



# Influence of Silver Nanoparticles on Selected Properties of Dental Porcelain: An *in vitro* study

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## Abstract

**AIM:** This study was conducted to reveal antibacterial effect of silver nanoparticles (AgNPs) incorporation on dental porcelain and its effect on flexural strength and surface hardness of porcelain.

**MATERIALS AND METHODS:** Two groups were prepared and classified according to the type of the liquid used for mixing with porcelain powder into Group 1 (Control group mixed with distilled water) and Group II (modified group mixed with AgNPs solution). A total of 60 samples were constructed (30 sample in each group). For each group, ten samples were evaluated for anti-bacterial activity, flexural strength, and surface hardness.

**ANTIBACTERIAL TEST:** The prepared porcelain disks of control group and modified group were placed in the prepared inoculum of *Streptococcus mutans*; then, the colonies forming units were counted. The three-point bending test was used to evaluate the specimens' flexural strength, and the surface hardness was assessed by micro-Vickers hardness test. Results were statistically analyzed and tabulated in this study.

**RESULTS:** Group II showed antimicrobial effect against streptococcus with a statistical significant  $p < 0.001$ , there is a statistical significant higher level of flexure stress with  $p = 0.02$  in modified group and no statistical significant difference between modified and control group as regards surface microhardness.

**CONCLUSION:** AgNPs inclusion into porcelain demonstrates a great effectiveness in inhibition of bacterial growth by mitigating the colonizing activity. Further, it supports flexural strength but has no effect on surface hardness of porcelain.

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## Introduction

Optimal optical and biological properties of dental porcelains make them the first selection for both dentists and patients. However, a hard brittle porcelain will be cracked or damage when surface comes under a significant load or impact, enhancement of strength and fracture toughness of dental porcelain through different modification methods have been recommended and one of these methods is the addition of ductile metal particles [1].

Ceramics are considered to be the most biocompatible material used in dental field. Chemical durability of ceramic materials is a principle property for intraoral use since it must resist the degradation in the presence of a wide range of solutions with a variable PH at different temperatures. Dental porcelains are increasingly used due to their excellent esthetics and enhanced performance [2].

The oral cavity is populated by a variety of microorganisms. Ideally, the restorative materials should exhibit antibacterial properties to limit the adhesion and proliferation of pathogens at a very early stage and therefore, prevent secondary caries [3].

One of the main problems facing the fixed prosthodontics is the marginal leakage which means failure of seal at the tooth restoration interface, which might lead to dental caries, pulp sensitivity, necrosis, periodontal disease, esthetic problem, and decrease the restoration's longevity. Increase the longevity of restorations through the incorporation of bioactive agents is a great demand as it enhances microbial destruction and combat recurrent caries while maintaining the loadbearing capability [4].

The development of biomaterials with antimicrobial effects helps to reduce biofilm formation and prevent diseases. Antibiotic substances can promote bacterial resistance; thus, the nanoparticles are an alternative with greater chemical reactivity and a smaller size to penetrate biofilm [5].

Antimicrobial efficacy of silver nanoparticles (AgNPs) has been confirmed against bacteria, viruses, and other micro-organisms. The small size of nanoparticles may provide advantages to the biomedical field through enhanced biocompatibility. Furthermore, bacteria are less likely to acquire resistance against metal nanoparticles than other conventional and narrow spectrum antibiotics. Resistance of antimicrobial activity is thought to occur, because the micro-organisms develop many mutations [6].

Replacement of dental restorations consumes approximately 60% of the average dentist's practice time as nearly half of restorations fail within 10 years, secondary caries could result from Plaque accumulation adjacent to the restoration margins which might ultimately lead to failure of the restoration. To make long-lasting restorations, the materials should be made antibacterial. Hence, methods to kill or slow down the growth of unwanted bacteria are of great demand. The use of metallic nanoparticles which exhibit bactericidal properties is considered an interesting alternative to existing methods [7].

Several forms of silver have been showed to inhibit the growth of a broad spectrum of microorganisms, so silver incorporation has attracted much attention. We have only recently started to understand the mechanisms by which silver inhibits bacterial growth although the antimicrobial properties of silver have been known for centuries [8].

Nanoparticles also improved the antibacterial effect by altering the hydrogen bonding, respiratory process, DNA unwinding, cell wall synthesis, and division by making "pits" in the wall and increasing the permeability resulting in a bacterial death [9].

Gadella *et al.*, in 2013 [10], clarify the benefits related to the use of AgNPs in the prevention of dental caries in children. They compared the antibacterial properties of AgNPs, chlorhexidine, and silver diamine fluoride to *Streptococcus mutans* (SM). They found that the tested substances showed effective bacteriostatic and bactericide activity. They concluded that AgNPs proved to have antimicrobial activity against SM; therefore, they may be an effective agent in the prevention of dental caries.

Abu-Eittah *et al.*, in 2013 [11], explored the anti-bacterial effect of AgNPs modified acrylic – based provisional fixed prosthodontics material against aerobic Gram-ve (*Escherichia coli*), and anaerobic Gram +ve (*Enterococcus faecalis*) bacteria. Four concentrations of AgNps were used (5 µg/mL, 10 µg/mL, 15 µg/mL, and 20 µg/mL). The colonies forming units (CFU) were counted after 14 days. Inverse proportion between the concentrations of added AgNps and the CFU count in both studied microorganisms was resulted.

As dental porcelain provides excellent esthetic results, simulating natural teeth, it has been widely used as a fixed dental prosthesis material, but it has functional limitations as chipping or fracture due to its strength. The improvement of strength through the advancement of technology and newer available nanomaterials to broaden their applications is the scope [12].

Veneered ceramic and metal ceramic crowns are widely used in dentistry due to their excellent esthetics and mechanical durability. They are fabricated by fusing porcelain onto a metal or ceramic coping. However, it is sometimes necessary to replace restorations due to fracture or chipping of the veneering

porcelain. Unlike metals, porcelain is not malleable or ductile and readily cracks or breaks with virtually no deformation when an excessive force is applied. One method to toughen glass ceramics such as porcelain is the addition of ductile metal particles, which improve toughness through crack bridging and deflection by the metal particles [13].

Porcelain materials were reinforced with other materials such as leucite crystals and fibers in the earlier years to nanoparticles in recent days to enhance the mechanical properties [12]. The use of nanoparticles has become a significant area of research in dentistry, the main use has been focused in increasing the mechanical properties such as surface hardness, ductility, and toughness.

A study showed that Ag/ZnO nanocomposite may have a potential effect on glazed ceramic as it significantly increases the performance of glaze. Furthermore, the Ag/ZnO nanocomposite powder improved crack resistance and whiteness [14].

The effect of AgNPs on the behavior of subcritical crack growth in dental porcelains was studied. Subcritical crack growth occurs and leads to strength degradation over time. It was shown that addition of AgNPs significantly increased the stress corrosion susceptibility coefficient of dental porcelain [1].

Toughening of alumina ceramics with AgNPs was tried using different amounts of Ag nanoparticles (0.0, 0.5, 1.0, 2.0, and 3.0 wt. %). It was claimed that the reinforcement mechanism of these materials is due to the deflection of cracks due to metallic bridges formed by the silver used as alumina strengthener [15].

Tokushi *et al.*, in 2012 [1], assessed the fracture resistance of porcelain after the addition of nanoparticles of precious metals of silver and platinum. They found enhancement of the mechanical properties of porcelain following the addition of silver and platinum nanoparticles since it increased both the Young's modulus and the fracture toughness of commercial porcelain.

Hence, the aim of our study was to evaluate the effectiveness of addition of AgNPs to dental porcelain and to study its effect on bacterial inhibition, flexural strength, and surface microhardness of dental porcelain.

## Materials and Methods

### Grouping and specimen preparation

Two groups were prepared and classified according to the type the liquid used for mixing with porcelain powder into:

1. (Group I) mixed with distilled water (control).
2. (Group II) mixed with AgNPs solution (modified).

All the specimens were mixed and fired as recommended by the manufacturer. Specially constructed mold for each test was prepared according to the American Dental Association. Disk-shaped specimens were prepared in a split teflon mold of 5 mm diameter and 2 mm thickness and were used in testing bacterial adhesion, cylindrical-shaped specimens were prepared in a split teflon mold of 4 mm diameter and 10 mm thickness and used in testing hardness and bar-shaped ( $2 \times 2.5 \times 25$  mm) specimens were prepared in readymade mold of 2 mm thickness, 25 mm length, and 2.5 mm width and were used for flexural strength measurements.

#### *Preparation of control samples (Group I)*

The porcelain powder was mixed with distilled water to form a past then using a moistened brush that it was applied in the mold. The samples were dried by heating slowly in the open entrance of the furnace. This is carried out to get rid of excess water, before it has a chance to form steam once the compact has been dried. The porcelain samples were fired in a porcelain furnace.

#### *Preparation of modified porcelain sample (Group II)*

Porcelain powder was mixed with AgNP-dispersed solution (15 g of porcelain powder were mixed with 25 ml of AgNP-dispersed solution) then manipulated as previously explained in control samples.

#### *Bacterial adhesion test*

Determination of minimum inhibitory concentration (MIC)

AgNPs of concentration 200 ppm were purchased from Nano gate, Egypt. This solution was diluted to obtain the different concentrations (30, 40, 50, and 80 ppm) through the addition of distilled water and used to determine the MIC. A modified Kirby–Bauer disk diffusion method was used. A filter paper of 8 mm diameter was soaked until it is saturated then introduced in the agar dish; the agar used is Mueller–Hinton agar. When a filter paper impregnated with the tested concentration was placed on agar, solution which diffused from the filter into the agar inhibited bacterial growth. The lowest concentration which inhibited visible growth of the microbial strain was considered the MIC of Ag NPs.

#### *Anti-bacterial test*

SM (EMCC 1815<sup>T</sup> (ATCC25175) supplied in lyophilized form was used in the current study. Hydrating fluid was added to lyophilized microorganism and shaken well forming a suspension (Three discrete representative colonies were inoculated into 2 ml sterile

saline to obtain turbidity equal to  $10^7$  CFU/ml which is equivalent to 0.5 McFarland turbidity standard solution to form standard inoculum). Each prepared porcelain disk from control and modified group was placed in a tube containing the prepared inoculum then incubated at 37°C for 7 days. Then, a swab was heavily saturated by hydrated suspension and gently rolled over agar plates corresponding to the broth, in which they were maintained (brain heart infusion agar) incubated at 37°C for 24h. CFU which reflects bacterial cells viability and activity was counted.

#### *Flexure strength test (FS)*

All samples were individually and horizontally mounted in a custom made loading fixture (three point bend test assembly; Two parallel stainless steel rods with span length 20 mm supporting the specimen, with the damage site centrally located on the tensile side) on a computer controlled materials testing machine, as shown in Figure 1 (Model 3345; Instron Industrial Products, Norwood, MA, USA) with a loadcell of 5 kN and data were recorded using computer software (Instron® Bluehill Lite Software). Then, the samples were statically compression loaded until fracture at a crosshead speed of 1 mm/min. The Stress-strain curves were recorded with computer software (Instron® Bluehill Lite Software).

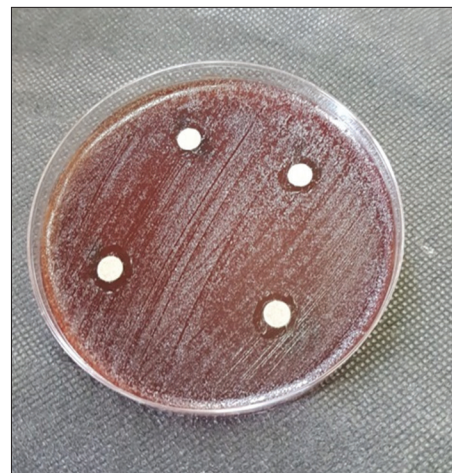


Figure 1: Hallos surrounding the filter paper

FS represents the limiting stress at which failure or instability is imminent. The value of the calculation of FS was guided by the formula

$$FS (\acute{o}) = 3 F (L)/2 wh^2$$

Where, F is the maximum load at the point of fracture, L is span, w is the width of the sample, and h is height.

#### *Micro-hardness test*

Digital Display Vickers Micro-hardness Tester (Model HVS-50, Laizhou Huayin Testing Instrument Co., Ltd. China) with a Vickers diamond indenter and

a 20X objective lens was used to determine Surface Micro-hardness of the specimens. A load of 100 g was applied to the surface of the specimens for 15 s. Three indentations, which were equally placed over a circle and not closer than 0.5 mm to the adjacent indentations, were made on the surface of each specimen. The diagonals lengths of the indentations were measured by built in scaled microscope and Vickers values were converted into micro-hardness values.

Micro-hardness calculation.

Micro-hardness was obtained using the following equation:  $HV=1.854 P/d^2$

Where, HV is Vickers hardness in  $Kgf/mm^2$ , P is the load in Kgf, and d is the length of the diagonals in mm.

## Results

### Determination of MIC

The lowest concentration which produced clear zone (inhibition zone) around filter paper was 40 ppm and this concentration considered the MIC. A distinct hallow surrounding the filter paper which means no microbial colonization, as shown in Figure 1.

### Anti-bacterial test

Comparison between control and modified groups revealed that control group showed the statistically significant higher mean count, as shown in Figure 2.

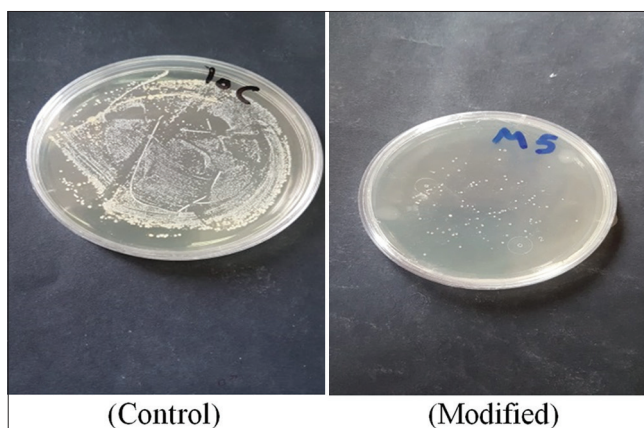


Figure 2: CFU for control (c) and modified (m)

Table 1 illustrates that there was a statistical significant low level of CFU with  $p < 0.001$  in modified group.

**Table 1: Comparisons of silver nanoparticles effect on the colony forming unit of dental porcelain**

Variables	Colony forming		p-value
	Mean	SD	
Modified	362	38.5	<0.001*
Control	1120	204.4	

### FS

Table 2 illustrates that there was a statistically significant higher level of flexure stress with  $p = 0.02$  in modified group.

**Table 2: Comparisons of silver nanoparticles effect on the flexure strength of dental porcelain**

Variables	Modified		Control		p-value
	Mean	SD	Mean	SD	
Flexure stress (MPa)	73.4	6.8	56.9	11.9	0.02

Table 2 illustrates mean flexural strength among different study groups with mean ( $73.4 \pm 6.8$ ) among modified group and ( $56 \pm 11.9$ ) among control group. There is statistically significant difference with  $p < 0.05$  between two study groups with higher value among modified group.

### Micro-hardness test

Table 3 illustrates that there was no statistical significant difference as  $p > 0.05$  – between modified and control group as regards micro hardness of specimen. That indicated both specimens show same level of hardness.

**Table 3: Comparisons of silver nanoparticles effect on the Micro-hardness of dental porcelain**

Variables	Micro-hardness		p-value
	Mean	SD	
Modified	409.26	13.46	0.9
Control	408.89	10.85	

Table 3 illustrates mean micro-hardness of dental porcelain among different study groups with mean ( $409.26 \pm 13.46$ ) among modified group and ( $408.89 \pm 10.85$ ) among control group. There is no statistical significance difference with  $p > 0.05$  between two study groups.

The mean colony forming unit in study groups is shown in Figure 3. Also, the mean flexure stress in the study groups is depicted in Figure 4. Further, the mean micro-hardness in the study groups is shown in Figure 5.

## Discussion

The inadequate sealing at the interface between the restoration and dental hard tissue can lead to marginal discoloration, secondary caries, and pulpitis so this area is of clinical concern. Protection of exposed dentine against bacteria and their toxins is considered a prime requirement of tooth restoration [16].

AgNPs antimicrobial mechanism was attributed to their ability induce membrane damage through production of free radicals. These free radicals which derived from the surface of AgNPs were responsible for

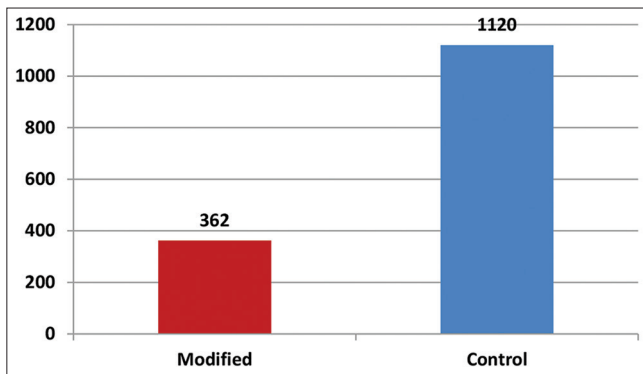


Figure 3: Mean colony forming unit in the study groups

the antimicrobial activity. AgNPs lead to the formation of “pits” in cell wall of the bacteria, through these pits it could enter into the periplasm and destroy the cell membrane. In addition, AgNPs resulted in the leakage of cytoplasmic component. Furthermore, the AgNPs could increase the decomposability of genome DNA. The antimicrobial effect of AgNPs could be attributed to their ability to inhibit enzymatic system of the respiratory chain and later DNA synthesis [17]. In our study, the antibacterial activity in the modified group may be attributed to contact of bacterial cells with surface of modified specimens as the SEM examination showed that there were less bacteria attached to the surface of modified specimens.

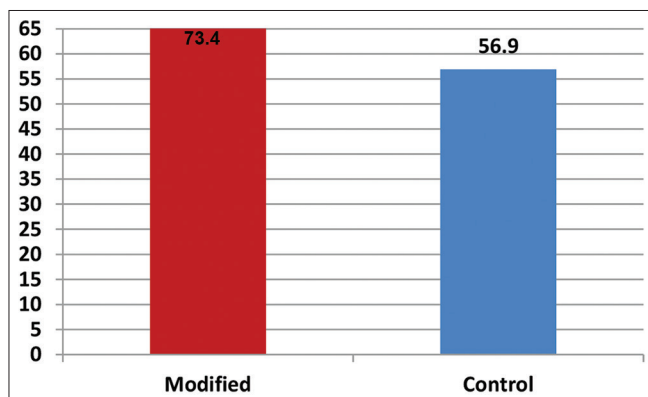


Figure 4: Mean flexure stress (MPa) in the study groups

Clinically, the common problems encountered with the use of porcelain-fused-to-metal and all-porcelain crowns are the fracture or detachment of the veneering porcelain such that more serious cases may necessitate the removal and remake of the entire restoration surface microcracks may inevitably form on porcelain crowns due to excessive occlusal force or cuspal interference. These surface microcracks can grow under subcritical fatigue loads and increase the risk of porcelain fracture, eventually resulting in premature failure of a restoration [1].

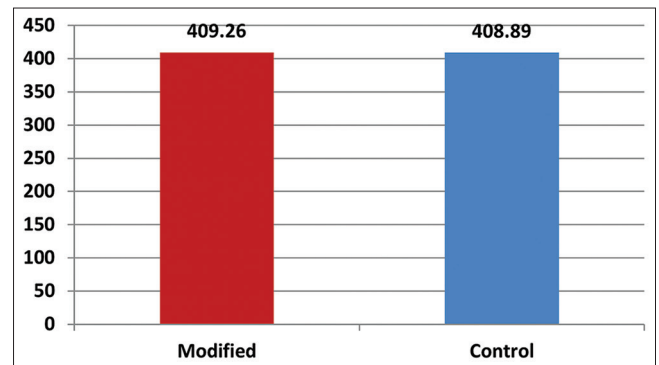


Figure 5: Mean micro-hardness in the study groups

Porcelain achieves appropriate optical properties in the oral cavity, so it is used in aesthetic dentistry. However, the requirements for porcelain are not purely cosmetic; it also needs to be adequately strong to withstand masticatory functions and to be durable [18]. Porcelain can be toughened by incorporation of fine particles of metals into the matrix. Toughening mechanisms act behind the crack tip to resist its opening.

The lamella structures hold the fracture surfaces together after the crack has propagated through the matrix. Cracks and microcracking – smaller cracks form in the material around the main crack – relieve the stress at the crack tip by effectively increasing the material’s compliance and transformation toughening [14]. The addition of AgNPs in the present study is mainly for its antibacterial effect; yet, it is expected to have influence on the flexural strength and microhardness of the studied porcelain. AgNPs modified porcelain samples (Group II) demonstrated higher flexural strength ( $73.4 \pm 6.8$  Mpa) than the control group (Group I) ( $56.9 \pm 11.9$ ). This increasing in flexural strength may be due to the effect of AgNPs addition to the glass matrix which generated a compressive stress when cooling down to room temperature. This is because the coefficient of thermal expansion of AgNPs is higher than that of glass. Another possible mechanism is the crack-bridging phenomenon that occurs when a ductile metal acts to inhibit the propagation of cracks. This inherent ductility of the metallic phase thus inhibited the propagation of cracks running through the AgNPs. Finally, an ion-exchange reaction between the sodium ions and silver ions in the glass matrix produced compressive stress at the surface [1].

In general, the strength of porcelains with particles dispersed in the glass matrix is enhanced by the suppression of cracking through deflection and bridging mechanisms [1], [18]. The results of this study were in accordance with the results obtained by Dlouhy *et al.* [19] as the inherent ductility of the metallic phase can be utilized to inhibit propagation of a running crack through the nanoparticles; in other words, a crack can be bridged by stretching the nanoparticles.

This study revealed that no significant difference between modified and control group as regards micro

hardness in contrary Mitsunori *et al.* [18] found that addition of AgNPs significantly increased Vickers hardness of the NS porcelain. The observed increase in Hv for specimens indicates that the specimens were more effectively sintered in the presence of the AgNPs. The difference in results might be attributed to specimen preparation.

## Conclusion

Under the limitations of this study, following conclusions could be detected:

1. AgNPs addition to porcelain was confirmed to be a very effective method in inhibition of bacterial growth through decreasing the colonizing activity.
2. The addition of AgNPs has been increased the flexural strength of dental porcelain.
3. The addition of AgNPs has no effect on surface hardness of dental porcelain.

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