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Original Research

Assessment of cytotoxicity of heat cure denture base resin modified with recycled PMMA-based denture base resin

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ABSTRACT

Background: A biocompatible material is one which has the quality of being non-destructive in oral environment. The present study was conducted to assess invitro cytotoxicity of heat cure denture base resin modified with recycled PMMA-based denture base resin. **Materials & Methods:** 60 disk-shaped specimens which were prepared and divided into six groups of 10 each. The group I, II, III, IV and V consisted of denture base resin processed with substitution of 10% 20%, 30%, 40%, and 50% (w/w) of recycled denture base resin (R_{10}, R_{20}, R_{30}, R_{40}, and R_{50}). The control group (group VI) consisted of unmodified processed denture base resin. In each group, eluates were prepared using five sterile specimens. The mean optical density (OD) values and cell survival/viability % in all groups was compared. **Results:** The mean OD at 570 nm in group I was 0.32, in group II was 0.31, in group III was 0.33, in group IV was 0.31, in group V was 0.32 and in group VI was 0.31. The difference was non- significant (P> 0.05). The mean of cell viability % in group I was 92.3%, in group II was 92.5%, in group III was 92.4%, in group IV was 92.8%, in group V was 92.6% and in group VI was 92.1%. The difference was non- significant (P> 0.05). **Conclusion:** It was found that modification of denture base resin using recycled PMMA does not have a cytotoxic effect on the mouse fibroblast cell line L929.

Key words: cytotoxicity, cell viability, Denture base acrylic

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INTRODUCTION

A biocompatible material is one which has the quality of being non-destructive in oral environment. Almost all dental materials have the characteristic feature of releasing substances into the oral environment to a varying degree, where denture base resins are not an exception.[1] Denture base acrylic resins were introduced to dentistry in the late 1930s. Since then, there have been reports of reactions to prostheses fabricated out of these materials.[2] Descriptions of oral reactions to denture base acrylic resins often include symptoms like stomatodynia, glossodynia, rubor, and mucosal erosion. In free-radical polymerization, the monomer to polymer conversion is not complete and the unreacted residual monomer released from the denture base may cause irritation or allergic oral reactions when in contact with the oral mucosa.[3]

Today's commercially available heat cure denture base resin materials are supplied in a 2-component powder-liquid system. The powder consists mainly of pre-polymerized poly (methyl methacrylate) and liquid component consisting of methyl methacrylate in monomeric form as the major component.[4] Many studies studied polymerization process of the denture base resins and its influence on the physical and biocompatibility properties of the resultant denture base.[5] Despite overwhelming research in the area to optimize the water: powder ratio, polymerization method, and curing cycle, it has been established that the conversion of monomer to polymer is incomplete.[6]The present study was conducted to assess invitro cytotoxicity of heat cure denture base resin modified with recycled PMMA-based denture base resin.

MATERIALS & METHODS

The present invitro study comprised of 60 disk-shaped specimens which were prepared and divided into six

groups of 10 each. The group I, II, III, IV and V consisted of denture base resin processed with substitution of 10% 20%, 30%, 40%, and 50% (w/w) of recycled denture base resin (R_{10}, R_{20}, R_{30}, R_{40}, and R_{50}). The control group (group VI) consisted of unmodified processed denture base resin. In each group, eluates were prepared using five sterile specimens. The mouse fibroblast cell line (L929) was seeded in a 96-well cell plate system at a concentration of 1×10^4 cells/well in the DMEM medium with 1× antibiotic and antimycotic solution and 10% fetal bovine serum at 37°C with 5% CO_2 and incubated in a CO_2 incubator for 48 hours. MTT assay was applied and the absorbance was measured at 570 nm using a microplate reader to assess the in-vitro cytotoxicity. The mean optical density (OD) values and cell survival/viability % in all groups was compared. Data thus obtained were subjected to statistical analysis. P value < 0.05 was considered significant.

RESULTS

Table I: The mean OD at 570 nm

Groups	Mean	P value
Group I	0.32	0.94
Group II	0.31	
Group III	0.33	
Group IV	0.31	
Group V	0.32	
Group VI	0.31	

Table I shows that mean OD at 570 nm in group I was 0.32, in group II was 0.31, in group III was 0.33, in group IV was 0.31, in group V was 0.32 and in group VI was 0.31. The difference was non- significant (P> 0.05)

Table II: The mean of cell viability %

Groups	Mean	P value
Group I	92.3	0.97
Group II	92.5	
Group III	92.4	
Group IV	92.8	
Group V	92.6	
Group VI	92.1	

Table II, graph I shows that mean of cell viability % in group I was 92.3%, in group II was 92.5%, in group III was 92.4%, in group IV was 92.8%, in group V was 92.6% and in group VI was 92.1%. The difference was non- significant (P> 0.05).

DISCUSSION

Cytotoxic effects of denture base resins are attributed to polymerto-monomer ratio, storage time, water immersion, polymerization method, and cycle.[7] It must not be inferred that by following heatpolymerizing method, release of residual monomer is completely eradicated.[8]Denture base acrylic resins have elicited various degrees of in vitro cytotoxicity and in vivo allergic reactions which are probably caused by unreacted residual monomer present after the polymerization.[9,10]The present study was conducted to assess invitro cytotoxicity of heat cure denture base resin modified with recycled PMMA-based denture base resin.

We found that the mean OD at 570 nm in group I was 0.32, in group II was 0.31, in group III was 0.33, in group IV was 0.31, in group V was 0.32 and in group VI was 0.31. Ajay et al[11] chemically characterized heat-cure denture by adding TCDDMDA to MMA and observed the disappearance of carbon–carbon double-bond (C═C) peak which indicated a clear reduction in the residual monomer. The tricyclodecane (TCD) tri-ring central group of TCDDMDA offers steric hindrance effect that slows the rate of polymerization and facilitates the monomeric conversion to polymer, thereby reducing the residual unpolymerized monomer (C═C double bond) content in the final polymerized specimens. Also, TCD tri-ring monomers are classified under cross-linking monomers. Cross-linking monomers upon polymerization reduces the residual monomer content. Ethylene glycol dimethacrylate (EGDMA), a commercially used cross-linking monomer, does not possess the steric-hindrance property which very well unique in the TCD tri-ring monomers (TCDDMDA).

We found that mean of cell viability % in group I was 92.3%, in group II was 92.5%, in group III was 92.4%, in group IV was 92.8%, in group V was 92.6% and in group VI was 92.1%. Krishnamoorthi et al[12]evaluated the in-vitro cytotoxicity of heat-cure denture base resin (PMMA) modified with recycled denture base resin at 10%, 20%, 30%, 40%, and 50% (w/w) concentration.A total of 30 disk-shaped specimens were prepared and divided into six groups (n = 5). The Control group (R0) consisted of unmodified processed denture base resin, the experimental group consisted of denture base resin processed with substitution of 10% 20%, 30%, 40%, and 50% (w/w) of recycled denture base resin (R10, R20, R30, R40, and R50). Eluates were prepared using five sterile specimens of each group. The mouse fibroblast cell line (L929) was seeded in a 96-well cell plate system at a concentration of 1 × 104 cells/well in the DMEM medium with 1× antibiotic and antimycotic solution and 10% fetal bovine serum at 37°C with 5% CO2 and incubated in a CO2 incubator for 48 h. MTT assay was applied and the absorbance was measured at 570 nm using a microplate reader to assess the in-vitro cytotoxicity. No statistically significant difference was observed in the mean and standard deviation of the optical density and cell viability % of the test groups that were compared.

Jorge et al[13] compared heat-cure denture base resin against microwave-cured denture base resin and concluded that all compared materials exhibited cell viability of more than 80%. Although a long curing cycle was used to cure the heat-cure denture base resin samples, UV radiation may have affected the

degree of polymerization and as a result the amount of residual monomer present in the material. This finding is in accordance with the results of the present study. While comparing auto polymerizing and heat-cure denture base resin, it was a general observation that heat-cured resin was less toxic.Urban et al[14] concluded that the short curing cycle with terminal boil promoted lower amount of residual monomer (0.08%) when compared with the long curing cycle without terminal boil (0.24%).
The limitation the study is small sample size.

CONCLUSION

Authors found that modification of denture base resin using recycled PMMA does not have a cytotoxic effect on the mouse fibroblast cell line L929.

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