## "EVALUATION OF THE TRANSVERSE STRENGTH OF THE HEAT CURE PMMA RESIN REINFORCED WITH VARIOUS CONCENTRATIONS OF TWO DIFFERENT NANOPARTICLES: AN *IN VITRO* STUDY"

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#### ABSTRACT

**Purpose:** The purpose of the study was to evaluate the effect of titanium dioxide and zirconia nanoparticles on transverse strength of heat cure PMMA resin routinely used for complete denture fabrication.

**Method:** One hundred samples of PMMA resin were made and divided into five groups (20 samples for each group) . The test specimens were divided into five groups depending on the concentration of reinforcing nanoparticles as Group 1,2,3,4 and 5; Group 1: PMMA unreinforced (control group), Group 2:

PMMA reinforced with 2.5% nanozirconia, Group 3: PMMA reinforced with 5% nanozirconia, Group 4: PMMA reinforced with 2.5% titanium dioxide nanoparticles, and Group 5: PMMA reinforced with 5% titanium dioxide nanoparticles. Universal testing machine was used to conduct a three-point bending test and evaluate the transverse strength of samples. Comparison of mean transverse strength for various groups was carried out by employing one way analysis of variance and Bonferroni *post hoc* tests.

**Results**: The highest and lowest mean transverse strength were observed in Group 3 and 1, respectively. Bonferroni *post hoc* test showed increase in transverse strength after reinforcement to be statistically significant between all the groups (P = .05) except between the samples of group G1 and G5 and G2 and G3.

**Conclusion:** Addition of nanoparticles in all concentrations significantly increased transverse strength of heat cure PMMA resin as compared to control group. The best result was obtained after adding 5% of nanozirconia particles to the conventional heat polymerized acrylic resin.

#### **KEYWORDS**

PMMA, nanozirconia, titanium dioxide nanoparticles, transverse strength

#### **1. INTRODUCTION**

Edentulism has been a matter of great concern to a number of people, be it partial or complete. Replacement of teeth by artificial substitutes plays a vital role in leading a normal life.<sup>[1]</sup> Since 1930, acrylic resin polymethyl methacrylate (PMMA), or acrylic, resin has been the most popular material for denture prosthesis fabrication. Not only does it provide an accurate fit, good esthetics, and stability in the oral environment but also is easy to handle in laboratory and clinic. Despite many advantages this material exhibits certain limitations which render failure to fulfill mechanical requirements for dental applications. These include low fracture resistance and plaque accumulation<sup>[2]</sup>, high coefficient of thermal expansion and relatively low modulus of elasticity. Fracture of maxillary dentures is twice more common than that of mandibular dentures, Fractures caused outside the mouth are usually a result of heavy impact forces or a high stress rate. On the other hand, denture fracture occurring inside the mouth are usually caused in function due to a fatigue phenomenon, i.e., low and repetitive stress rate which

commonly occurs over a period of time and results in such functional fractures. This type of fracture is typically seen in midline of maxillary dentures than in mandibular dentures.<sup>[3]</sup>

Release of residual monomer from the dentures has reported to cause irritation to mucosa.<sup>[5]</sup> Also being a radiolucent material it cannot be imaged using standard radiographic techniques hence in cases of accidental ingestion of prosthesis, aspiration or traumatic impaction of dental appliance, their detection can become painstaking and invasive procedures may need to be carried out.<sup>[4]</sup>

In order to improve the mechanical properties of acrylic resins, several attempts in the past have been made. These include chemical modification or reinforcement with glass fibers, metal oxides and nanoparticles.

Recently, incorporation of nanofillers has been suggested to improve mechanical properties of PMMA. Fine particle size enables the homogenous distribution of nanofillers in PMMA matrix and has reportedly improved the thermal properties of PMMA by increasing its thermal stability compared with PMMA. However, size, shape, type and concentration of nanoparticles added affect the properties of resin.<sup>[5]</sup>

The few studies conducted on the effect of nanoparticles on the transverse strength have been more or less conclusive and unclear.<sup>[6]</sup>

Titanium dioxide and zirconium oxide nanoparticles have become popular as reinforcement nanofillers recently. Titanium is used since it increases the surface hydrophobicity, reduces the adherence of biomolecules, aids in colouring, has antimicrobial properties and improves mechanical properties of PMMA resins. Spherical particles of titanium dioxide have been used to improve the flexural strength as spherical particles increase the polishability and mechanical properties. Other structures such as nanotubes and fibers which have been recently discovered show much better properties.<sup>[7]</sup>

Zirconia has exhibited excellent biocompatibility and being white in colour it is less likely to interfere with esthetics.<sup>[4]</sup>

Zirconium oxide nanoparticles mechanically reinforce the polymers and allows for high impact strength, fracture toughness, hardness and density of the reinforced PMMA matrix.<sup>[8]</sup>

Modifying nanozirconia powder by coating with a layer of trimethoxysilypropylmethacrylate (TMSPM) renders more radiopacity as it increases the radiographic density and allows more absorption of radiation.<sup>[4]</sup>

Hence, the purpose of this study was to evaluate and compare transverse strength of heat cured PMMA resin after its reinforcement with zirconium oxide and titanium dioxide nanoparticles in concentrations 2.5 wt % and 5wt % each.

#### 2. MATERIAL AND METHODS

#### 2.1 The study proceeded as follows:-

i. Fabrication of metal dies-

Three stainless steel metal dies of dimensions 65mm x 10mm x 3 mm were fabricated. The selection for dimensions of the dies was based on ADA specification no. 12.

ii. Fabrication of test samples-

Preparation of moulds for fabrication of wax pattern:

- a. The stainless steel metal dies were impressed upon putty material (Affinis, New Delhi, India) so as to create a mould space. Molten wax (No.2, Rolex, Ashoosons Dental Care Pvt. Limited, Delhi, India) was then poured onto the mould spaces so created and left to cool. The wax patterns of dimension 65 x 10 x 3mm as per ISO 1567 standardization were obtained after cooling. In this manner, a total of 100 wax patterns were obtained.
- b. These patterns were invested in Type II dental plaster (Dentex, India). After the investing material had set, the flasks were placed for dewaxing in a conventional water bath. The moulds so created were thoroughly flushed with hot water. The flasks were left to cool followed by application of a layer of separating media (DPI Cold Mould Seal, Bombay Burmah Trading Corporation Ltd., Mumbai, India) to prepare the flasks for packing.
- c. Appropriate amount of heat cure acrylic resin was weighed with the help of electronic weighing machine (Jeejex Digital Electronics SF-400, Jiya Sales, India).and mixed with monomer in the ratio 3:1 by volume i.e. 3 parts of polymer and one part of monomer

iii. Division of samples into various groups (Fig.1):

G1 - Control (DPI Heat Cure, Bombay Burmah Trading Corporation Ltd., Mumbai, India)

G2- DPI Reinforced with 2.5 wt% nanozirconia particles (ZrO2, Purity 99.9%, Average particle

size: 30-50nm, NanoResearch Lab, Jamshedpur, Jharkhand, India)

G3 - DPI Heat Cure with 5 wt% nanozirconia particles

G4 - DPI Heat Cure with 2.5% titanium dioxide nanoparticles (TiO<sub>2</sub>, Anatase, Purity: 99.9 %,

Average particle size : 10-20nm, NanoResearch Lab, Jamshedpur, Jharkhand, India)

G5 - DPI Heat Cure with 5 wt% titanium dioxide nanoparticles



- Fig. 1 Specimens for each group
  - a. Preweighed nanoparticle powder was separately added to the acrylic resin powder to form the desired formulation and thoroughly mixed using a mortar and pestle (Uniteck Scientific & Electronic Industry, Chandigarh, India).
  - b. Proportionate amount of the monomer and the polymer was taken in the mixing jar and thoroughly mixed until dough stage was attained. The acrylic dough was kneaded and packed

into the flask (Varsity Flask, No-7, S.S. Products, India). Trial closures were carried out using a hydraulic press (Unident Instruments India Private Limited, India) to remove the excess flash and provide uniform distribution of acrylic dough throughout the u cavity. The flasks were then secured tightly to maintain the pressure and bench cured for 30 minutes to allow proper penetration of monomer into polymer. The flask was then immersed in a thermostatically controlled water bath such as acrylizer (Unident Instruments India Private Limited, India) at room temperature. The temperature of water bath was raised to 74 C, held for 8 hours, then raised to 100 C for an hour. After the completion of the curing cycle, the flask was removed from the water bath and bench cooled for 30 minutes, immersed in cool tap water for 15 minutes prior to deflasking. Samples were then contoured using carbide bur, finished with sandpaper and polished using slurry of coarse pumice. The width and thickness of each samples was measured using a digital vernier calliper (PRECISE, Sudershan Pvt Ltd, Delhi, India) with a least count of 0.01mm. Since width and thickness were factors assessed for determining transverse strength, only the resin samples with a slight variation in dimensions up to 0.3mm were included in the study.

# In this manner, a total of 100 acrylic samples divided into five groups each containing 20 samples of compression moulded heat cure acrylic denture base resin were produced.

All the specimens were stored in distilled water at a temperature of 37° C for 48 hours prior to transverse strength testing.<sup>[9]</sup>

#### 2.2 Calculation of Transverse Strength:

To determine transverse strength, fracture load was measured using the three-point bending test according to ISO 178 on a universal testing machine (ASIAN Test Equipments, Micronix Intruments, India). Then specimens were placed on a 3-point flexure apparatus with the support span of 50 mm. Load was applied at the midpoint of the sample with a crosshead speed of 5mm/min until the specimen fractured and fracture load was recorded (Fig. 2).<sup>[7]</sup> The transverse strength values of each specimen were derived using formula:

 $2bd^2$ 

where TS is the transverse strength (in MPa), W is the fracture load (N), L is the distance between the two supports, b is the specimen width, and d is the specimen thickness. Data was analysed using computer software STATA and SPSS-20.0, IBM software, Chicago.



Fig. 2 - Testing the specimen on Universal Testing Machine

#### 2.3 SCANNING ELECTRON MICROSCOPIC (SEM) ANALYSIS:

SEM (FEI Nova NanoSEM 450, USA) was used to examine the surface of fractured cross-section of the specimens. The acceleration voltage, used to perform SEM evaluation, was set at 10 kV and the working distance was 5.3 mm with a 3 spot size. The specimens were gold sputtered to provide conductivity to the material. Images were recorded at magnifications 500x and 5000x to study distribution of particles (Fig. 3a-e and Fig. 4a-e).



- Fig. 3 a SEM image for sample G1 at 500x
- Fig. 3 b- SEM image for sample G2 at 500x
- Fig. 3 c SEM image for sample G3 at 500x
- Fig. 3 d SEM image for sample G4 at 500x
- Fig. 3 e- SEM image for sample G5 at 500x



- Fig. 4 a SEM image for sample G1 at 5000x
- Fig. 4 b- SEM image for sample G2 at 5000x
- Fig. 4 c SEM image for sample G3 at 5000x
- Fig. 4 d SEM image for sample G4 at 5000x
- Fig. 4 e- SEM image for sample G5 at 5000x

#### 3. RESULTS

In the present study all reinforced groups showed a definite increase in mean transverse strength compared to that of the control group.

The mean transverse strength (**MPa**) obtained for control and reinforced groups have been summarized in **Table 1.** Group 3 (**G 3**) showed maximum transverse strength, followed by **G2**, **G4**, **G5** and minimum strength was shown by **G1**.

One-way ANOVA (Table 2) showed significant differences among the groups (P<0.001).

For the comparison between groups, i.e., for multiple group comparisons, a post hoc Bonferroni test (Table 3) was applied, which showed that the increase in transverse strength was statistically significant for all experimental groups in comparison with samples of the unreinforced PMMA group, except between G1 and G5 (P= 1.000) and G2 and G3 (P=.74).

Table 1- Descriptive statistics of transverse strength values (MPa) obtained for control and two types of nanoparticles:

Groups	N	Mean	Standard	Standard	
		(MPa)	<b>Deviation</b>	Error Mean	
G1	20	94.800	9.83483	2.19913	
G2	20	134.82	12.66516	2.83202	
G3	20	148.96	28.65139	6.40664	
G4	20	119.26	6.91676	1.54663	
G5	20	101.98	14.43716	3.22825	

 Table 2: Intergroup comparison (One-way ANOVA) of mean transverse strength (MPa) among various studied groups:

Transverse	Sum of	df Mean		F	Sig.
strength (MPa)	Squares		Square		
Between Groups	40373.034	4	10093.258		
Within Groups	25351.795	95	266.861 37.822		0.000*
Total	65724.828	99			

Table 3: Multiple group comparison (post hocBonferroni test) of mean transversestrength (MPa) among various studied groups .

(I)	(J)	Mean	Std.	Sig.	95% Confidence Interval	
Group	Group	Difference	Error		Lower	Upper Bound
		(I-J)			Bound	
G1	G2	-40.02500 <sup>*</sup>	5.16586	.000*	-54.8721	-25.1779
	G3	-54.15700 <sup>*</sup>	5.16586	.000*	-69.0041	-39.3099
	G4	-24.46100 <sup>*</sup>	5.16586	.000*	-39.3081	-9.6139
	G5	-7.17825	5.16586	1.000**	-22.0253	7.6688
G2	G1	40.02500*	5.16586	.000*	25.1779	54.8721
	G3	-14.13200	5.16586	.074**	-28.9791	.7151
	G4	15.56400 <sup>*</sup>	5.16586	.033*	.7169	30.4111
	G5	32.84675 <sup>*</sup>	5.16586	.000*	17.9997	47.6938
G3	G1	54.15700 <sup>*</sup>	5.16586	.000*	39.3099	69.0041
	G2	14.13200	5.16586	.074**	7151	28.9791
	G4	29.69600*	5.16586	.000*	14.8489	44.5431
	G5	46.97875 <sup>*</sup>	5.16586	.000*	32.1317	61.8258

G4	G1	24.46100 <sup>*</sup>	5.16586	.000*	9.6139	39.3081
	G2	-15.56400 <sup>*</sup>	5.16586	.033*	-30.4111	7169
	G3	-29.69600*	5.16586	.000*	-44.5431	-14.8489
	G5	17.28275 <sup>*</sup>	5.16586	.012*	2.4357	32.1298
G5	G1	7.17825	5.16586	1.000**	-7.6688	22.0253
	G2	-32.84675*	5.16586	.000*	-47.6938	-17.9997
	G3	-46.97875 <sup>*</sup>	5.16586	.000*	-61.8258	-32.1317
	G4	-17.28275*	5.16586	.012*	-32.1298	-2.4357

\*statistically significant

\*\*statistically non-significant

#### 4. DISCUSSION

Polymethylmethacrylate (PMMA) is the most popular material used widely in various fields of Prosthodontics. However, the impact strength and fatigue strength of this material are not found to be entirely satisfactory. The ultimate flexural strength (also called transverse strength or modulus of rupture) of a material reflects the potential of the material to resist fatal failure under a flexural load.<sup>[7]</sup> Hence, high flexural strength can be regarded as an important determinant of the success of dentures. Compressive, tensile and shear strengths collectively form the flexural strength of a material. As the flexural strength increases, the force required to fracture the material also increases.<sup>[10]</sup>

Transverse strength represents the type of loading borne by a denture in the mouth. A high value of transverse strength of denture base acrylic is desirable as it provides a superior clinical performance by the dentures.<sup>[1]</sup> Fatigue phenomenon is the low and repetitive stress rate which commonly occurs over a period of time. This fatigue failure is not dependent on strong biting forces. Relatively small stresses caused by mastication over a period of time can contribute to formation of a small crack , which propagate through the denture thereby resulting in a fracture. Fractures of denture occur essentially because of concentration of stresses and increased flexing.<sup>[2]</sup>

Recently, incorporation of nanofillers has been suggested to improve PMMA properties. The structure of material that has particles of a nanometer size possesses special properties. It can be rendered to the high ratio of surface area to volume. <sup>[11]</sup> Amongst a variety of nanoparticles available like silver, copper, zinc, silicon, titanium and their oxides, titanium dioxide has gained importance recently because of its higher photocatalytic activity, high stability, low cost and safety towards both humans and the environment. On the other hand some studies found that titanium dioxide nanoparticles did not improve the transverse strength of PMMA. This could be attributed to clustering of the particles within the resin matrix that weakened the denture prosthesis. <sup>[12]</sup>

# It was found that $TiO_2$ nanoparticles have an effect on thermal stability of PMMA resin and that they caused a decrease in the thermal expansion coefficient and contraction. A decrease in elastic modulus, transverse strength and toughness was reported. Addition of nanozirconia was suggested to improve the mechanical properties of PMMA. This helped in increasing the impact strength, transverse strength, compressive strength, fatigue strength, as well as its fracture toughness and hardness.<sup>[11]</sup>

Use of nanoparticles is based on the principle that reduction of filler size increases the mechanical properties of resins. Spherical particles of titanium dioxide have been reported to improve the transverse strength as they increase polishability. It has also shown an increased surface hydrophobicity, reduced adherence of biomolecules has aided in colouring.<sup>[7]</sup>

The quest has been on for the most suitable concentration of different nanoparticles which can be added to the acrylic resin so that transverse strength is improved manifolds. Massive changes in the colour of acrylic have been reported with use of nanoparticles above 5% concentration. Therefore, two concentrations 2.5% and 5% were selected.<sup>[13]</sup>

The increase in transverse strength at 5% concentration of nanozirconia can be attributed to the high interfacial shear strength between nanofiller and matrix due to formation of cross-links or supra molecular bonding which covers or shields the nanofillers. This in turn prevents propagation of crack. Also the crack propagation can be changed by improving the bonding between nanofiller and resin matrix. An increase in transverse strength that occurred with addition of 2.5wt% zirconium dioxide nanoparticles can be attributed to the uniform distribution of the very fine size of nanoparticles that allows them to enter between linear macromolecular chains of the polymer.<sup>[4]</sup>

The SEM micrograph studies in this study showed good surface characteristics with different nanozirconia concentrations. Moreover, a uniform distribution of particles was assumed as no big agglomeration was found. These SEM analytical studies revealed that, as the concentration increased, the polymer matrix got filled with nanoparticles that stopped crack propagation, resulting in stronger material. Uniform dispersion of the nanoparticles into the resin matrix filled the inter-polymeric chain spaces, which shows the importance of the additive content of the nanoparticles.<sup>[14]</sup>

Increase in transverse strength on addition of titanium dioxide nanoparticles in concentration 2.5% in PMMA matrix can be attributed to uniform dispersion of the small sized filler particles. This is responsible for the improved fracture resistance of PMMA. Addition of titanium dioxide nanoparticles up to 2.5 % increased the strength, above which the strength decreased. Dispersion of TiO<sub>2</sub> nanoparticles in PMMA matrix adversely affects the degree of polymerization which in turn leads to increase in the level of residual unreacted monomer that acts as plasticizer. Incorporation of nanoparticles causes these particles to agglomerate and aggregate. The agglomerated compounds can act as stress concentrating center in the matrix and adversely affect mechanical properties of the polymerized material. It is easily noted that the content of nano additives is of critical importance.<sup>[15]</sup>

By using scanning electron microscopy in our study, it was observed that, at a concentration of 2.5%, nanoparticles of titanium dioxide were well distributed in the specimen. The particles maintained their original size and had an active role in improving the mechanical properties. However, when the concentration was increased to 5.0%, the SEM images demonstrated that the nano-oxides had agglomerated to a different extent, which resulted in a decrease in the mechanical properties of the resin, as reported earlier. <sup>[11][15]</sup>

In the present *in vitro* study, only one mechanical property, i.e., transverse strength was taken into consideration. Therefore, further studies should be carried out in *in vivo* conditions to understand its effect in the oral environment.

#### 5. CONCLUSION

Within the limitations of this study, following conclusions were drawn:

- Addition of titanium dioxide nanoparticles beyond the concentration of 2.5 % decreased the transverse strength of conventional heat polymerized acrylic resin.
- The best result was obtained with incorporation of 5% nanozirconia particles to the conventional heat polymerized acrylic resin.
- According to the results of this *in vitro* study, it can be concluded that reinforcement of PMMA resulted in significantly higher transverse strength as compared to that of unreinforced resin.
- However, only one mechanical property i.e., transverse strength was taken into consideration in this *in vitro* study, and the effect of nanoparticles on other mechanical properties was not studied.
   Further studies considering other mechanical, esthetic and biological properties can be carried out.

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#### **COMPETING INTEREST**

Authors have no competing interest

#### **AUTHORS' CONTRIBUTIONS**

Author 1- Executed the study and performed statistical analysis

- Author 2 and 3- Designed and planned the study
- Author 4- Managed the analyses of the study
- Author 5 and 6- Managed the literature searches

All authors have read and approved the final manuscript.

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