche et al, 1994).

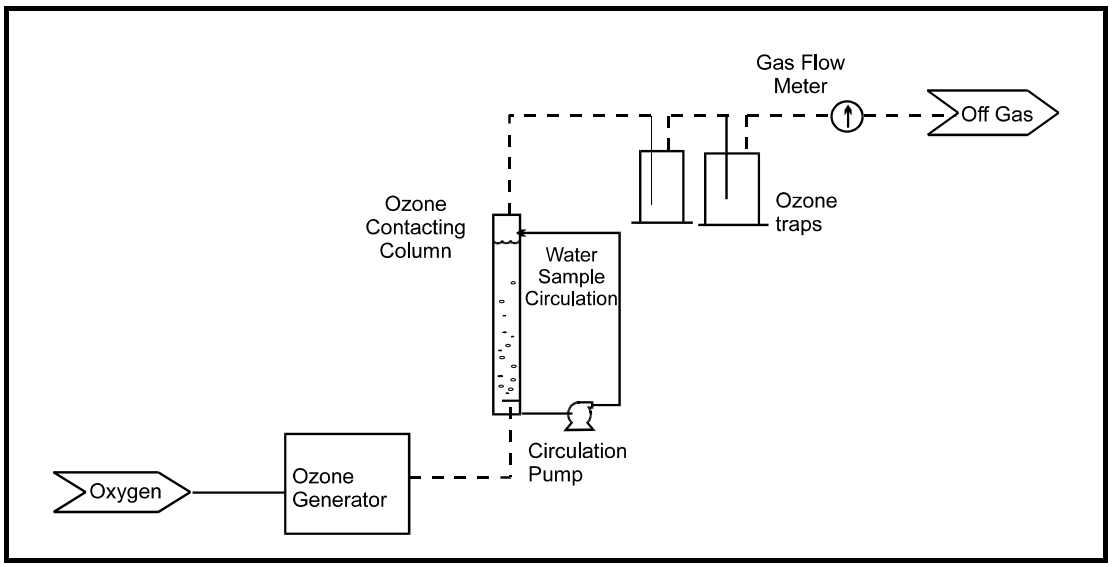


Figure 6.1: Flow Diagram of Laboratory Scale Ozonation Apparatus.

Samples were collected after each incremental ozone dose and a contact period of 30 minutes was allowed after sample extraction before adding a small amount of sodium thiosulphate to destroy any ozone residual that might still have been present. However, because of the procedure used, the actual ozone contact time increased with increasing ozone dose.

6.5.5. Disinfectant Demand Tests

Disinfectant demand tests were conducted for hydrogen peroxide and peracetic acid as well as for chlorine and ozone requirement tests. These tests were conducted as described above for chlorine demand, but dosing increasing increments of the required disinfectant, rather than of chlorine. The residual after a 30 minute contact time was then determined as for the chlorine demand test using the DPD tests for free and total chlorine.

6.5.6. Operation of Low Pressure UV Unit with Lamp Positioned above Effluent

Two low pressure UV systems were used in pilot plant tests. The first system consisted of a shallow pan constructed from highly reflective stainless steel with the low pressure UV lamp positioned just above the water level and not immersed in the water as is the more usual design. A submersible pump with a control valve was used to pump effluent at a flow rate matching that required by the UV unit to a feed tank. The effluent flowed from the storage tank through the unit under gravity and remained in the disinfection chamber for a period varying between 14 and 72 seconds, with flow rates of between 800 and 150 *l*/h and a nominal power consumption of 40 watts.

6.5.7. Operation of Low Pressure UV Unit with Lamp Immersed in Effluent

The second low pressure UV unit investigated consisted of a polycarbonate chamber surrounding a UV lamp encased in a quartz sleeve. The polycarbonate chamber had a convoluted design which forced the water closer to the lamp transversely at various points. A submersible pump delivered effluent to the unit and the flow was controlled by a regulating valve. The unit was operated in both upflow and downflow modes using flow rates varying between approximately 1800 L/h and 800 L/h giving rise to residence times in the unit of between 5 and 12 seconds. Only a flow rate of approximately 1800 L/h was used in the downflow mode and it was then necessary to restrict the outflow from the unit in order to keep the UV irradiation chamber completely filled. According to the manufacturers, added benefits of *mixed oxidants* due to air being drawn in through a venturi would be gained by operating the unit in this mode. The UV lamp in this unit had a nominal power consumption of 6 Watts.

6.5.8. Operation of Low Pressure UV Unit in Laboratory Batch Tests

The low pressure UV unit used in the continuous flow through tests described in **section 6.5.7** was also used in batch mode for laboratory tests. It was operated in batch mode so that the wastewater effluent could be exposed to different UV doses based only on time, without the need to vary the flow rate. When testing the disinfection efficiency of the UV irradiation emitted by this unit, it was completely filled with secondary clarified effluent and then the unit was switched on for a specified time. Thereafter, samples of water both before and after UV irradiation as described above, were analysed.

6.5.9. Medium Pressure UV Unit

A medium pressure UV unit was used in pilot plant tests to determine the efficacy of various disinfectants for wastewater disinfection. The medium pressure UV unit used was a Photon PMD 100A1/2 supplied by Hanovia Limited with a capacity of between 300 mJ/cm^2 (at 1,4 m^3/h) and 32 mJ/cm^2 (at 13 m^3/h) and a nominal power consumption of 0,6 kW. It required a flow rate of between about 1,4 and 13 m^3/h and was microprocessor controlled, displaying UV intensity (% and mW/cm^2), UV dose (mJ/cm^2 or mW.s/cm^2), flow rate, water temperature and lamp time.

6.5.10. Multiwave UV Unit

Pilot plant tests to investigate the disinfection efficacy of medium pressure UV irradiation were also conducted using a Multiwave UV unit. A Berson In-Line HXFS (W) 1Multiwave medium pressure UV unit was used in place of the Hanovia PMD 100 A1/2 unit for some of the medium pressure UV pilot plant tests. This unit had a manual wiper so that the UV tube could be cleaned as required while the UV unit was in operation. The unit was calibrated to produce a UV dose of 25 mJ/cm^2 at a flow rate of 22 m^3/h for a water/effluent with a transmittance of 100%. The dose could be adjusted according to a calibration table provided by the suppliers. For example a dose of double that above could be achieved by reducing the flow rate by half. A control panel on the unit listed the power level, hour counter, flow rate and UV absorbance/ % transmittance of the water.

6.5.11. Peracetic Acid and Peracetic Acid with Hydrogen Peroxide

Peracetic acid was obtained from Fluka as an approximately 39% assay in acetic acid. The peracetic acid was standardised using an iodometric titration procedure described in Vogel's Textbook of Quantitative Inorganic Analysis (1993) under the procedure for benzoic acid. Hydrogen peroxide was also standardised using an iodometric procedure described in this textbook. Peracetic acid was added to secondary clarified wastewater effluent at concentrations of up to 60 mg/L. A mixture of peracetic acid and hydrogen peroxide was prepared containing 150 g/L peracetic acid and 230 g/L hydrogen peroxide as described in an Air Liquide brochure (1997). Wastewater effluent was then dosed with this mixture using peracetic acid concentrations of up to 60 mg/L with a contact time of 30 minutes. The effluent both before and after disinfection at the various concentrations was analysed for the bacterial indicator organisms. Tests using sodium hypochlorite solutions were also conducted on the wastewater effluent for comparative purposes. All these tests were carried out in the laboratory.

Pilot plant tests were also conducted using peracetic acid on the pilot plant described in **section 6.5.15.** These tests were carried out using a peracetic acid solution produced according to a procedure described in Vogel's Practical Organic Chemisty by adding 1 part 30% percent hydrogen peroxide to 3 parts glacial acetic acid in the presence of a catalytic amount of sulphuric acid. The concentration of the peracetic acid solution was then determined as described above. Peracetic acid was added to the effluent in various quantities based on the chlorine demand. Peracetic acid was used in concentrations that were equivalent to those being used for chlorine disinfection in terms of mg/L.

Both laboratory and pilot plant tests were conducted using peracetic acid in conjunction with UV and ozone.

6.5.12. Bromine

A bromine solution, consisting of potassium hypobromite and supplied by Israeli Irrigation Co-ordinators was used in disinfection tests. The total bromine concentration was determined using an iodometric titration described in **sections 6.4.1** and **6.4.2**. Available bromine forms release iodine quantitatively at pH 4 (White, 1992).

Bromine was used at concentrations equivalent to those being used for chlorine disinfection in terms of mass concentration. Bromine was also used in conjunction with medium pressure UV irradiation and ozone.

6.5.13. Mixed Oxidant Generator

A mixed oxidant generator was obtained from a company known as Radical Waters. According to the manufacturers this unit can produce up to 40 L/h of an anolyte stream which has disinfection capabilities superior to those of a hypochlorite generator stream, since it is claimed to contain oxidants other than hypochlorite such as peroxide compounds and radicals, ozone, hydrogen peroxide and metal oxide hydrates. The unit was operated at an anolyte flow rate of approximately 700 mL/min, using a pressure of approximately 0,5 bar (this was determined by the flow rate) and using a 5% sodium chloride solution to supply the unit. Tests using a 5% sodium sulphate solution were also conducted in an effort to determine the effect of mixed oxidants other than chlorine species.

The anolyte stream was standardised using the iodometric titration procedure described in **section 6.4.1** (Standard Methods for the Examination of Water and Wastewater, 1998). This

titration measures all oxidising species, but it was not possible to speciate the various oxidants present in the anolyte stream. The anolyte was then dosed into secondary clarified wastewater effluent at various doses and allowed to react for 30 minutes prior to addition of sodium thiosulphate. The bacterial indicator organisms present in the water before and after disinfection were measured and for the purposes of comparison, the same wastewater effluent sample was also treated at equivalent sodium hypochlorite concentrations, as determined from the iodometric titration procedure, and the effluent before and after chlorination analysed for the various bacterial indicator organisms.

6.5.14. Klor-Free

Klor-Free is the trade name of a disinfectant manufactured by Master Pool Services containing potassium persulphate (500 g/kg) and copper sulphate (25 g/kg) as the active ingredients, as well as ammonium chloride (approximately 40%) and boric acid (approximately 10%). It is intended for swimming pool use, but claimed by the manufacturers to also be suitable for potable water treatment applications. It is supplied as a powder, and since it is easy to apply, it was assessed as it might have applications for small rural applications. The Klor-Free was dosed into secondary wastewater effluent at concentrations of up to 100 mg/L using contact times of 30 minutes and 4 hours after which the effluent both before and after disinfection was analysed for the various bacterial indicator organisms. Only laboratory scale tests were conducted.

6.5.15. Pilot Plant

A pilot plant was constructed in order to assess the disinfection efficacy of ozone and medium pressure UV, separately, together and in conjunction with other disinfectants such as chlorine, peracetic acid and bromine. The size of the pilot plant was determined by the medium pressure UV unit which required a flow rate of between about 1,4 and 13 m^3/h. The ozone generator was a Prosep M30G model supplied by O_3 Ozone Systems, capable of producing up to 30 g /h ozone at an oxygen flow rate of 10 L/minute. Initially the ozone generator consisted of three water-cooled glass dielectric tubes, but these were later replaced with two air-cooled ceramic dielectric tubes due to problems experienced with cooling water flow and the glass tubes breaking, but this did not have any significant impact on the ozone output of the generator.

A schematic of the pilot plant is shown in **Figure 6.2**. Pilot plant tests were conducted on secondary treated wastewater effluent taken from the secondary overflow channel of the Darvill Wastewater Works, a works with a nominal design capacity of approximately 75 ML/day treating a predominantly domestic wastewater. The effluent was pumped out of the

overflow channel using a Grundfos multistage in-line centrifugal pump through a sieve and non-return valve system. When using ozone, this was injected into the system through a venturi prior to an in-line static mixer. This venturi could also be used for the addition of other disinfectants, provided that ozone was not being used. Ozonated water entered a separator or contact column with a total capacity of approximately 500 L, although it was generally only filled to about 250 L. Unreacted ozone and oxygen was separated from the water in the ozone separator/contactor tank from which it was passed through a thermal destructor before being vented to atmosphere A series of valves allowed the water to be diverted through the medium pressure UV unit prior to discharge. Sampling taps throughout the pilot plant allowed for sampling at various stages in the process.

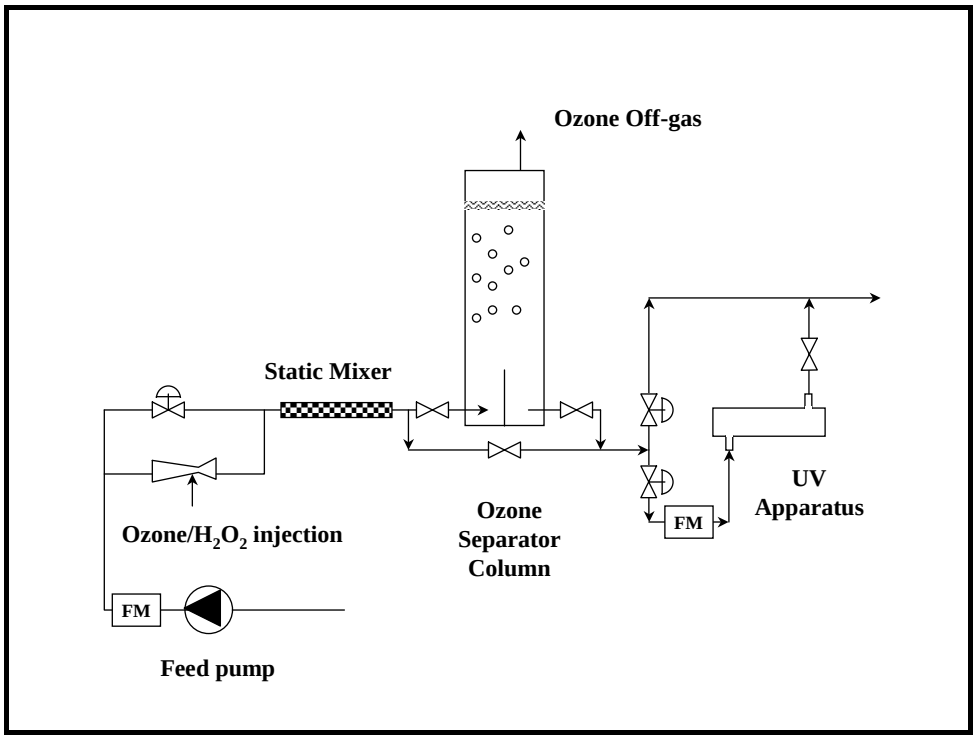


Figure 6.2: Flow Diagram of pilot plant consisting of ozonation and medium pressure UV facilities.

6.5.16. Tracer Tests

Tracer tests were conducted on the pilot plant using sodium hypochlorite and determining the total chlorine concentration of the effluent. A concentrated solution of sodium hypochlorite was introduced into the pilot plant through the venturi for a period of 5 seconds. Sampling at various points through the pilot plant started simultaneously with the addition of hypochlorite solution, with samples being collected every 5 seconds for a period of 45 seconds. The flow rate through the plant and the uptake rate of the venturi were recorded. These tests were

conducted for the Multiwave UV unit, the ozone contact/separator column and the medium pressure UV system.

7. Photographs

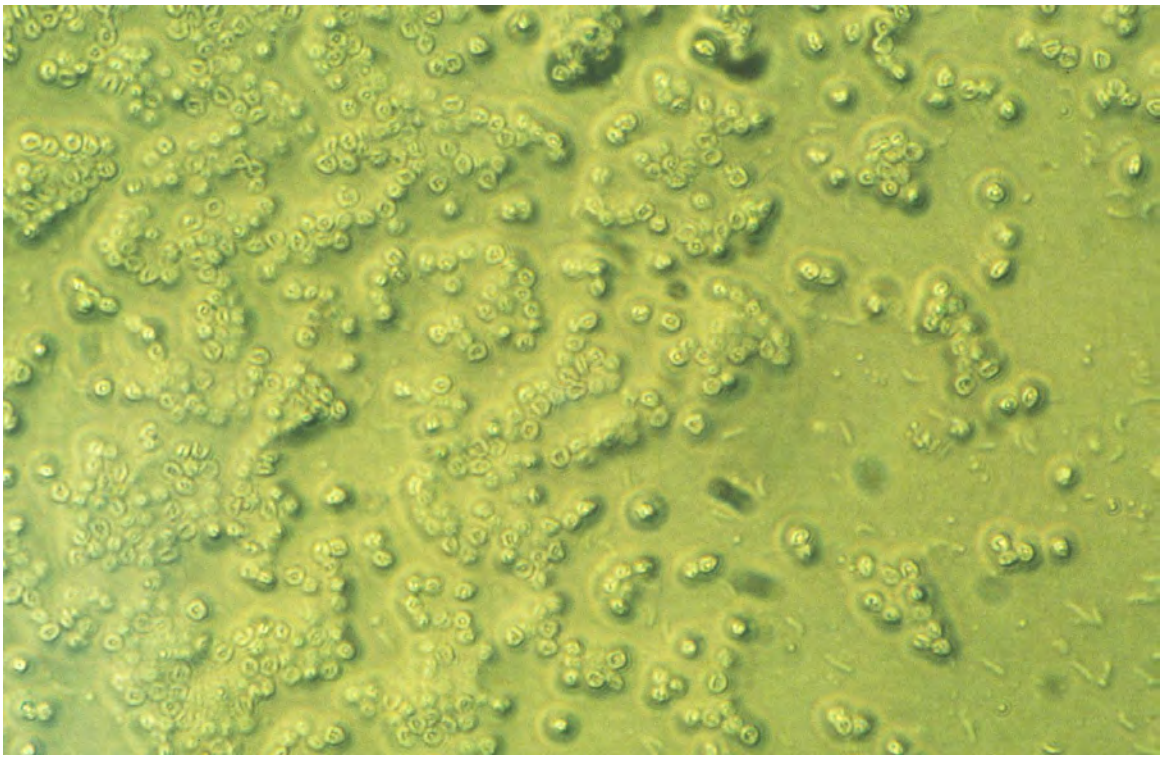
7.1 Laboratory Ozonation Equipment

7.2 Pilot Plant

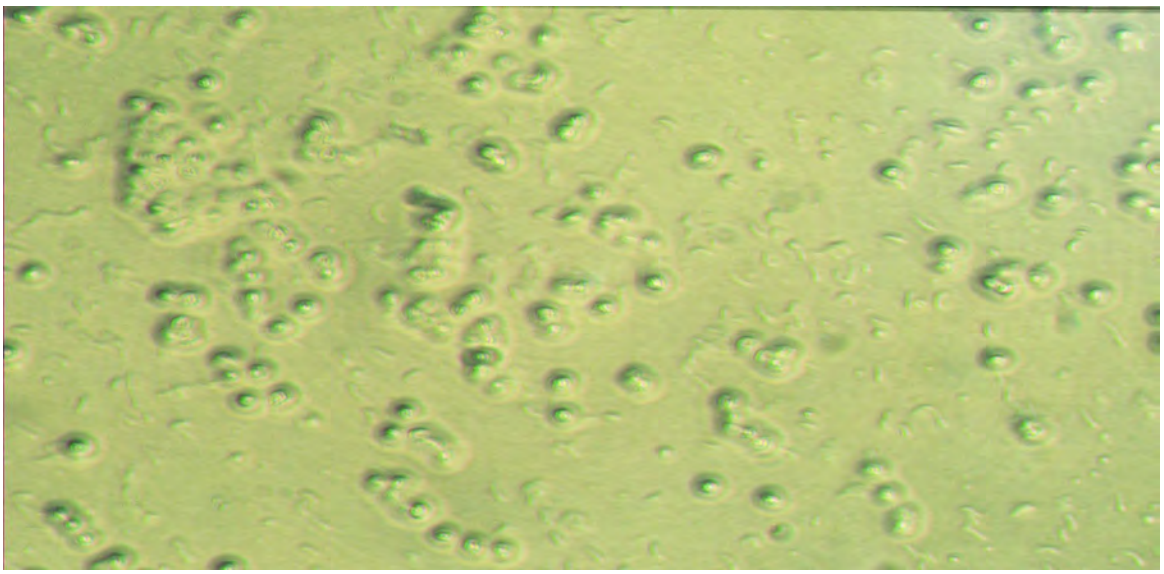
7.3 Medium Pressue UV Unit

7.4 Ozone Generator and Destructor

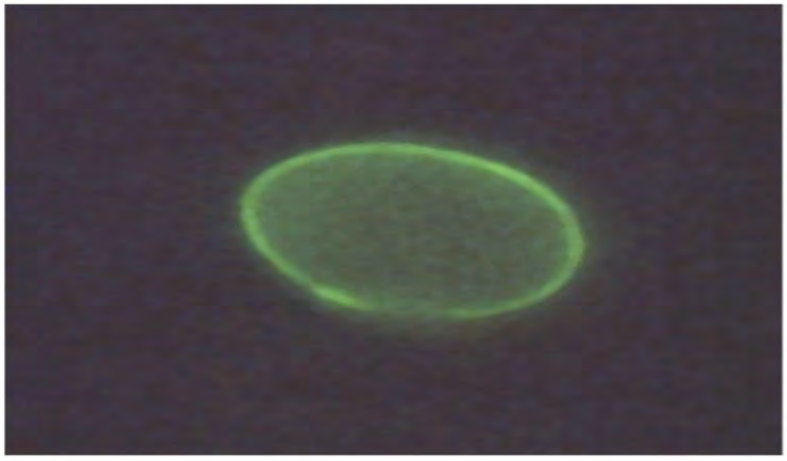
7.5 *Cryptosporidium Oocysts* Following In Vetro Eexcystation Viewed Under DIC

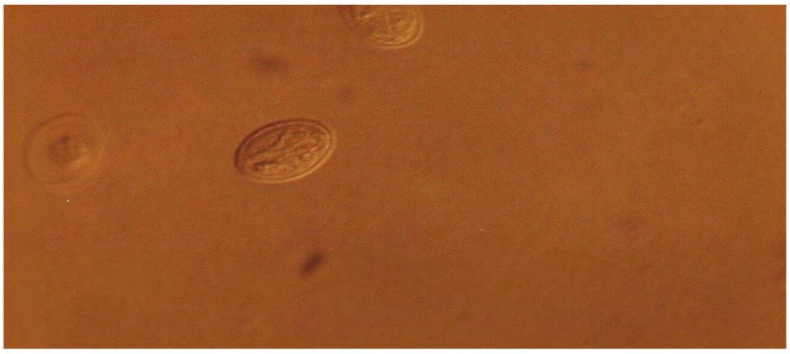


7.6 Crytosporidium Oocysts Following In Vetro Excystation Viewed Under DIC



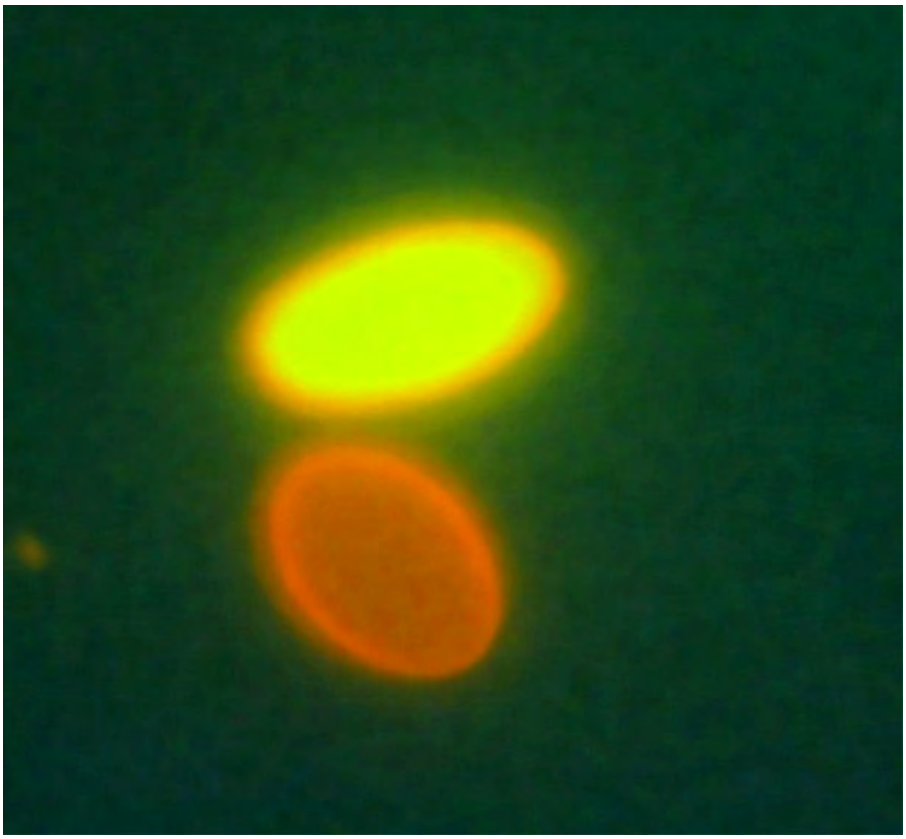
7.7 FITC Stained *Giardia Cyst*





Giardia Cyst Viewed Under DIC

7.8 TMR-FDA Stained *Giardia Cysts*



7.9 SEM of a *Cryptosporidium Oocyst*

7.10 SEM of a *Giardia Cyst*