ORIGINAL RESEARCH

Chemical Structure and Physical Properties of Heat-cured Poly(methyl methacrylate) Resin Processed with Cycloaliphatic Comonomer: An *In Vitro* Study

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Abstract

Aim: The purpose of this *in vitro* research is to chemically characterize polymethyl methacrylate (PMMA) processed with 10% and 20% (v/v) tricyclodecane dimethanol diacrylate (TCDDMDA) comonomer. It also aimed to assess the degree of conversion (DC) and glass transition temperature (T_g) of the formed copolymers.

Materials and methods: The experimental groups were processed with the TCDDMDA comonomer (10% and 20% v/v), whereas the control group was processed only with the methyl methacrylate monomer. The copolymerization was studied by nuclear magnetic resonance (NMR) spectroscopy. The surface characteristics and composition (wt%) were studied by field-emission scanning electron microscopy (FESEM) and energy dispersive X-ray (EDX) spectroscopy (cuboidal specimen; 5 mm × 5 mm × 3 mm), respectively. The DC and T_g of the formed copolymers (powdered form) were analyzed by Fourier transform infrared spectroscopy and differential scanning calorimetry, respectively. One-way analysis of variance with *post hoc* Bonferroni test was used to compare the mean values of DC% and T_g among the groups.

Results: The newly formed copolymer [P(MMA-*co*-TCDDMDA)] was chemically characterized by NMR and FESEM-EDX. The DC and T_g of the experimental groups were higher than the control. Tricyclodecane dimethanol diacrylate at 20% (v/v) concentration showed the highest DC and T_g.

Conclusion: The addition of TCDDMDA comonomer improved the DC and T_g of the formed copolymer.

Clinical significance: The P(MMA-*co*-TCDDMDA) copolymer is expected to improve the mechanical properties and biocompatibility of the denture base acrylic resin. This would result in improved denture quality and durability, thereby, imparting a better quality of life to the geriatric population.

Keywords: Comonomer, Copolymer, Degree of conversion, Glass transition temperature, Tricyclodecane dimethanol diacrylate.

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Introduction

Knowledge about the composition of a polymer plays a pivotal role in understanding the chemistry and clinical application of the material. The polymer composition (physical, chemical, and structural information of the molecules) and its distribution can be deduced using the most common method called nuclear magnetic resonance (NMR) spectroscopy. This is achieved by observing and studying the peaks of the NMR spectra.[1]

Polymethyl methacrylate (PMMA) is a widely used denture base resin material for the past 70 years. Although there are several advantages, unpropitious reactions to PMMA have been documented in the literature.[2,3] The cell viability is chiefly influenced by the unreacted residual monomers in the resin specimens. The residual monomer content is predominantly governed by the degree of conversion (DC) achieved in the polymerization reaction.[4,5] In denture base (vinyl) polymers, the DC is calculated from the ratio of aliphatic C═C double bonds remaining in a cured specimen to C═C bonds in the uncured specimen. Fourier transform infrared (FTIR) spectroscopy is one of the most commonly and widely used methods to evaluate the DC.[6] This monomer to polymer conversion affects the mechanical properties of the polymerized resin.[7–9] Improper or incomplete conversions affected the glass transition temperature (T_g) of the polymers. Hence, these polymers may be considerably more unstable at oral temperatures than highly converted polymers.[9]

From a clinical point of view, high credits are ceded to T_g, as temperature generally influences the physicomechanical properties of polymers. Denture base polymers are amorphous in nature which at sufficiently low temperature possess rigidity with brittleness. With the increase in temperature, they typically behave as flexible solids, rubber, or even as a viscous liquid.[10] This transition from rigid brittle to flexible material is indicated by T_g.[11] Thermal properties inclusive of T_g of denture base polymers have been primarily investigated by differential scanning calorimetry (DSC).[12–14]

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Numerous modifications were attempted to improvise the physicomechanical and biological properties of PMMA. In recent years, monomeric modifications are becoming popular.[15] However, the added comonomer has to interact and copolymerize with the proprietary methyl methacrylate (MMA) monomer. This is because copolymerization determines the properties of the denture base resins.[16] To ameliorate the mechanical properties of the denture base polymer with a comonomer, it mandates a high interaction between the proprietary and the comonomer. This can be asseverated using field-emission scanning electron microscopy (FESEM) with energy dispersive X-ray (EDX) spectroscopy. For attaining detailed, high-resolution images of denture base surfaces, scanning electron microscopy has been widely replaced by FESEM. A microanalytical system is used in EDX, which depends on the X-ray excitation of specimens for peak characterization based on the atomic structure.[17]

Tricyclodecane dimethanol diacrylate (TCDDMDA) is a novel cycloaliphatic, difunctional, dual-reactive, and cross-linking monomer. It has a unique steric hindrance property, whereby it slows down the polymerization rate and increases the monomer to polymer conversion. Hence, this study aims to deduce the organic structure and composition of the PMMA processed with 10% and 20% (v/v) TCDDMDA comonomer by NMR (^{1}H-NMR and ^{13}C-NMR) spectroscopy and FESEM-EDX, respectively. It also aims to assess the DC and T_g of the formed copolymers.

Materials and Methods

Nuclear magnetic resonance and DSC were carried out at the Department of Chemistry and Centralized Instrumentation and Service Laboratory, respectively, Annamalai University, Tamil Nadu. Field-emission scanning electron microscopy was performed at the University Science Instrumentation Center, Alagappa University, Tamil Nadu. Degree of conversion was executed at Centre for Nanoscience and Technology, KSR Institutions, Tamil Nadu.

Heat-cured denture base acrylic resin (DPI® Heat Cure; Dental Products of India, Mumbai, India) and cycloaliphatic monomer, TCDDMDA (Sigma-Aldrich, Germany; CAS Number 42594-17-2), were used in this study. Experimental monomers were prepared by substituting 10% and 20% (v/v) of TCDDMDA in MMA. Methyl methacrylate without TCDDMDA served as control. The monomers were then stored in three separate identical dark glass bottles and labeled with appropriate concentrations. The investigator was blinded from the concentrations by covering the labels with opaque stickers. The bottles were then randomly numbered from one to three (Bottles 1, 2, and 3) to avoid expectation bias.

Specimen Preparation

One representative cuboidal specimen of each group (5 mm × 5 mm × 3 mm) was prepared for FESEM-EDX. For all other experiments, disk-shaped (22 mm × 2 mm) specimens (n = 10 for each DC and DSC experiment) were fabricated. To obtain mold space, plastic acrylic dies of appropriate dimensions were used. The polymer and the monomer were proportionated at a 3:1 ratio in a porcelain jar. The dough forming time was 8 minutes for the control group and 15 minutes for experimental groups. The mold spaces were then packed with the dough at a packing pressure of 3,500 psi in mechanical press (Sirio Dental Srl, Meldola FC, Italy) for 10 minutes and then heat-cured at 74°C for 2 hours followed by terminal boiling treatment at 100°C for 1 hour in an acrylizer (Unident Instruments India Pvt Ltd). For NMR, DC, and DSC analyses, the polymerized disk specimens of all groups were ground to powder particles with a clean tungsten carbide bur. The obtained powder was then sieved to obtain uniform particle size.[18] All the polymeric specimens (powder/solid) were prepared by a single investigator to avoid bias. The study groups and their abbreviations are tabulated in Table 1.

Table 1: Groups for the experiments

		Groups		
Experiment		*Control*	*Experimental*	
NMR	^{1}H-NMR	H NMR0	H NMR10	H NMR20
	^{13}C-NMR	C NMR0	C NMR10	C NMR20
FESEM-EDX		P(MMA)	P(MMA-*co*-10TCDDMDA)	P(MMA-*co*-20TCDDMDA)
DC				
DSC				

NMR, nuclear magnetic resonance; FESEM, field-emission scanning electron microscopy; EDX, energy dispersive X-ray; MMA, methyl methacrylate; TCDDMDA, tricyclodecane dimethanol diacrylate; DC, degree of conversion; DSC, differential scanning calorimetry

A digital NMR spectrometer (Ultrashield™ 400 Plus; Bruker BioSpin, Karlsruhe, Germany) was used for the ^{1}H- and ^{13}C-NMR spectra of the groups. A 20 mg and 30 mg powdered resin specimen was taken in a thin glass tube for ^{1}H- and ^{13}C-NMR, respectively. One milliliter of deuterated chloroform ($CDCl_3$) was added to the tube and shaken well to completely dissolve the powdered resin. Tetramethylsilane was preferred as an internal standard.

Field-emission scanning electron microscopy (FEI™ Quanta™ FEG 250, Oregon, USA) was used for characterizing the surface morphology of the groups. This machine was able to take images with high resolution. The cuboidal unfinished specimens were placed on the specimen mounts with appropriate labeling. Beam to specimen surface distance was 10 mm. The specimen surfaces were scanned at 10 kV with 3,000× magnification. The chemical composition of the specimens was measured with the aid of an EDX spectroscopy (Bruker India Scientific Pvt Ltd, Mumbai, India). The X-ray incident photons bombard the electrons of atoms, and the distribution of bombarded particles was detected through EDX. The graphical representation of the same was processed by in-built microanalysis software (Bruker Quantax Esprit).

The DC of monomer to polymer was determined by FTIR (Spectrum 100, Perkin Elmer, Singapore) with 4 cm^{-1} resolution and 16 scans at absorbance (Abs) mode. All heat-cured specimens, as well as the uncured material (n = 10 for each group), were analyzed. To obtain the FTIR spectrum of cured specimens, 2 mg of specimen powder was mixed with 100 mg of dry potassium bromide (KBr) powder. The mixture was pressed into a disk of 0.5 mm thickness using a special mold and hydraulic pellet press. This pellet was placed vertically in the specimen compartment, and an infrared beam was transmitted. To prepare the uncured material, the prepolymerized resin powder was heated at 70°C for 30 hours.[18] This led to degradation of benzoyl peroxide and eschewed polymerization reactions of the material after mixing with the monomer. After mixing, the resin paste was placed between two polyethylene films, pressed to form a very thin film, and the infrared beam was passed. The DC was determined from the ratio of the Abs intensities observed for aliphatic C═C and the carbonyl bond C═O at 1,638 cm^{-1} and 1,720 cm^{-1} wavenumbers, respectively. These wavenumbers are frequently used as the internal standard of polymerized material for both the cured and uncured materials. The DC was calculated using the following equation:

$$DC\% = \left[1 - \frac{\left(\frac{Abs\ 1640}{Abs\ 1720}\right)_{Cured}}{\left(\frac{Abs\ 1640}{Abs\ 1720}\right)_{Uncured}}\right] \times 100$$

For T_g, the specimens were tested in the DSC instrument (Netzsch, STA 449 F3 Jupiter®, Selb, Germany) under nitrogen. Twenty milligrams of the powdered specimen were placed in an aluminum holder and heated from 50°C to 200°C. The heating rate was set to 20°C minutes^{-1}. In order to erase the thermal history of the specimen, all calculations were done using the data acquired from the second cycle. The T_g was estimated with the in-built software (Proteus®) by drawing tangents on the heat flow curve at temperatures above and below the glass transition. The tangents were selected carefully because they have a significant effect on the reproducibility of the evaluation. It can be determined as the point of intersection of the bisector of the angle formed between the tangents with the measurement curve.

Statistical analysis was performed using SPSS Version 20.0 software. Preliminary results of the Shapiro–Wilks test indicated that the data were normally distributed ($p > 0.05$). Descriptive statistics, including mean, standard deviation (SD), standard error, maximum, and minimum, were calculated. Concerning inferential statistics, the level of significance between the three groups for DC and T_g was tested with one-way analysis of variance (ANOVA). To compare the groups, the *post hoc* Bonferroni test ($\alpha = 0.05$) was performed. $p < 0.05$ was considered for statistical significance.

Results

NMR Spectroscopy

The peak signals of ^{1}H-NMR of control and experimental groups are shown in Figure 1. The protons in $-OCH_3$ (δ 3.533–δ 3.532), $-CH_2$ backbone (δ 1.980–δ 1.744), and $-CH_3$ (H NMR0: δ 1.372–δ 0.769; H NMR10, 20: δ 1.370–δ 0.773) were seen in both control and experimental groups. Additional protons in $-OCH_2$ (δ 4.964), aliphatic ring (δ 4.062–δ 4.044, δ 3.937–δ 3.897, and δ 2.108), and CH (δ 2.986–δ 2.947) were observed exclusively in experimental groups. The peak signals of ^{13}C-NMR of control and experimental groups are shown in Figure 2. The carbon (C) atoms in >C=O (δ 178.14–δ 177.00), $-O-CH_3$ (δ 51.84), quaternary carbon (δ 44.88 and δ 44.53), saturated alkane CH (C NMR0: δ 25.01; C NMR10, 20: δ 29.69), and $-CH_3$ (δ 18.72 and δ 16.46) were seen in both control and experimental groups. Additional C-atoms in $-O-CH_2$ (δ 64.89) and tricyclic C-atoms (δ 40.51–δ 31.27) were observed exclusively in experimental groups. There were no significant spectral differences between the experimental groups. The presence of these additional protons and carbons in experimental groups affirms the copolymerization of TCDDMDA with MMA in PMMA.

FESEM-EDX

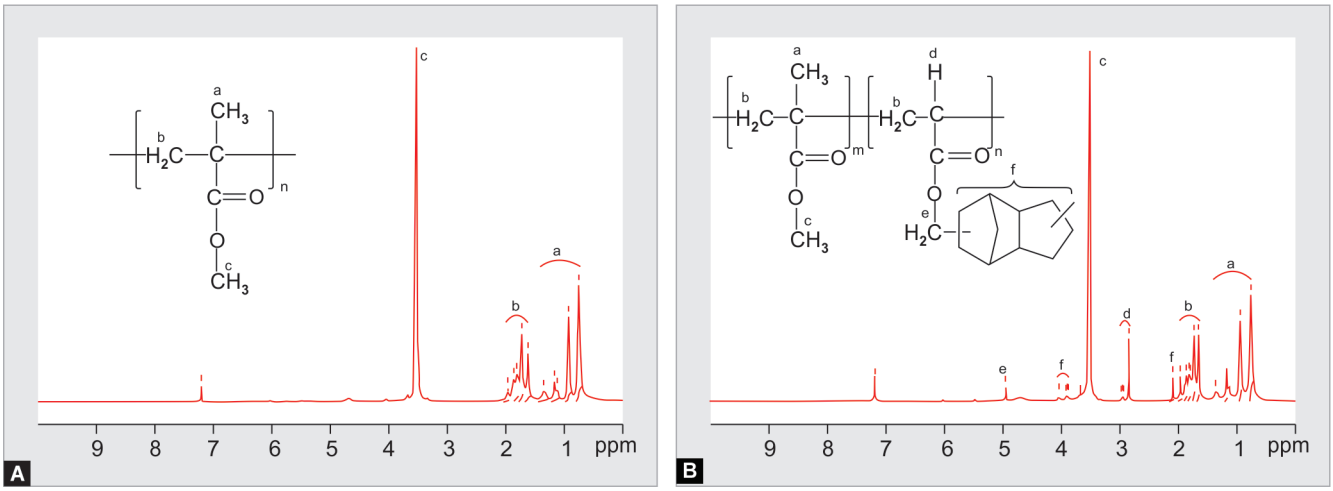
In FESEM, the P(MMA) group has a highly irregular rough surface (Fig. 3A) with a "continental" appearance. Experimental groups rather have a regular lamellar surface (Figs 3B and C) with a "sand-dune" appearance. The lamellar pattern is more regular in P(MMA-*co*-20TCDDMDA; Fig. 3C) than the other. On the basis of the EDX analysis, C (60.52 wt%) and O (39.48 wt%) were the elements in the P(MMA) group. The C-atoms in the P(MMA-*co*-10TCDDMDA) and P(MMA-*co*-20TCDDMDA) groups were 61.97 wt% and 66.18 wt%, respectively. This increase in the C-atoms in the experimental groups is due to the additional TCDDMDA monomer.

Degree of Conversion

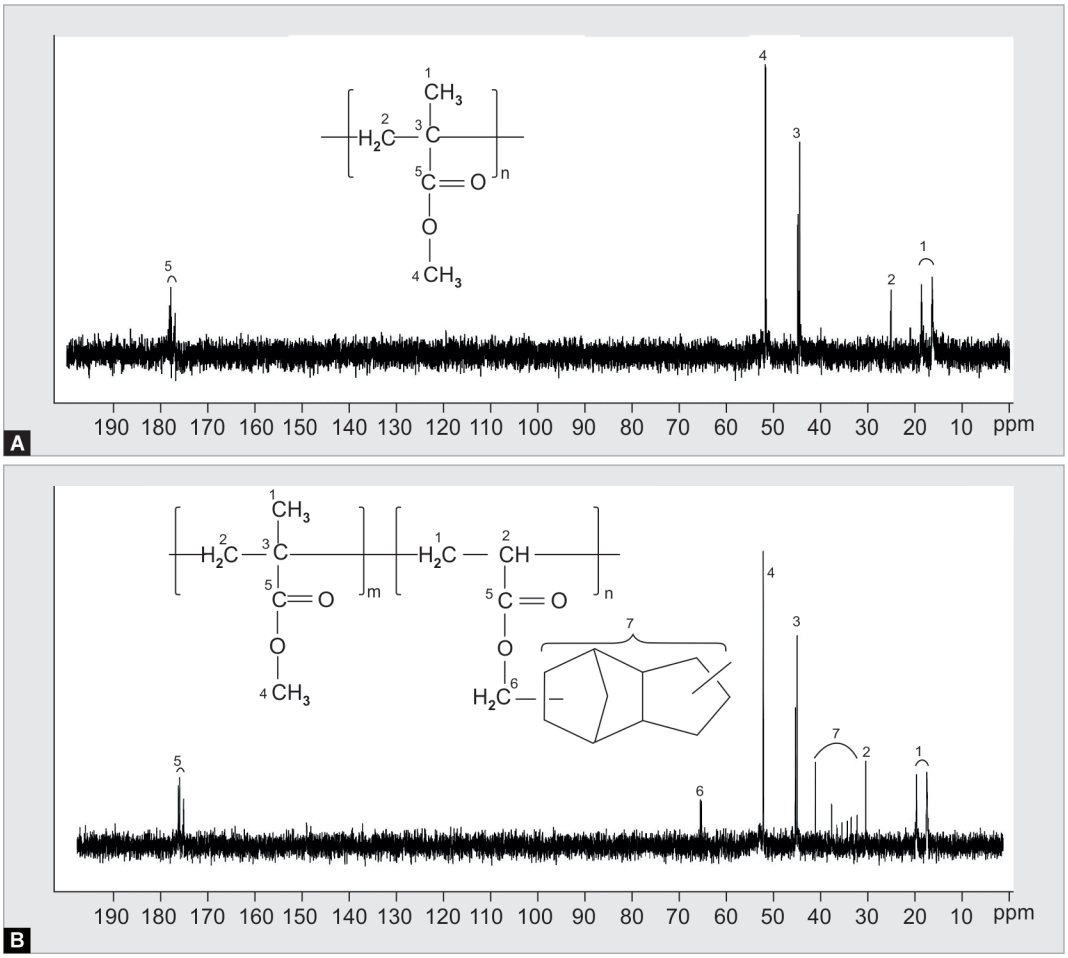
In the infrared charts of all groups (Fig. 4), two Abs peaks appeared at 1,638 cm^{-1} and 1,720 cm^{-1} for uncured specimens. This implies the presence of unreacted carbon–carbon double bond (C=C). The Abs peak at 1,638 cm^{-1} dramatically disappeared with the cured specimens of both experimental groups. This is inferred as a reduced residual monomer. However, for the cured specimen of the P(MMA) group, a weak Abs peak at 1,638 cm^{-1} did appear, which suggests the presence of residual monomer. Table 2 presents the mean (SD) of the control and experimental groups. One-way ANOVA showed the level of significant differences among the groups ($p = 0.000$). Bonferroni multiple comparison tests (Table 3) showed a statistically significant difference between the groups ($p = 0.000$). The DC of experimental groups was higher than that of the control group. The DC of P(MMA-*co*-20TCDDMDA) was greater than that of P(MMA-*co*-10TCDDMDA).

Differential Scanning Calorimetry

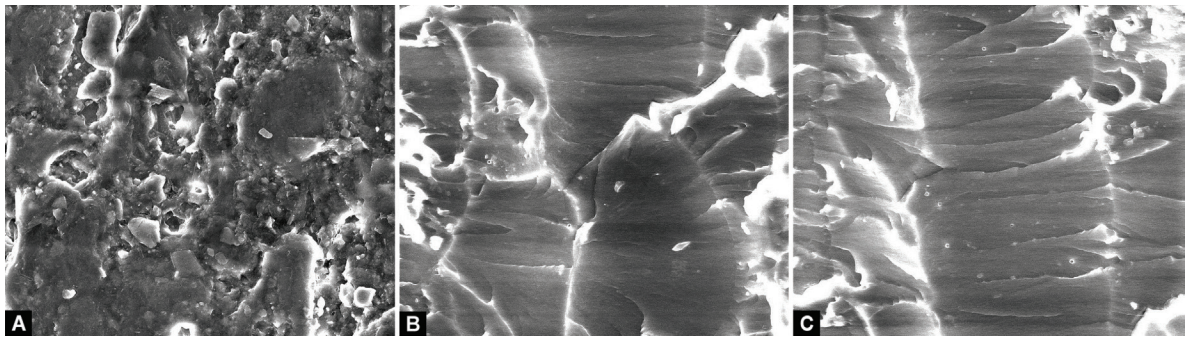
One-way ANOVA (Table 2) showed the level of significant differences among the tested groups ($p = 0.000$). When compared between



Figs 1A and B: ^{1}H-NMR spectrum: (A) control group; (B) experimental groups



Figs 2A and B: ^{13}C-NMR spectrum: (A) control group; (B) experimental groups

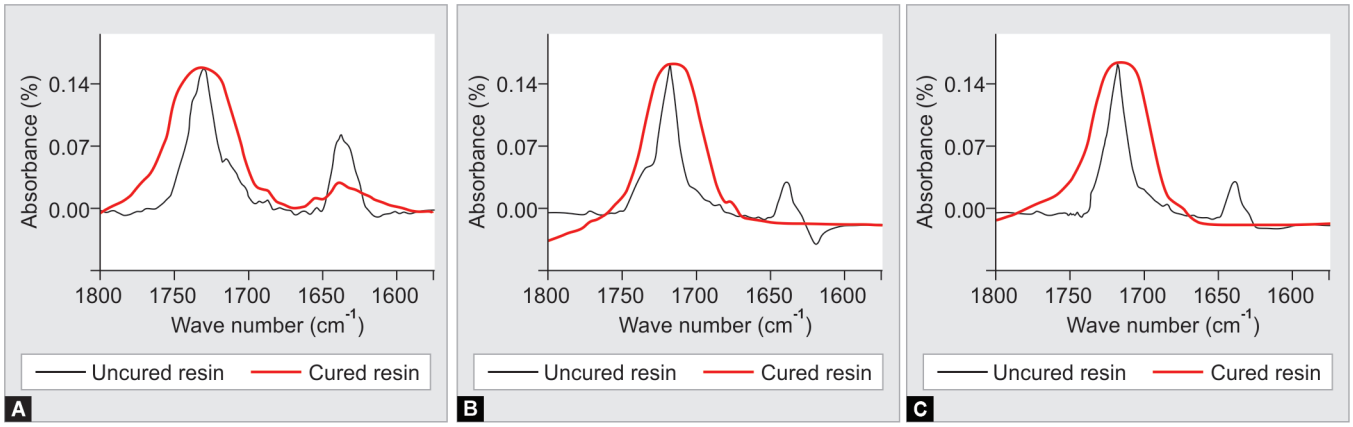


Figs 3A to C: Field-emission scanning electron microscopy: (A) P(MMA); (B) P(MMA-*co*-10TCDDMDA); (C) P(MMA-*co*-20TCDDMDA)

the groups by Bonferroni multiple comparison tests (Table 3), there existed a statistically significant difference ($p = 0.000$). The T_g of experimental groups was higher than that of the control group. The T_g of P(MMA-*co*-20TCDDMDA) was slightly higher than that of P(MMA-*co*-10TCDDMDA). Hence, the addition of TCDDMDA in MMA increased T_g. Figure 5 shows the DSC curves of the groups with the T_g plot. Exothermic post/cold crystallization peak appeared at around 160°C (T_c) in the experimental groups, which is absent in the P(MMA) group.

Discussion

The present research demonstrated the influence of the copolymerization, which was caused by adding the TCDDMDA monomer, on the DC and T_g of heat-cured PMMA material. The structure of the PMMA was chemically modified by incorporating TCDDMDA (10% and 20% v/v) to produce the P(MMA-*co*-TCDDMDA) copolymer. Nuclear magnetic resonance spectroscopy of the experimental groups showed higher protons and C-atoms than the control group. This was further affirmed by EDX spectroscopy, which showed a higher C weight percentage in experimental groups than the control. He et al.[19] used antibacterial agents in methacrylate dental composites to evaluate the antibacterial activity, and they characterized the structure of the materials using NMR. Ayaz et al.[20] copolymerized the acrylamide monomer with MMA. This was confirmed by the appearance of a new peak of the $-NH_2$ functional group in the experimental groups. Therefore, ^{1}H- and ^{13}C-NMR spectroscopy along with EDX spectroscopy confirmed the formation of the P(MMA-*co*-TCDDMDA) copolymer.



Figs 4A to C: Fourier transform infrared charts: (A) P(MMA); (B) P(MMA-*co*-10TCDDMDA); (C) P(MMA-*co*-20TCDDMDA)

Table 2: One-way ANOVA

Groups	*Mean (SD)*	*F value*	*p value*
I. DC (%)			
P(MMA)	78.56 (0.84)	305.577	0.000
P(MMA-*co*-10TCDDMDA)	83.77 (0.79)		
P(MMA-*co*-20TCDDMDA)	87.05 (0.67)		
II. DSC (°C)			
P(MMA)	107.69 (0.55)	2644.431	0.000
P(MMA-*co*-10TCDDMDA)	123.35 (0.55)		
P(MMA-*co*-20TCDDMDA)	125.33 (0.67)		

ANOVA, analysis of variance; SD, standard deviation; MMA, methyl methacrylate; TCDDMDA, tricyclodecane dimethanol diacrylate; DC, degree of conversion; DSC, differential scanning calorimetry

Table 3: *Post hoc* Bonferroni test

Group	*Compared group*	*Mean difference*	*Significance*
I. DC (%)			
P(MMA)	P(MMA-*co*-10TCDDMDA)	−5.21*	0.000
	P(MMA-*co*-20TCDDMDA)	−8.49*	0.000
P(MMA-*co*-10TCDDMDA)	P(MMA-*co*-20TCDDMDA)	−3.28*	0.000
II. DSC (°C)			
P(MMA)	P(MMA-*co*-10TCDDMDA)	−15.66*	0.000
	P(MMA-*co*-20TCDDMDA)	−17.64*	0.000
P(MMA-*co*-10TCDDMDA)	P(MMA-*co*-20TCDDMDA)	−1.98*	0.000

*The mean difference is significant at the 0.05 level
MMA, methyl methacrylate; TCDDMDA, tricyclodecane dimethanol diacrylate; DC, degree of conversion; DSC, differential scanning calorimetry

The experimental groups had greater DC than the P(MMA) group. This increase in DC is attributed to reactive moieties. The acrylic moieties of diacrylates (TCDDMDA) are generally more reactive than methacrylic moieties of the MMA/ethylene glycol dimethacrylate cross-linker (EGDMA) and are thus used when faster reaction kinetics is desired. In the EGDMA cross-linker, after the first methacrylate group has reacted, the second pendant methacrylate group is estimated to be less reactive by 7.5–10 times. This leads to reduced final conversion for copolymerization of MMA with EGDMA.[7] However, the faster reaction kinetics of acrylic moieties in TCDDMDA cause increased conversion thereby reducing the residual monomer content. In the FTIR spectra, the Abs peak at 1,638 cm^{-1} corresponds to the presence of unreacted residual monomer (C=C). In this study, the disappearance of Abs peak at 1,638 cm^{-1} for the P(MMA-*co*-TCDDMDA) copolymer suggests greater DC and lesser residual monomer than the P(MMA).

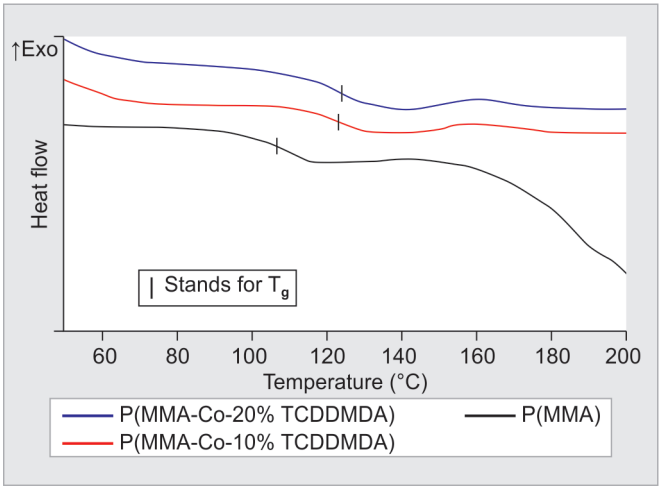


Fig. 5: Differential scanning calorimetry curves with T_g plot

The addition of TCDDMDA to MMA increased the T_g of the P(MMA-*co*-TCDDMDA) copolymer when compared to P(MMA). The exact relationship between the T_g and cross-linkers has not been established yet in the dental literature. From this study, it may be inferred that the cross-linker type determines the T_g. However, Jerolimov et al.[21] concluded that cross-linking PMMA by divinyl cross-linking agents can significantly alter T_g. However, the relationship between T_g and cross-linker chain length and cross-linker concentration is complex. Also, different curing cycles produced T_g variations up to 20°C.[22] Apart from the TCDDMDA comonomer, certain other monomers have been experimented with MMA and studied its influence on T_g.[15] For example, the addition of fluoro monomers in MMA increased the T_g.[23,24] Kubota et al.[24] concluded that increase in the C-chain length increased the T_g. However, itaconate[25,26] and nitro-monomers[14] decreased T_g. Hence, from the aforementioned studies, it is apparent that the T_g is not only influenced by the C-chain length but also by the type of comonomer added.

In this study, concentrations of 10% and 20% (v/v) of TCDDMDA were substituted in MMA. The copolymerizing capability of TCDDMDA with MMA, DC, and T_g more than 20% have not experimented yet. This is the only research with the cycloaliphatic monomer in PMMA, and therefore, the results should be interpreted with caution. Further studies ought to be executed by increasing the concentration of TCDDMDA and its effect on the mechanical properties and biocompatibility.

Conclusion

Within the limitations of this research, the following conclusions can be deduced:

- There was evidence of copolymerization of the TCDDMDA comonomer with PMMA resin.
- Unfinished surface topography appeared lamellar along with high C wt% in P(MMA-*co*-TCDDMDA) copolymers.
- The DC and T_g were increased after the incorporation of TCDDMDA in the PMMA resin matrix. The P(MMA-*co*-20TCDDMDA) copolymer had the highest conversion and T_g.

Clinical Significance

The study of copolymerization by adding a comonomer in denture resins provides a modification or reinforcement method, which is expected to improvise the mechanical properties and biocompatibility of the material. This will result in improved prosthetic (dentures) quality and durability, thereby, imparting a better quality of life to the geriatric population.

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