

Vitreous System $\text{Ag}_2\text{O} - \text{ZnO} - \text{B}_2\text{O}_3$ Action Against Gram Negative Bacteria

Razvan Stefan, Marina Spinu, Silvana Popescu, Georgeta Taralunga

University of Agricultural Science and Veterinary Medicine-3400-Cluj-Napoca, Calea Manastur, 3-5, Romaniaia

Abstract

In this work the ability of the system $x\text{Ag}_2\text{O} \cdot (100-x) [45\text{ZnO}55\text{B}_2\text{O}_3]$ oxide glasses to inhibit the growth of bacteria *Escherichia coli*, *Pseudomonas* and *Salmonella* was investigated. Using the diffusimetric method there were measured the diameters of inhibition, in order to classify the sensitivity of gram negative bacteria to oxide compounds containing silver, for their use as filters.

Vitreous samples were processed as powders with grain between 45 and 75 μm and less than 45 μm for a large of contact area with the microorganisms and to make possible the study of biological effect of grain addition. Action of the investigated oxide system against the gram negative bacteria is strictly related to the presence of silver oxide in glass composition.

Keywords: antibacterial, chemical durability, glass

1. Introduction

Lately based B_2O_3 oxide glasses were given increasing importance due to their ability to form easily vitreous structures. Based on boron oxide glass is composed of structural units of boroxol (B_3O_6), and with the introduction of alkali or alkaline earth metal oxides network breaks into smaller structural units di-, tri-, pentaborate.

Borate oxide structure can absorb [1, 2] to encompass Ag_2O . As already known boron and phosphorus vitreous structures can be used in the medical field [3], even if it presents a high bioactivity and can not connect directly to living tissue by hydroxyapatite formation layers.

Silver oxide glass network is embedded in the glass to become useful in electronics [4], but also in medical applications can be used as filters for air purification in shelters or in operating rooms. Because the silver antibacterial capacity irrespective of where it is can be used to wrap materielele with caare medical instruments, light industry for the impregnation of textiles wish to use to get clothes with silver nano particles.

The introduction of this material in body may cause danger of a local infection. No matter on the mechanism by which silver ions come in contact with the bacteria released by ions in fluids or by another mechanism, their presence is necessary when there is possibility of occurrence of an infection. Mechanisms of action of silver against bacteria are not clarified, but there are a few assumptions namely: silver ions penetrate the cell DNA macro cell molecule [5], that cause to lose its ability to replicate or if the silver metallic form it is assumed that they inactivate proteins [6, 7]. Also the shape and size [7] silver particles can produce different effect on microorganisms in general are lower but the effects are more important [8].

Many results have shown that size in experimental powdered oxide action on Gram negative bacteria is dependent on particle size that interact with bacteria. Also the size and shape can be modified and their biocompatibility [9].

Gram negative bacteria such as *Escherichia coli* is a class of bacteria present in the air in shelters being transported by dust particles. Its adaptability to antibiotics is very high and the practical need to

¹ Corresponding author: Razvan Stefan

obtain this new antibacterial compounds that bacteria are not adapted or that one can not adapt. In the present work we intended to study the effect $x\text{Ag}_2\text{O} (100-x) [45\text{ZnO}55\text{B}_2\text{O}_3]$ system against gram negative bacteria using difussimetrical method. We folow also to obtain some grain as fine powder to maximize surface contact between the powder and cultural environment of microorganisms investigated.

2. Materials and methods

Oxide glass belonging $x\text{Ag}_2\text{O} \cdot (100-x) [45\text{ZnO}55\text{B}_2\text{O}_3]$ vitreous system with $0 \leq x \leq 7$ have been obtained from Ag_2O , ZnO , H_3BO_3 of reagent grade putrity. Mixtures were melted in air at 1230°C , sintered corundum crucibles, and maintained 15 min. at this temperature.

Molten pieces were broken in an agate mortar and turned into fine powder. Powders thus obtained were passed through the site with pore diameters of $45\ \mu\text{m}$ and $75\ \mu\text{m}$ to obtain material with controlled grain.

Antibacterial efficacy of the compounds investigated was tested on Gram negative germs (*E. coli* standard strains ATCC 10536, *Salmonella enteritidis* ATCC 13076, *Pseudomonas aeruginosa* ATCC 27853), the diffusion method in agar gel 2%. Agar poured into Petri dishes with a diameter of 12 cm, was sown with bacterial inoculum, prepared from 24-hour culture on agar, diluted in saline tube density of 1 McFarland scale. Immediately after sowing were charged 3.5 mm diameter wells, the compounds were assigned a concentration of 15% in quantity of 40 ml / well. Each compound was tested in duplicate. The diameters of inhibition zones were read after incubation for 24 hours at 37°C , being expressed in mm.

3. Results and discussion

Action of vitreous compounds with the composition $x\text{Ag}_2\text{O} \cdot (100-x) [45\text{ZnO}55\text{B}_2\text{O}_3]$ with $0 \leq x \leq 7$ mol% of the inoculum of *Escherichia coli* (Fig. 1) is clearly evidenced by the inhibition diameters between 10 and 12 mm.

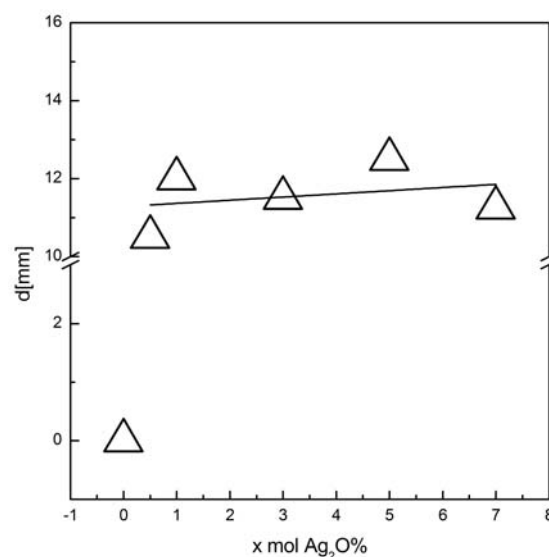


Fig. 1 The dependence of inhibition diameter in case of *E. coli* bacteria on Ag_2O concentration in vitreous compounds $x\text{Ag}_2\text{O} \cdot (100-x) [45\text{ZnO}55\text{B}_2\text{O}_3]$

The compounds investigated was compared with base glass $44\text{ZnO}55\text{B}_2\text{O}_3$ (note and $44\text{Zn}55\text{B}$) to investigate its inhibitory effect and its value is given by the value $x = 0$ for each graph. Investigation of the inhibitory mechanical capacity on the inoculum is excluded as can be seen (Fig. 1) because the sample without Ag_2O not present any form of inhibition in conditions where grit is identical to that used for other concentrations, inhibition of mechanical bacterial development is one of possible mechanisms. Figure analyzed data found in the area of concentration. Also there is the possibility of *E. coli* adhesion to the surface electrostatic powder leading to its impossible development[10]. Thanks so different between the molar content of silver ions (molar ratio of 1:14) between the ends of the compositional range investigated, report that is preserved and the surface, there are very different between the expectancy effect between the edges of the compositional range.

As can be seen (Fig. 1) the effect of compounds $x\text{Ag}_2\text{O} (100-x) [45\text{ZnO}55\text{B}_2\text{O}_3]$ is not directly proportional to the molar content of silver in the case of *E. coli* bacteria, but increase with increasing silver oxide as was obtained and in the usual borosilicate glasses [11] or a glass deposit [12] for medical active substances. Trough $45\text{ZnO}55\text{B}_2\text{O}_3$ dissolved in the matrix, $x = 0.5$ mol% is sufficient to inhibit bacterial growth on a diameter of 10.5 mm, a new increase of 0.5 mol% induces an addition of only 1.5 mm diameter

inhibition. Experimental points of glasses containing silver were linear fitted and showed a minor increase of 8% in terms of scientific content principle inhibitor (silver ions) of 14 times.

In Figure 2 the evolution of diameters inhibition are presented made by vitreous zincborat compounds containing up to 7 mol% Ag_2O if *Pseudomonas bacteria*. For comparison (Fig. 2) is presented as a matrix of the experimental and $\text{ZnO-B}_2\text{O}_3$ and as you can see it does not cause any inhibtie on *Pseudomonas* growth.

The data were experimented with a second degree polynomial function to trace the evolution trend of inhibition rays generated by a molar content Ag_2O increasingly higher oxide matrix investigated. The diameter mean inhibition are found in the range $d \in [11.5, 15]$ mm and have an upward trend throughout the compositional range in which content Ag_2O vitreous matrix increase. Because the disc diameter of inhibition is higher for any $x \in [0.5, 7]$ mol% Ag_2O if only if *Pseudomonas bacterium* and *E. coli* can be said that the first of which is more sensitive to direct one of the silver ions inactivation mechanisms, the respiratory chain termination [5].

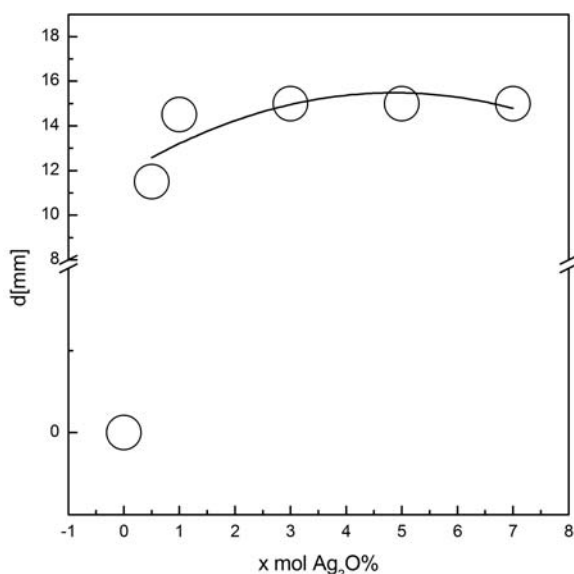


Fig. 2 The dependence of inhibition diameter in case of *Pseudomonas bacteria* on Ag_2O concentration in vitreous compounds $x\text{Ag}_2\text{O} \cdot (100-x) [45\text{ZnO}55\text{B}_2\text{O}_3]$

Also differences between the average diameter of inhibition respectively for $x = 0.5$ and $x = 1$ is ~ 3 mm denoting the existence of lower bacterial sensitivity threshold as a 7-fold increase of silver concentration is an increase of radius of only 5

mm. Increasing molar content in the samples leads to growth Ag_2O antibacterial properties of glass zincborate if *Pseudomonas* as in other vitreous [1, 11], because of a greater number of available silver ions for diffusion on the material surface. Diffusion happening in this case a longer late, due to a compound concentration in the area.

Areas of inhibition due to oxide compounds based on silver ion doped B_2O_3 in the case of *Salmonella* (Fig. 3) have average diameters d within the range $[11.5, 14.5]$ mm very close to those measured in the case of *Pseudomonas bacteria*, the trend is somewhat different. There is no positive dependence of inhibition area in relation to concentration mol% Ag_2O .

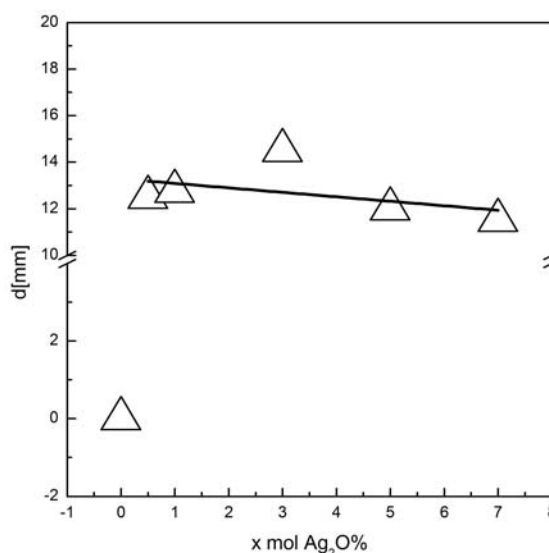


Fig. 3 Diameter of inhibition on *Salmonella* bacterium cultures vs. Ag_2O concentration in $45\text{ZnO}55\text{B}_2\text{O}_3$ glass matrix

Glass of the $\text{ZnO-B}_2\text{O}_3$ a not inhibit bacterial growth at all if the gram negative species, and vitreous material containing 0.5 mol% inhibiting agent performs an inhibition diameter of 12.5 mm. Indicating an inhibition threshold is reached at much lower concentrations of silver than 0.5 mol%, the compounds containing metal ions there is a constant investigation of the relationship between antibacterial and toxic to the body. Growth of *Salmonella* environments investigated is comparable measures if *Pseudomonas bacteria*.

To highlight the effect of powdered grain interacting with bacterial culture sample $x = 3$ mol% was Ag_2O mortar and diversity in the sieve $d = 45 \mu\text{m}$ and separately from that of $d \in [45, 75] \mu\text{m}$ and inserted in *Salmonella* cultures as well as

in previous samples. At these dimensions no different antibacterial actions can be highlighted.

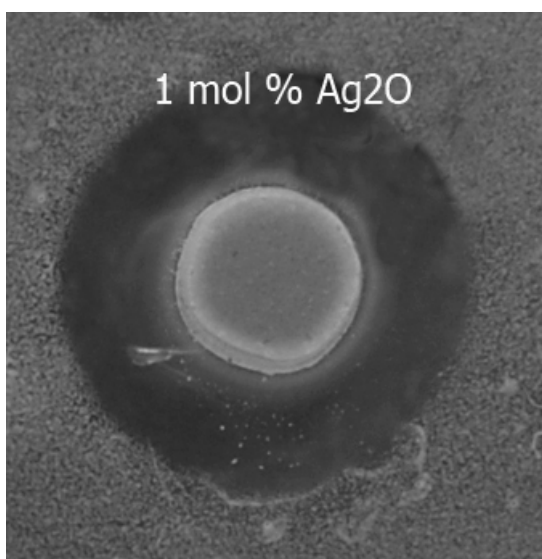


Fig. 4 Action of $1\text{Ag}_2\text{O}\cdot 99\% [45\text{ZnO}55\text{B}_2\text{O}_3]$ vitreous compound with $d \leq 45 \mu\text{m}$ on stem *Salmonella* could investigate the differences of antibacterial action.

4. Conclusions

Oxide glass were prepared in glassy system $x\text{Ag}_2\text{O} (100-x) [45\text{ZnO}55\text{B}_2\text{O}_3]$ with $0 \leq x \leq 7$ mol%. The action of compounds on gram negative bacteria *Escherichia coli*, *Salmonella*, *Pseudomonas* is strictly related to the presence of silver oxide homogeneously embedded in oxide matrix.

Effect of inhibition is proportional with a silver concentration on the surface, the inhibition was required much lower amounts.

The lowest effect of investigated glasses appear on *E. coli*.

The inhibition area created by the sample $1\text{Ag}_2\text{O}99\% [45\text{ZnO}55\text{B}_2\text{O}_3]$ smashed fine in *Salmonella* agar in culture is illustrated in Figure 4. The ratio of area of inhibition and area of the well $A_i / A_w = 5.29$, but would not specify exactly when grain is due to diffusion or diffusion of silver ions released in body fluids.

Antibacterial effect is the sum of Ag^+ ion diffusion effect in the material released [13] in the diffusion medium and powdered in liquid diffusion effect, silver ions in inhibiting the early stages of colonization bacteria.

The investigated $45\text{ZnO}\cdot 55\text{B}_2\text{O}_3$ based glass has no bactericidal or bacteriostatic inhibition areas as any studied species.

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