The use of aqueous ozone for cleaning operations in breweries

Ozone clean in place in food industries - ozone as an alternative sanitising agent for cleaning operations in food industry

Miguel Ángel Prieto Arranz, Dr. G. Schories

Technologie Transfer Zentrum Bremerhaven, Umweltinstitut, An der Karlstadt 6; D-27568 Bremerhaven

Abstract

The production of beer is characterized by the consumption of large volumes of fresh water. Whereas, cleaning operations are the highest water consumption demanding activities in breweries. Commonly “Cleaning In Place” (CIP) systems are used in closed processing lines. These consist of automatic cleaning and disinfection programs based on a succession of several solutions of water, cleaning chemicals and disinfection agents. Typical disinfection chemicals are based on Chlorine, other halogens or H_2O_2. These chemicals are discharged into sewer systems together with large amounts of water necessary to rinse out residual chemicals, which are being questioned for their unhealthy effects on humans. Ozone has the potential to replace these chemicals currently employed in the different CIP processes. Ozone is a powerful broad-spectrum antimicrobial agent that is active against bacteria, fungi, viruses, protozoa, and bacterial and fungal spores, by attacking various cell membrane, wall- and cell constituents, this is due to the high oxidation potential of ozone (2.07 V) far greater than that of other chemical compounds currently applied for cleaning and disinfection operations in the brewing industry. The European research project OZONECIP, aims to demonstrate the environmental advantages of the use of ozone instead of the conventional chemicals used in “Cleaning In Place” operations in different food processing industries. In order to achieve these objectives a prototype will be designed, built and prepared for assaying CIP processes which will allow obtaining environmental indicators and values for its evaluation. The prototype enables the simulation of industrial CIP processes carried out in breweries (also in diaries and wineries) and essay equivalent processes based in ozone.

Key-words: Ozone, cleaning operations, brewing industry,

Introduction and current situation in breweries

The production of beer is characterized by the consumption of large volumes of fresh water. Breweries use almost all the water consumed for cleaning and disinfection operations, thus becoming wastewater effluents. It has been reported, that a brewery needs about 2.5-6 hl water/hl beer sold (Rüffer and Rosenwinkel 1991). The most important cleaning tasks are related to the washing of process vessels, tanks and the net of pipes that are involved in the production process. Commonly “Cleaning In Place” systems (CIP) are used, these are characterized by automatic cleaning programs based on a succession of several solutions of water, cleaning chemicals and disinfection agents. Typical disinfection chemicals are based on Chlorine, other halogens or H_2O_2. These chemicals are discharged into sewer systems together with large amounts of water necessary to rinse out residual chemicals, which are being questioned for their unhealthy effects on humans.

Current practices for cleaning and disinfection in breweries vary according to the target surface:

- Closed equipment: Cleaning In Place processes are the most employed procedure for cleaning and disinfection operation in breweries. It is applied in closed processing lines, such as pipes, vessels and tanks.
- Open surfaces: The cleaning and disinfection operations of open surfaces in breweries, such as e.g. fillers or conveyors in the bottling cellar, are usually performed using low-pressure foam systems or thin film cleaning, following the cleaning and disinfection pattern showed in Table 1. The use of hot solutions or strong chemicals is in many cases limited for safety reasons. (Holah, 1992).

Action	Agent
Prerinsing	Water
Foaming	Foam cleaner
Soak time	Foam cleaner
Intermediate rinsing	Water
Spraying	Disinfectant solution
Final rinsing	Water

Table 1: Foam cleaning and disinfection programme

During the production of beer, several microorganisms are likely to grow because of the nutrient-rich environment they are exposed to. On the other hand, the characteristics of beer: alcohol, carbon dioxide and sulphur dioxide as well as low pH reduce the range of organisms most likely to be found to relatively few species, among others, *lactobacillus (Lactobacillus brevis, L lindneri, L. brevisimilis, L. frigidus, L. coryniformis, L. Casei, L. pediococcus damnosus and L. pediococcus incotinatus) pectinatus cerevisiiphilus* or different yeasts cultures. These organisms may grow in beer causing off-flavours, turbidity and precipitates. (Storgards, 2000) Besides, biofilms are an additional source of contamination for the whole food and beverage sector. The formation of biofilms takes place when a solid surface comes into contact with a liquid medium in the presence of microorganisms. In the food industry, biofilms could be defined as a layer of bacteria on surfaces covered and protected by extracellular polymeric substances. Biofilms mainly develop gradually in areas, which are hard to access by cleaning and disinfection operations (Rinsing, cleaning and disinfecting processes). Organisms growing as biofilms are far more resistant towards these operations than free cells, making such deposits even more difficult to remove, accumulating on floors, wastewater and freshwater pipes, bends and dead ends in pipes and stainless steel surfaces.

Therefore, hygienic design practices are crucial aspects to be taken into consideration in breweries, in order to ensure that all hygienic standards are satisfied, guaranteeing the quality of the final product. These practices comprise suitable selection of equipment, materials and accessories, while warranting the correct construction, process layout and process automation of the installation. The equipment design should take into consideration that all surfaces which will be in contact with the beer or its intermediates are easily accessed for cleaning and disinfection purposes, being smooth and avoiding crevices, pits, edges or blind ends.

It was reported, that chemical cleaners have been found to be more effective in eliminating attached bacteria from surfaces than disinfectants. Furthermore, a complete removal and disinfection of attached biofilms will be only achieved once the surface is initially satisfyingly cleaned before the disinfectant is applied. As a consequence, removal of adherent microorganisms or biofilms should involve first cleaning followed by disinfection.

For choosing adequate disinfectants in breweries, the following characteristics should be considered (Donhauser et al. 1991):

- Effectiveness against Gram-positive and Gram-negative bacteria and against yeasts and moulds
- Effectiveness in the presence of proteins
- Effectiveness at low temperatures
- Wetting ability
- CIP-suitability
- Environmental aspects such as easily rinsable, readily biodegradable, etc.
- Economy: effectiveness at low concentrations, reusable, easily rinsable
- Health aspects: safe to use
- Product compatibility: no adverse effects on the product.

The brewing industry is currently making use of different types of disinfectants such as: hydrogen peroxide, halogens or halogenated carbonic acids, alkylamines, biguanides, chlorine dioxide, etc.

These chemicals are discharged into sewer systems together with large amounts of water necessary to rinse out residual compounds, which are being questioned for their unhealthy effects on humans. Ozone has the potential to replace these chemicals currently employed in the different CIP processes and in further cleaning and disinfection operations in breweries.

Ozone as an alternative sanitising agent

Ozone is one of the most powerful disinfectants available. It has been used for water disinfection since the early 1900s. Ozone is a very efficient broad-spectrum antimicrobial agent that is active against bacteria, fungi, viruses, protozoa, and bacterial and fungal spores, by attacking various cell membrane, wall- and cell constituents. The antimicrobial action of ozone is based on its high oxidation potential (2.07 V) far greater than that of other chemical compounds currently applied as disinfectants, such as H_2O_2, HOCl, Cl_2, ClO_2 or I_2. Therefore high disinfection efficiency is expected against microorganisms typically found in beer processing plants. Moreover, ozone appears to be an alternative which would also solve the key problems found in the most widespread disinfection technologies, such as hazardous by products or microorganisms resistance. Complementing this effectiveness, is the fact that ozone, unlike other disinfectants, leaves no chemical residual and degrades to molecular oxygen upon reaction or natural degradation. This, on the one hand ensures that no by-products are present in the beer, which could cause taste or odour in the final product, while on the other hand leads to better wastewater quality, as it contains additional oxygen enhancing the performance of biological wastewater treatment processes. Additionally the use of aqueous ozone for disinfection purposes, increases the water recycle and reuse possibilities in breweries. Provided adequate microbiological control, the ozonated disinfection rinse can be re-used in e.g. for the initial rinse sequences in CIP processes. Ozone also enables energy savings as it is normally applied at low temperatures, this leads to an increase of ozone's half-life and to a decrease in its natural conversion back to Oxygen. As it is generated on-site, ozone avoids the need of storing hazardous chemicals, which could cause accidents endangering human and environmental health and safety. (Lagrange, 2004)

On the other hand, it has to be considered, that ozone is a toxic gas, being harmful to health even at low concentrations. Therefore the ozone concentration should be monitored in the workplace when it is used to disinfect equipment and installations. The toxicity of ozone varies, depending on its concentration and the length of exposure. Ozone irritates eyes, skin and throat and can lead the severe damages. Special attention is required when using ozone in the workplace, as harmful concentration in the air will be reached very quickly on loss of containment. Consequently, ozone ambient monitoring devices, ventilation, local exhaust, cold-insulating gloves, face shields, or eye protection in combination with breathing protection are some recommended measures to take at the workplace. In the United States, the current permissible level for ozone exposure in the workplace was set at 0,1 ppm by the Occupational Safety and Health Administration (OSHA). This is the concentration to which a worker may be continuously exposed to the gas under normal working conditions (8 h a day or 40h per week) without adverse effects. In Europe this value may vary among the different member states, whereas in Germany this maximum permissible level, called MAK-Wert, was also set at 0,1 ppm of ozone in the working place. However, ozone is readily detectable by human smell at 0,01 to 0,04 ppm concentration in air, far below the maximal permissible level for ozone exposure, and ozone is not listed as a carcinogen.

Ozone is a strong oxidising agent with high corrosion potential, which could cause severe damages to the brewing processing equipment. Although the chemicals compounds currently used for the cleaning and disinfection operations also possess strong oxidising and corrosive properties, the reactivity of ozone and its high oxidation potential is even higher, and should be taken into consideration. Therefore all materials in contact with the ozone enriched solution should have good compatibility with ozone. Not only because of the eventual corrosion of the equipment's surface, but also because of losses in the concentration of the ozone enriched water, with ozone reacting with the equipment's surface. The corrosive effects of ozone in contact with different materials, depends mainly on the applied ozone concentration. Stainless steel is the most common surface currently used in CIP systems and in production equipment in the brewing industry. Most of them are compatible with a continuous ozone use at moderate ozone concentrations of about 1 – 3 ppm dissolved in water. It has been reported, that stainless steel type 316L (X2CrNiMo18-14-3) is the material of choice when considering corrosion of materials in contact with ozone enriched solutions. Consequently it is advisable to identify all materials that should come in direct or indirect contact with ozone and check their corrosion resistance towards ozone.

The OZONECIP project

The three year's European research project OZONECIP, which started in December 2005, aims to demonstrate the environmental advantages of the use of ozone instead of the conventional chemicals used in "Cleaning In Place" operations in the food processing industry. In order to achieve these objectives a prototype will be designed, built and prepared for assaying CIP processes which will allow obtaining environmental indicators and values. The prototype consists on three subsystems: CIP-subsystem, which is constructed following the traditional CIP system's design; the ozone-subsystem, which is responsible for the production of the ozone and its dilution into the water and the target-subsystem, which will simulate different parts of the processing equipment. The prototype will allow to simulate industrial CIP processes carried out in breweries (also in diaries and wineries) and essay equivalent processes based in ozone. Several trail series will be carried out with different products and equipment surfaces, to obtain qualitative and quantitative indicators regarding environmental and non-environmental aspects. Such study at pilot scale is necessary in order to collect all the necessary data under reproducible conditions with a selected number of variable parameters of operation being monitored for the establishment of quantitative indicators (water consumption, volume of wastewater produced, chemical characterisation of the wastewater, disinfection efficiency, etc.)

Within this scope, the first trial series will be carried out with the OZONECIP prototype following two reference CIP sequences, which represent the most applied CIP processes currently applied in breweries. Following table shows the first reference CIP sequence to be essayed.

Action	Temperature	Duration
Prerinsing	Cold	5min
Alkaline cleaning	Cold	10 min
Intermediate rinsing	Cold	5 min
Acidic cleaning	Cold	5 min
Intermediate rinsing	Cold	5 min
Disinfection	Cold	5 min
Final rinsing	Cold	5 min

Table 2: First reference CIP sequence to be essayed

A second CIP sequence will follow the same pattern but avoiding the alkaline cleaning cycle. This is due to the fact, that in breweries, all equipment that is likely to contain a CO_2 atmosphere, such as fermentation tanks and pipes, storage tanks, filtration tanks, etc. are mainly cleaned using CIP cleaning sequences avoiding the use of the alkaline chemicals, while acid-based compounds are preferred because of the following practical advantages:

1) Acids are not affected by carbon dioxide atmospheres, so they do not loose their cleaning efficiency when used on a recovery system.
2) Acids prevent carbon dioxide losses by allowing cleaning and disinfection without the need of tank ventilation, facilitating carbon dioxide top pressure cleaning.
3) Acids reduce the risk of tank implosion.
4) Acids are efficient in removing and preventing beer stone and hard water deposits.
5) Acids reduce water consumption as they are easily rinsed away.
6) Acids are energy efficient because hot cleaning is not necessary.

However, sodium hydroxide has shown to have high cleaning efficiency and a high antimicrobial activity against organisms likely to be encountered in breweries. Therefore, alkaline based CIP sequences are in many cases unavoidable and have to be run. Although this practice is carried out in general with a lower frequency than the "acid-only" CIP sequence. Following table shows the second reference CIP sequence that will be essayed with the OZONECIP prototype.

Action	Temperature	Duration
Prerinsing	Cold	5min
Acidic cleaning	Cold	30 min
Intermediate rinsing	Cold	5 min
Disinfection	Cold	5 min
Final rinsing	Cold	5min

Table 3: Second reference CIP sequence to be essayed

In a second stage of the first trials series, ozone enriched solutions will replace the different rinses carried out in the reference CIP processes essayed (Table 2 and 3). The prototype itself and the different trials were designed in order to allow the progressive introduction of ozonated rinses in the subsequent ozone based CIP sequences to be essayed. Disinfection cycles and the different pre-, intermediate and final rinses will be tested using different ozone concentrations, and cycle durations, evaluating its performance and efficiency towards the reference cycles.

The trials will be carried out using diverse types of polluted beer. The beer samples will be polluted with different type of microorganisms, likely to be found in beer processing plants, and in order to get reliable and accurate results, they will be essayed separately. The trials will be carried out in a first stage using different types of lactobacillus; *pectinatus cerevisiiphilus* and different yeast cultures.

During the running of the trials several aspects are to be monitored accurately in order to allow its reproducibility and evaluation. The trails' results will determine to which extend the different ozone based CIP sequences comply acceptably with the expected objectives and goals within the project's scope. Following parameters will be recorded:

a) Water consumption and wastewater production
b) Energy consumption
c) Duration of each cleaning cycle and total duration of each CIP sequence
d) Concentration and dose of the chemical compounds applied
e) Conductivity of the cleaning solutions at the target sub-system's inlet and outlet
f) Ozone concentration in the ozone sub-system's buffer tank
g) Ozone concentration at the target sub-system's inlet and residual ozone concentration at the target sub-system's outlet
h) Microbiological parameters and disinfection efficiency

Based on the results and initial tendencies of these first trail series, further trail series will be designed and essayed, in order to adapt and improve the CIP sequences essayed. This will allow to best define the different environmental indicators and representative values to update European and national *Best Available Technology* (BAT) reference documents, which is also one of the project's main objectives. Non-environmental factors that could affect the applicability of the ozone CIP alternative will also be essayed, suggesting solutions to overcome them. Models of technical and economical viability will be generically defined to facilitate the large scale implementation in the food and beverage industry.

Apart from CIP processes, ozone can also be applied in the brewing sector for pre-treatment and disinfection of the brewing process water, replacing the chemicals applied in bottle rinsers, it can also be employed in Pasteur tunnels or even for the cleaning and disinfection of walls, floors, tables and conveyors. However, and due to the toxicity of ozone, these practices have to be performed in adequate way under controlled conditions, in order to comply with the regulations and the maximal permissible exposure level of ozone at the working place.

Several publications have already stated the disinfection efficiency of aqueous ozone on different surfaces; some of them are listed in table 4:

Application	Aqueous Ozone Treatment	Targeted microorganisms	Disinfection efficiency	Author, year
CIP system	Not specified	Staphylococcus aureus, Pseudonomas aeruginosa, Candida albicans	99% microbial count reduction	Lagrange, Reiprich & hoffmann, 2004
Dairy biofilms on stainless steel surfaces	0,5 ppm O_3 for 10 min	Pseudomonas fluorescens, Alcaligenes faecalis	5,6 log and 4,4 log reduction respectively	Greene, Few, Joao & Serafini, 1993
Stainless steel mixing kettle	2 ppm O_3 for 1 min	Not specified	Microbial plate count reduction from 63,1% to 99,9%	Hampson, 2000
Floor areas	2 ppm O_3 for 1 min	Not specified	Microbial plate count reduction from 67,0%-95,6% (High traffic floors) and 84,3%-99,9% (Low traffic floors)	Hampson, 2000
Equipment, walls, floors, drains, tables and conveyors	3 - 3,5 ppm O_3	Trichophyton mentagrophytes, Salmonella choleraesuis, Staphylococcus aureus, Pseudonomas aeruginosa, Campylobacter jejuni, Listeria monocytogenes, Aspergillus flavus, Brettanomyces bruxellensis, E. Coli	Reduction ranging from 4 log – 6 log	Boisrobert, 2002

Table 4: Publications stating different disinfection efficiencies using aqueous ozone

Based on this table it can be expected, that moderated ozone concentrations, ranging from 0,5 ppm to 3,5 ppm are capable to reach remarkable disinfection efficiencies, ranging up to a 6 log disinfection efficiency (Pascual et al. 2006). However, it has to be taken into consideration the quite diverse values observed (ozone concentration and contact times versus disinfection efficiency), which indicates the varied effectiveness of ozone against the different specific microorganisms targeted, and its strong dependency regarding its concentration, contact time and surfaces types on which it is applied. Therefore, no definitive conclusions or tendencies can be drawn prior the finalisation of the trials series that are to be performed within the OZONECIP project. Based on these results the general pathway will be established, however tailor made study should be carried out in every brewery before an ozone based CIP or other ozone based cleaning and disinfection system is run.

Conclusion and outlook

Based on the information exposed in this paper it can be concluded that the application of aqueous ozone for cleaning operations in breweries, is a promising alternative to traditional disinfection agents currently use. Whereas the different trials series to be performed within the OZONECIP project, will contribute to test and evaluate the efficiency of aqueous ozone as alternative disinfectant in breweries.

Project Consortium
Ainia - Instituto Tecnologico Agroalimentario (Spain, project coordinator), Technologie Transfer Zentrum Bremerhaven, Umweltinstitut (Germany), Gdansk University of Technology (Poland), Allied Domecq bodegas (Spain, wine processing), InBev. (Germany, beer processing) and Meierei-Genossenschaft e.G. Langernhorn (Germany, dairy processing)

Acknowledgements

The OZONECIP project is co-funded by the European Union’s LIFE Environment Programme, EC contribution 394,924.00 € (LIFE 05 ENV/E/000251).

References

1. Boisrobert, C. (2002). U.S. regulatory review of ozone use in the food industry, Ozone III: Agricultural & food processing applications of ozone as an antimicrobial agent.
2. Donhauser, S., Wagner, D. and Geiger, E. 1991. Zur Wirkung von Desinfektionsmitteln in der Brauerei. Brauwelt, Vol. 131, pp. 604–616.
3. Greene, A. (1993) A comparison of ozonation and chlorination for the disinfection of stainless steel surfaces. Journal of Dairy science, 76, 3617-3620.
4. Hampson (2000). Use of ozone for winery and environmental sanitation. Practical winery & vineyard, January-February, 27-30.
5. Lagrange F., Reiprich W., Hoffmann M., 2004. CIP-cleaning and disinfection with ozone water, Journal article, 84(2):112-114
6. Occupational Safety and Health Administration's (OSHA) Regulations for ozone. www.osha.gov
7. Pascual, A. et al., (2006) Use of ozone in food industries for reducing the environmental impact of cleaning and disinfection activities, Trends in food Science & Technology
8. Rüffer, H., Rosenwinkel, K.H., (1991)Taschenbuch der Industrieabwasserreinigung, Oldenbourg Verlag, München
9. “Toxicologisch arbeitsmedizinische Begründungen” Deutsche Forschungsgemeinschaft Band IV
10. Storgards, E. 2000. Process hygiene control in beer production and dispensing. VTT Biotechnology
11. Holah, J.T. 1992. Industrial monitoring: hygiene in food processing. In: Melo, L.F., Bott, T.R., Fletcher, M. and Capdeville, B. (eds.). Biofilms – Science and Technology. Kluwer Academic Publications, Dordrecht. Pp. 645–659.