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Air disinfection and food preservation by ozone gas

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Abstract

Most food spoilage is the result of bio-chemical reactions between microorganisms such as bacteria, funguses, molds, yests and food as carbon based organic matters. Food preservation by ozone gas slows or prevents this biochemical reaction. Ozone consumed for these reactions causes its reduction or decomposition. The amylaceous food (bread and other) and some kinds of fruits were chosen for research. The shape of the ozone reduction curves, its rate and half life time (HLT) reflects the rate of reaction between ozone and microorganism on food. The bacteriobiological and photographic analysis of the samples before and after ozone treatment show that ozone gas can kill or prevent the growth of funguses and mould so this gas can be effectively used to preserve the food.

Keywords. Food preservation, ozone decomposition, ozone treatment, mould, funguses, Aspergillus.

1. INTRODUCTION

Food preservation is a concern not only for the housewives but also for those working in the food industry. Most food spoilage is the result of bio-chemical reactions between microorganisms such as bacteria, funguses, molds, yests and carbon based organic matters which are the main components of foods. Therefore, methods used for food preservation must either to kill the microorganism or to make them inactive. Chemical methods are based on oxidation or denaturation mechanisms. It is concerning to the use of ozone gas, alcohol vapor ect. Physical methods include series of techniques such as irradiation, UV, plasma or heat and so on. One of the most polular methods is low temperature preservation. Cold-processing slows down the bio-chemical reaction rate in the food, but does not kill most microbes or denature enzymes. For every 10 °C increase in temperature, bio-chemical reaction rates are roughly doubled. When chilled foods are brought back to room temperature, the rate of the spoilage process will increase. Dioxide carbon, nitrogen gases are also used mainly for grains because insect does not live in the oxygenless conditions. Recently, one began to use ozone for air disinfection because it is a powerful oxidant. Ozone is an unstable compound and decomposes to molecular oxygen spontaneously without leaving a toxic residue. Lack of toxic residues makes ozone a favorable sanitizer. Ozone has been used since the late nineteenth century to purify water. Ozone as a disinfectant is declared to be generally recognized-as-safe (GRAS) for food application in 1997. Since that time, interest in developing ozone applications in the food industry has increased.[1,2] Ozone can be applied to food products as a gas or as an ozone-water solution. It has been used for process water sterilization and recycling, inactivation of bacterial growth, prevention of fungal decay, washing and storage of fruits and vegetables, reducing microbial populations on stainless steel surfaces, control of storage pests, destruction of pesticides and chemical residues, and control of microorganisms on poultry and meat products. In the gaseous phase application, a series of advantages distinguish ozone from others like to UV radiation or HEPA filters. Ozone is a gas that could penetrate to every corners of the room, thus it could disinfect the entire room effectively. It needs to emphazise that ozone can kill both bacterium and its spore and it differs ozone from vaporized alcohol. A series of studies of these authors on water disinfection by ozone have been shown in Refs. [3-5]. This work is focusing on air disinfection by using ozone gas.

2. MATERIALS AND METHODS

The amylaceous food (bread and others) and some kinds of fruits were chosen for test (hereinafter we consider as the samples). They were put into a closed box of ~ 300 dm^3 in volume. The box connects with an ozone generator BKIDT using oxygen rich inlet gas. The power of the ozone generator is approximately 3 grams ozone per hour. An ozone monitor of a range 0-100 ppm was connected with the box. Every day, ozone gas was pumped in to the box three times, every time lasted 5 min. Average ozone concentration was approximately 60 ppm during 5-10 minutes. The similar samples were laid in air for comparison. Experiment was carried out in June - July at average temperature near 34-36 ^oC, the humidity was kept at 70-80 %. In a few days, the funguses, moulds were appeared in surface of samples that were laid in air. The pictures of mouldy bread and fruits were taken by an ordinary camera. The type of the funguses was analyzed at The Biological Lab of National Institute of Nutrition. Morphology of the funguses was observed by an optical microscope.

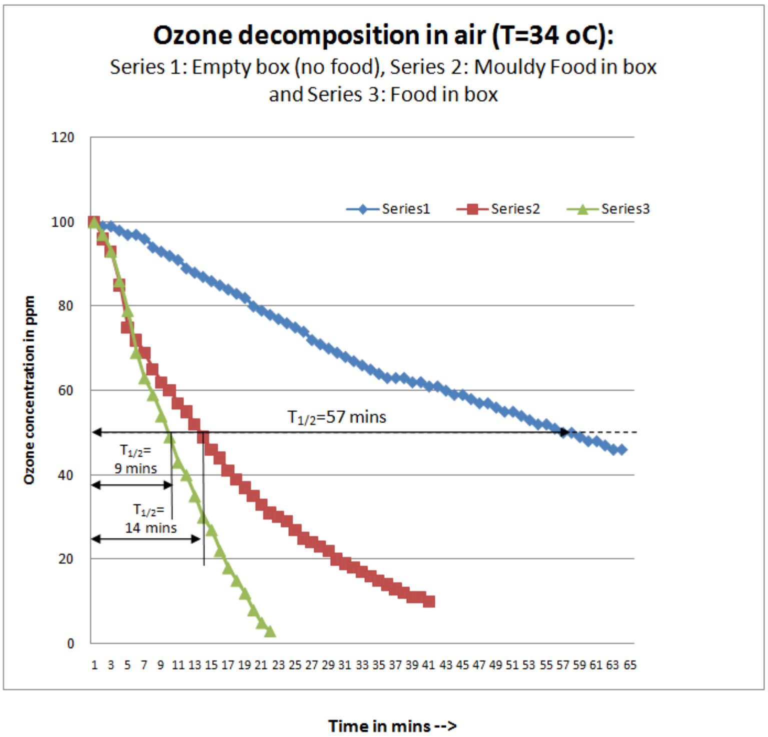


Figure 1: Ozone decomposition curves in air

3. RESULTS AND DISCUSSION

3.1. The ozone decomposition curves

Ozone molecular is unstable, so it spontaneously decomposites to oxygen by $2O_3 \rightarrow 3O_2$. The decomposition rate increases with the temperature increase. It is called as thermal decomposition. Half life time (HLT) of thermal decomposition in air at 30 ^oC is near 2 days. Besides, HLT also depends on the experimental condition.

First, we pump ozone into the box and stop pumping when the ozone concentration in box reached to 100 ppm. From this moment we began to observe the reduction of ozone concentration for getting the time dependant concentration. We call it as decomposition curve. We measure the decomposition curve for (i) empty box (the box without food), (ii) the box with fresh food and (iii) the box with mouldy food. The slopes of the curves are very different. The slope of the curve will determine the ozone reduction rate (ppm per min) and the HLT (min). In the case of empty box, ozone reduction is low, the HLT is long (57 min). In contrast, for the box containing fresh food, the reduction rate is high and the HLT is short (9 min., i.e. near 6 times shorter in comparison with the empty box). As mentioned above, ozone in the empty box decomposites only by thermal reason. In contrast, for box containing food, besides thermal decomposition, some amount of ozone needs to use to kill the mould spores and other microbes often existing in food surfaces. It consumes additional ozone and therefore causes faster ozone reduction, consequenly, the decomposition rate increases and HLT reduces. It is interesting to see that the curve for the box with mouldy food is lying between the two others. The rate for this case is higher for empty box but lower than box containing fresh food. This evident fact will be explained as follows: ozone can easily kill the spores of moulds but hardly destroy the grown up moulds. Consequently, ozone must be consumed more to kill spores than to destroy the grown-up mould. Further, additional experiments needed to assert it.

Thus, generally, the curves in Fig. 1 show that, the slope of the ozone reduction curves reflects the degree or the rate of bio-chemical reaction of ozone gas with the microrganism.

Table 1: Total spores (funguses, moulds, yests)

Samples	Unit	Quatity	Type	Method
F1 (Mandarin), 8 days	CFU/g	1.2×10^{8}	Found at least: - *Aspergillus flavor* - *Aspergillus fumigatus*	TCVN 8275-2, 2010. 52 & 53 TCN TQTP 0001-2003
F2 (Sweet potato), 8 days		3.4×10^{8}		
F3 (Bread), 6 days		5.6×10^{7}		

Tests have been done at the VILAS 307, National Insitute of Nutrition. Bureau of Accreditation issuses, July 2018. CFU: Colony Forming Units.

3.2. Bacteriobiological analysis

Visually, one can observe both groups of samples that were exposed in air and in the box supplied of ozone. While all samples in ozone box look normal i.e. not any trace of mould is seen, in contrast, the samples kept in air were covered by a color layer like mould. The color layers are different in shape and in color for bread and fruit samples.

Mouldy samples were tested in a microbiological laboratory to classify the grown up bodies in foods. The results of biological analysis were tabulated and shown in table 1.

The moulds developed on all samples exposed in air for a few days. On most samples, at least, two funguses were found and classified, they are *Aspergillus flavus* and *Aspergillus fumigatus* which are the genus Aspergillus. These funguses are a heterotrophic microorganism that can not produce its own organic matters and need to takes organic carbon from other living bodies (microscopic parasite). These funguses may cause a wide variety of diseases e.g aspergillosis, especially in individuals with an immunodeficiency.

3.3. Microphotographic observation

We show a series of photographs of food's surfaces (Figs. 2 and 3). All samples (starch and fruits) without ozone treatment were covered by moulds. In contrast,

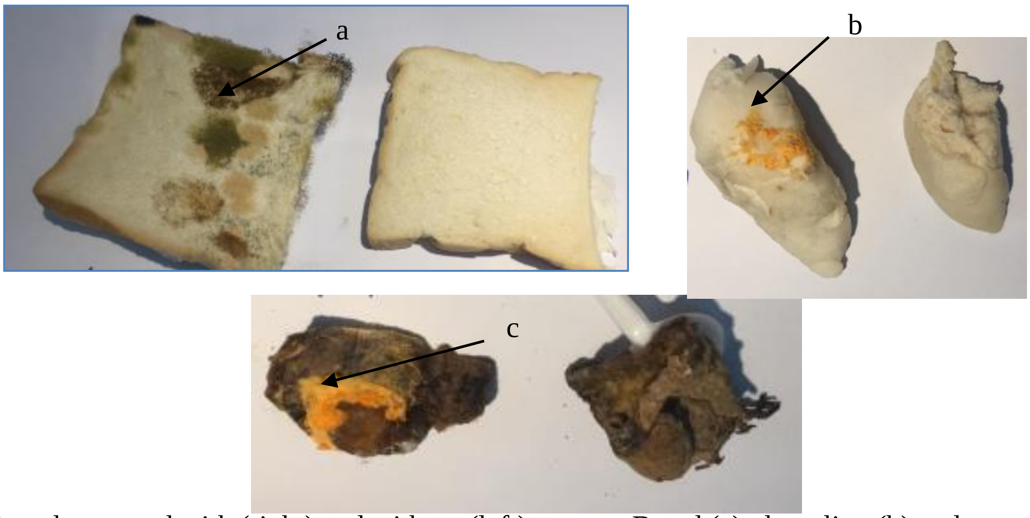
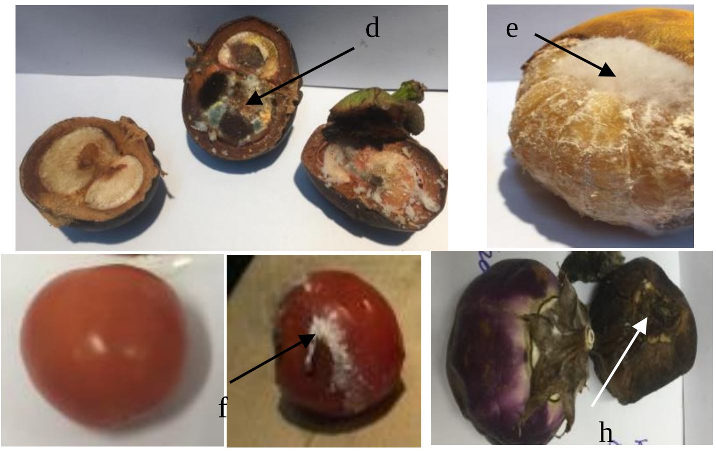


Figure 2: Samples treated with (right) and without (left) ozones: Bread (a), dumpling (b) and sweet potato (c) samples kept in air for 6-9 days. All samples kept in air without ozone became moldy.

no traces of mould was seen in all samples kept in ozone gas. High humidity allows for extensive mold growth. So we try to research the relation between ozone gas, humidity and mould growth. The moulds on the samples kept in ozone gas 6-9 days at the humidity of ~ 70 % and > 90 % were not observed.

At the same time all samples kept in air without ozone at humidity of > 90 % were mildewed (Fig. 3). Image in Fig. 4 showed the morphology of a mould found in one of the samples without ozone treatment. Its morphology is similar to an *Aspergillus flavus* fungus. For comparison, an image of an *Aspergillus* flavus is shown in Fig. 4b.



Fruit samples: mangosteen (d), orange (e), tomato (f) and eggplant (h) kept in air for 9-15 days without ozone (right) and with ozone (left). All samples kept in air without ozone became moldy

Figure 3: All samples were kept in ozone box look normal. On them not any trace of moult is seen. In contrast, all samples kept in air became spoiled, on their surface the bodies of filamentous shade are observed visually. At least two mould types are found: they are *Aspergillus flavus* and *Aspergillus fumigatus* according to biological analysis. Their density is in the range of 10^8 CFU/g. Ozone treatment duration is shown in the text.

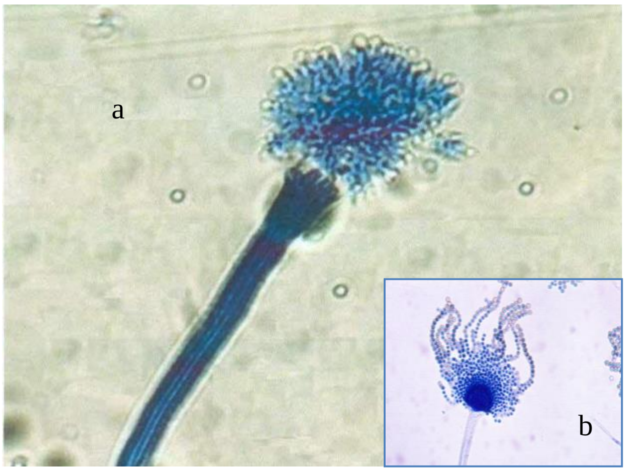


Figure 4: Optical microphotography of a fungus found in sample without ozone treatment (a). Its morphology is similar to the *Aspergillus flavus* shown in image (b), Source: MBBS Medicine (Humanity First)

4. CONCLUSSIONS

1. The ozone reduction curve reflects the rate of biochemical reaction of ozone with the microorganism in gaseous phase. The parameters deduced from the curves can be used to establish the technical parametres for food reservation by ozone.

2. Food (bread and fruits) kept in air for a few days are spoiled by moulds and fuguses. In contrast, not any trace of moulds was seen on all samples kept in ozone even at the high humidity (> 90 %). So, obviously, ozone gas can be used for food preservation.

3. The *Aspergillus flavus* and *Aspergillus fumigatus* funguses found in food kept in air for a few days may cause a wide variety of diseases e.g aspergillosis, especially in individuals with an immunodeficiency.

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