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# The Use of Ozone as a Disinfectant in the Dairy Industry

### What is Ozone and How does it work

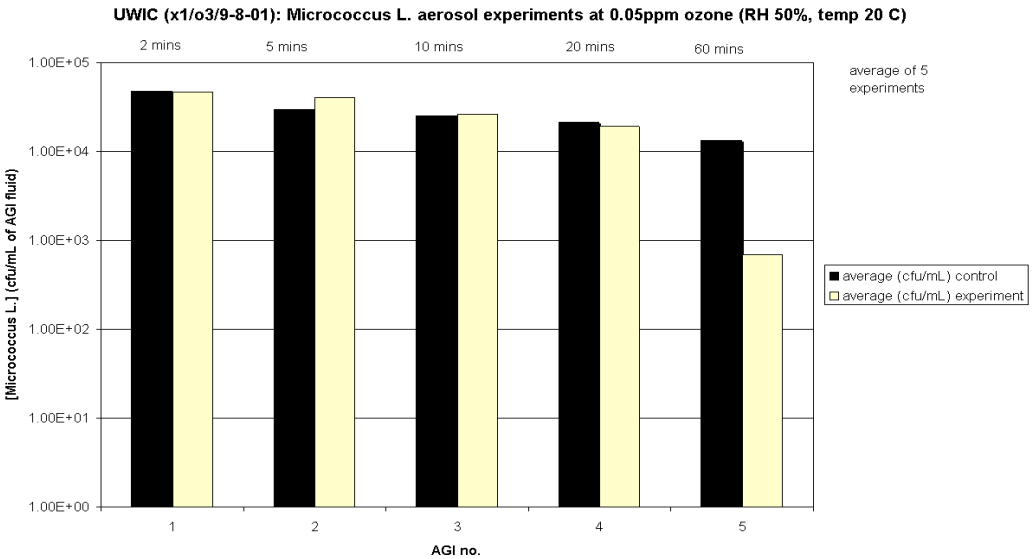
Ozone occurs naturally high in the atmosphere as a result of the action of ultra violet radiation from the sun acting upon the oxygen molecules. At lower levels it is created as a result of lightning and by the solar initiated photochemical processes that occur in polluted environments. Ozone can also be artificially generated by passing natural oxygen, which is a bi-atomic molecule ($O_2$), through a high energy electrical corona replicating the action of lightning. This raises the energy levels of some of the bi-atomic oxygen molecules causing them to dissociate into free oxygen atoms. These free atoms then loosely attach themselves to other unchanged bi-atomic molecules creating unstable tri-atomic oxygen ($O_3$) that is more commonly called ozone.

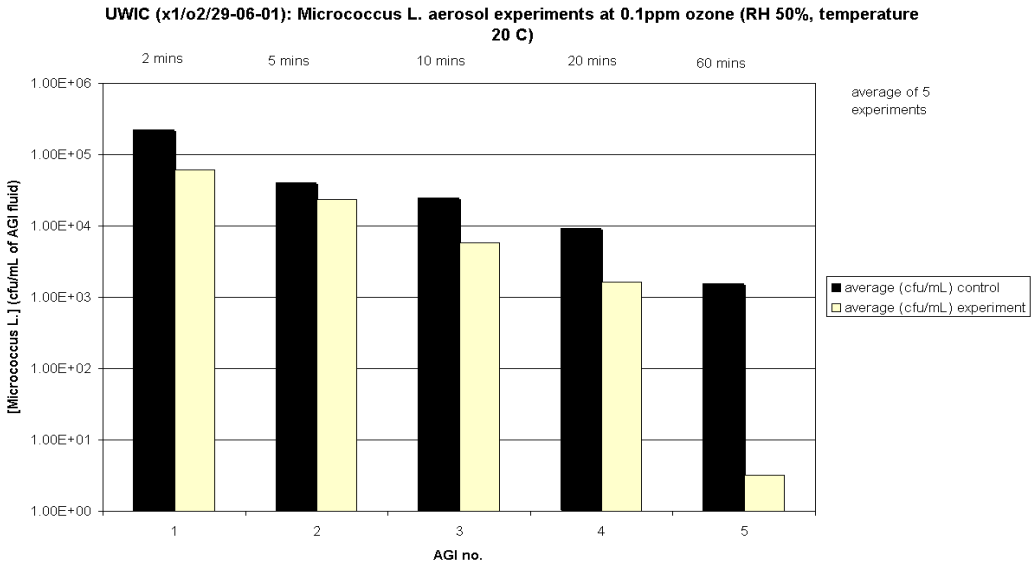
When ozone comes into contact with an organic substance such as mould, yeast, bacteria or bacteriophage then the ozone dissociates and the free highly reactive oxygen atom reacts with the cell membrane. Initially the ozone attacks the bacterial membrane glycoproteins, glycolipids and certain amino acids such as tryptophan. It also acts upon the sulfhydral groups and some enzymes resulting in the disruption of normal cellular activity. Bacterial death is rapid and is often attributed to changes in the cell permeability followed by cell lysis. If the ozone comes into contact with a VOC or an odour the free oxygen atom reacts with the VOC oxidising it into a harmless or non odorous substance. As each of these reactions occur the amount of ozone present continuously reduces until all the ozone has decayed back to the original normal bi-atomic oxygen making it a safe and environmentally friendly disinfectant with no harmful residues.

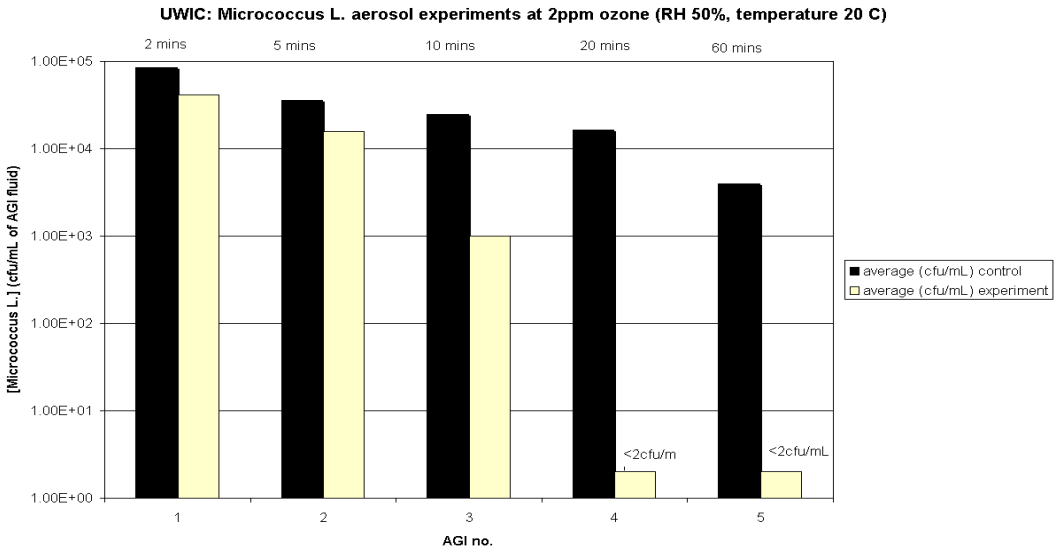
### Effectiveness against Airborne Micro-organisms

Extensive research has been conducted with the Food Research and Consultancy Unit at the University of Wales Institute, Cardiff (UWIC) into the affect of ozone on airborne micro-organisms. These trials showed that with an airborne bacteria and using ozone concentrations of between 0.05 ppm and 2 ppm reductions in viable bacteria of between 99% and 99.99% could be achieved within 60 minutes.

### Ozone disinfection aerosol results - 20°c RH 50% *(Micrococcus Luteus)*





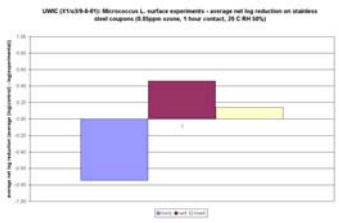




## Effectiveness against Micro-organisms on Surfaces

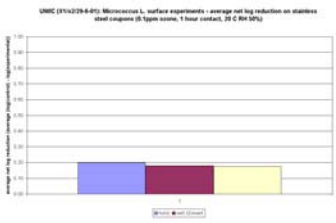
During the trials at UWIC it was also demonstrated, using stainless steel coupons, that the effective disinfection of surfaces, including the normally difficult to clean exposed undersides, could be achieved within an hour using ozone levels of 2 ppm. It was also shown that during these experiments that very effective disinfection could still be achieved even when the stainless steel coupon had not been subjected to any cleaning and could be considered to be dirty.

## Ozone surface disinfection results - 20°C RH 50%, 1 hour exposure *(Micrococcus Luteus)*

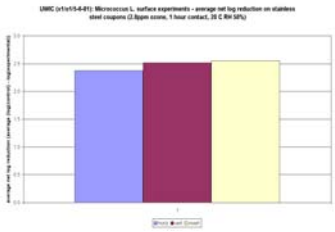
0.05 ppm ozone



0.1 ppm ozone

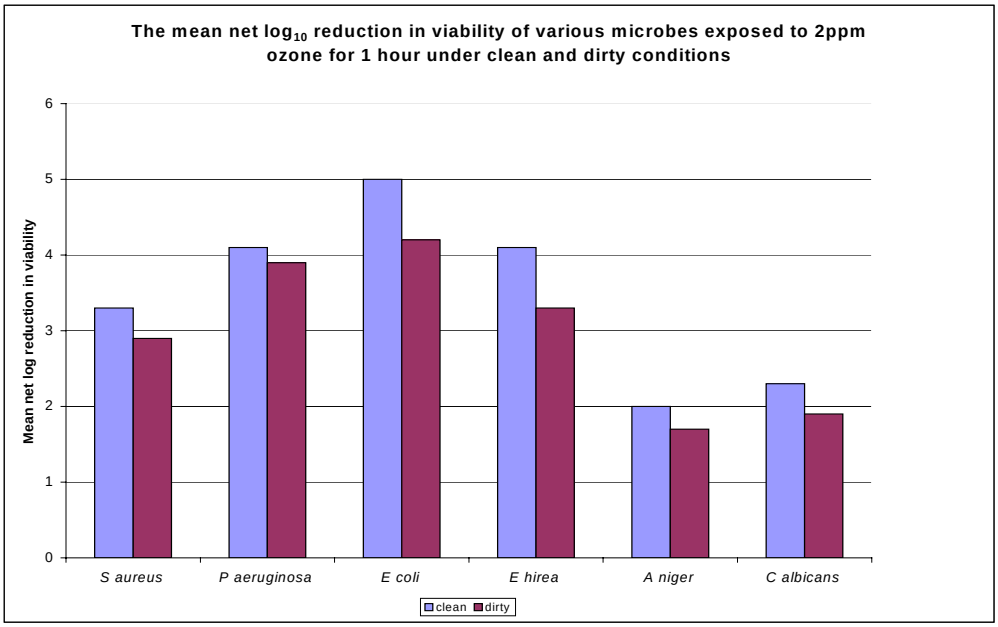


2 ppm ozone



Other work on surfaces has shown that ozone is similarly effective across a wide range of micro-organisms including *Escherichia Coli, Serratia Liquefacens, Enterococcus Hirae, Candida Albicans, Pseudomonas Aeruginosa, Staphylacoccus Aureaus, Listeria Innocua, Aspergillus Niger, Saccharomyces Cerevisae* and *Rhodotorula Rubra.* Percentage reductions ranging from 97% up to 99.999% have been demonstrated

using 2ppm of ozone for between 1 and 4 hours with gram negative bacteria such as *E. Coli* being more sensitive to ozone than gram positive ones such as *S. Aureas*. Yeasts *(Saccharomyces Cerevisae, Rhodotorula Rubra)* and mould *(Aspergillus Niger)* were found to be the most resistant to ozone probably due to the thick cell walls of these organisms presenting a more effective barrier to the initial penetration of the gas.




## Affect of Ozone on Insect Infestation

It has also been noticed in the field that regular ozone treatment in cheese stores eliminates mite damage although the actual causal mechanism of this effect is still unclear. There are several anecdotal field reports of mites seeking refuge away from the ozone by entering the interstice between the cheese and the shelf that the ozone cannot penetrate where previously they have occupied all surfaces. While these reports are so far uncorroborated scientifically the field results being demonstrated by users are impressive. These same users are increasingly confident that they will be able to completely abandon the use of chemical fumigants such as methyl bromide in their cheese stores by the regular use of ozone.

There are three species of mite that attack cheese, the Flour Mite *(Acarus Siro)*, the Cheese Mite *(Tyrophagus Casei)* and the Mould Mite *(Tyrophagus Putrescentiae)*.

The Flour Mite is the commonest mite to infest a wide range of foodstuffs, including cheese, and it can be harmful if consumed in any quantity by humans or animals. It has the unusual characteristic of a *hypopus* stage in its development during which it becomes almost immobile, grows suckers with which it attaches itself to surfaces and its body wall hardens. During this phase of its life cycle it becomes very much more resistant to insecticides and other adverse conditions such as surface cleaning. It is likely that this characteristic is what makes this particular mite so hard to eradicate. It thrives at 28 $^{0}$C and 90% RH but becomes inactive below 17 $^{0}$C and 65% RH.

Cheese mites are larger than flour mites, very common and just as cosmopolitan in their diet although are particularly known for the holes that they gnaw in cheese leaving behind a grey dust made of their dead bodies, cast skins and faeces that is a recognised cause of dermatitis amongst cheese industry workers. They like the cool temperatures and high humidity generally associated with cheese storage and can even survive refrigeration but they do however lack the *hypopus* stage of the flour mite in their development that makes them easier to manage.

Finally there is the Mould mite that unlike either of the others two does not appear to be injurious to health but can be responsible for causing tremendous losses within foodstuffs. It is very common although as its name implies it is only found in numbers where mould and fungi will flourish such as cheese stores.

The regular use of ozone to eliminate mould would clearly have a beneficial impact on the presence of the mould mite by removing what is presumably its food source. Exactly how ozone deters the other two species is less clear but the presence or lack of mould may play an important part and more research is needed.

## Equipment

Ozone Industries Ltd was formed in 1994 by venture capitalists to research into and develop devices to generate ozone and it found its first market niche in the field of odour control in washrooms and the like. Shortly after a farmhouse cheese producer contacted the company to see if ozone could provide a means of eliminating unwanted moulds in a cheese store. As a result of this contact a dialogue commenced with the cheese producer and a long term trial was set up using an existing design of air phase ozone generator that had been developed for odour control applications. Two units were installed in the roof space of the cheese store controlled by time clocks that turned them on each night for eight hours after people had vacated the building. An immediate improvement was noted but this improvement slowly reduced over a period of a couple of months and it was found that the ozone output of the generators had declined to almost zero. After servicing the improvement returned but again slowly declined as the output of the generators fell away.

It was clear that the impact of ozone was beneficial if the generator output could be kept stable and a product development programme ensued. It became obvious that the generators trialed could not withstand the damp conditions present in the cheese store and that the moisture seriously affected their electrical properties. After many trials of different configurations spread over the next two years it was concluded that the only solution was to eliminate the moist atmosphere entirely by using concentrated oxygen fed directly into a reaction chamber.

Ozone Industries had been experimenting with this idea in the laboratory for some time and the indications were that an economically viable product could be built that would have the right output levels, controllability and reliability that would be necessary for this demanding application. A prototype unit was built and installed and within days the improvement was obvious.

## Results

The cheese store immediately lost its usual musty smell, the mould rapidly vanished and by the end of May the cheese mites had disappeared completely. Cheeses that without ozone would be covered with mould within six weeks were completely free and required no cleaning. Indeed the improvement was so great that it was found that cheeses could be despatched to customers without the need for any final cleaning processes for them to meet standards set by customers. Not only were there substantial labour savings from the reduction in handling during the whole production process but it also resulted in less blemishes and damage to the rind which coupled with the elimination of the mite damage resulted in higher yields being achieved.

The annualised cost savings from using ozone per tonne of cheese produced were shown to be in the range of £350-£400 per tonne enabling a payback of two years or less to be achieved even by a small producer.

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## Legislation and Health & Safety

The more traditional methods of disinfection in the cheese industry have been to use chemical fogging but this is not used greatly by small producers and is rapidly disappearing amongst the larger ones as production demands and the Biocidal Products Regulations (2001) that are progressively restricting the use of chemicals. For example, methyl bromide, has been the workhorse for the industry for many years is due to be withdrawn by 2005 because of the damage it causes to the ozone layer. The use of these chemicals is hazardous and expensive with the serious problem of the elimination of the harmful residues that they tend to leave behind. As a result of these problems industry utilises these chemical treatments as infrequently as possible leading to huge peaks and troughs in the amount of viable microbiological contamination and insect infestation.

Ozone on the other hand while it must be used carefully especially in high concentrations leaves no residues at all and can be automatically applied without exposing people so it is normally used on a daily basis. This ensures that the microbiological and insect life is kept tightly under control at all times preventing unexpected blooms that cause product to be spoiled, reject batches or customer complaints.

Ozone is classified as a toxic gas with a 15 minute TWA maximum exposure of 0.2 ppm and an 8 hour TWA maximum exposure of 0.1 ppm so when used at concentrations above these levels precautions need to be taken. The safe way of using ozone as a disinfectant is to use a time clock to switch on the generator after everyone has vacated the room to be treated and turn it off again about two hours before anyone re-enters. Provided that there is at least three hours between the on and off times then the ozone will have had sufficient time to reach its disinfectant level and stay there long enough for it to do its work. The time period between switch off and the first re-entry to the room allows the ozone to decay naturally to a safe level.

Control of the ozone output is now so advanced that in a number of food production plants very low levels of ozone, that are well below the health and safety limits, are being used on a continuous basis as a sanitiser while people are present. This technique has been shown to further keep the bacterial growth under control by reducing the number of viable bacteria by around 80%. This enables plants that operate 24/7 can still benefit from using ozone thereby improving product quality and reducing spoilage. Long term field trials in a large cheese processing plant producing bags of grated cheese for the catering industry resulted in the number of customer complaints caused by mould contamination falling from 90/day to less than 1/day.