Regeneration, Reconstruction & Restoration

Original Article

Open Access

Effect of Glucose on Attachment and Proliferation of Human Gingival Fibroblasts on Zirconia and Titanium Surfaces

Sadaf Keshavarz[1] | Maryam Torshabi[2] | Amirhosein Zamanian[3*] | Zeinab Rezaei Esfahrood[4]

1. School of Dentistry, Shahid Beheshti University of Medical Sciences, Tehran, Iran.
2. Department of Dental Biomaterials, School of Dentistry, Shahid Beheshti University of Medical Sciences, Tehran, Iran.
3. Department of Prosthodontics, School of Dentistry, Shahid Beheshti University of Medical Sciences, Tehran, Iran.
4. Department of Periodontics, School of Dentistry, Shahid Beheshti University of Medical Sciences, Tehran, Iran.

Article Info

Received: 16 September, 2025
Accepted: 28 October, 2025
Published: 15 November, 2025

***Corresponding Author**
Amirhosein Zamanian, Department of Prosthodontics, Dental School, Shahid Beheshti University of Medical Sciences, Tehran, Iran.

Email: zamanian.amirhosein@gmail.com

Abstract

Background and objectives: Diabetes-induced delayed wound healing compromises dental implant success by impairing junctional epithelium formation. This study evaluated the effects of various glucose concentrations on human gingival fibroblast (HGF) proliferation and attachment to titanium and zirconia surfaces.

Materials and methods: Titanium and zirconia discs (2 × 10 mm) were prepared. HGFs were cultured on the discs or empty polystyrene plates (control) at glucose concentrations of 5.5, 11, 22, and 33 mM. Cell viability/proliferation and attachment were evaluated after 24 and 72 h using MTT assay and scanning electron microscopy (SEM). Statistical analysis was performed using one-way ANOVA and post-hoc Tukey test ($P < 0.05$).

Results: Glucose concentration did not significantly affect cell viability/attachment within each material group. However, HGFs proliferated significantly between 24 and 72 h at 5.5 and 11 mM glucose on titanium, and at all concentrations on zirconia. Cell viability was similar between titanium and zirconia at all concentrations except 11 mM, where zirconia performed better. At 72 h, zirconia showed significantly higher cell attachment than titanium at high glucose concentrations (22 and 33 mM).

Conclusion: Glucose did not affect initial cell attachment, but prolonged incubation (72 h) reduced cell-surface affinity on both metals compared to polystyrene. Nevertheless, zirconia promoted better HGF proliferation and attachment than titanium under high glucose conditions, suggesting zirconia as a preferable abutment material for diabetic patients.

Keywords: Glucose, Proliferation, Attachment, Human gingival fibroblast, Zirconia, Titanium.

Please cite this paper as:
Keshavarz S, Torshabi M, Zamanian AH, Rezaei Esfahrood Z. Effect of Glucose on Attachment and Proliferation of Human Gingival Fibroblasts on Zirconia and Titanium Surfaces. Journal of "Regeneration, Reconstruction & Restoration" (Triple R). 2025; Volume 10: e21. Doi: 10.22037/rrr.v10.50316

1. Introduction

Diabetes mellitus, as one of the most common health problems, has several complications, one of which is delayed wound healing (1,2). Deficient insulin production or an improper cell response to blood insulin causes an increase in blood glucose levels in diabetic patients. It has been shown that collagen biosynthesis impairment in intact skin (3), reduced fibroblast proliferation potential (4), connective tissue repair deficiency, superstructure changes in connective tissue contraction, delayed presence of insulin-like growth factor I and II during wound repair, and microvascular damage (5,6) occur due to high-glucose concentrations. Therefore, it seems that uncontrolled diabetes may adversely affect peri-implant soft tissue formation and maintenance.

Zirconia is a white crystalline oxide of zirconium. Its mechanical properties are similar to those of metals, and its color is similar to that of natural teeth. Impressive mechanical features, chemical stability, versatility, acceptable esthetic outcomes, and favorable biocompatibility have put zirconia-based restorations at the forefront of current interest in dental

clinics (7-10). Titanium is a relatively light metal with a high strength-to-weight ratio. The mechanical behavior of titanium is acceptable at high temperatures. The anodic titanium oxide layer forms on the titanium surface when it comes into contact with body fluids. The presence of this protective anodic titanium oxide layer results in remarkable corrosion resistance, even up to 353 ˚C (11,12). Compared with other alloys and materials used for implant production, titanium has great bone compatibility and, in addition to stimulating formation, allows for bone growth through the implant surface pores (11). Fibroblasts are the most common cells in connective tissue; they are differentiated from undifferentiated mesenchymal stem cells. These cells play an essential role in the production and maintenance of the extracellular matrix composition, including collagen fibers, the most abundant protein in the human body. The fibroblasts' function can be divided into two stages: 1. Cells are highly synthetic and detectable in morphology. 2. Inactivated cells are in their preformed matrix (13). Human gingival fibroblasts (HGFs) are predominant cells in healthy gingival connective tissue and are critical for the gingival system's evolution. These cells have roles in the formation, survival, and repair of the gingival connective tissue and its extracellular matrix components (e.g., various types of collagens, glycoproteins, and proteoglycans). They remodel the collagen fibers during connective tissue repair (13).

High-glucose culture media are commonly used in studies to simulate the diabetic biological environment and evaluate its complications (14,15). Considering the role of fibroblasts and their collagens in implants' cell-surface affinity, this study aimed to assess the morphology, viability, proliferation, and attachment of HGF to zirconia and titanium surfaces in high- and low-glucose culture media.

2. Materials and Methods

2.1. Cell culture

HGF1 PI 1 (NCBI: C165) cells were purchased from the National Cell Bank of Iran (Iran Pasteur Institute). Cells were cultured using 25ml Dulbecco's modified Eagle's medium (DMEM, GIBCO, UK) supplemented with 10% fetal bovine serum (GIBCO, UK) and 1% penicillin-streptomycin (GIBCO, UK). The cultures were incubated with 5% CO2 at 37 °C in a 95% humidified atmosphere.

2.2. Glucose cytotoxicity assay

The MTT (3-[4,5-dimethylthiazol-2-yl] -2,5-diphenyl tetrazolium bromide) assay was used to evaluate the effect of glucose on HGFs' viability and proliferation. 3500 cells with 100 µL culture medium were transferred to each well of a 96-well culture plate (SPL, Korea) and incubated for 24 h (with 5% CO2 at 37 °C in a 95% humidified atmosphere). On the second day of culture, 5.5, 11, 22, and 33 mM glucose were added to the wells (6 wells per concentration). Cultures containing 5.5 mM glucose are considered the negative control (no cytotoxicity, 100% viability). The plates were incubated for 24 and 72 h to evaluate acute and chronic cytotoxicity, respectively. At the end of each incubation period, culture plates were removed from the incubator; each well's medium was aspirated and substituted with 100 µL of fresh culture medium without FBS and antibiotics containing 10% MTT dye solution (5 mg/mL). After 3 h of incubation at 37 °C and 5% CO2, DSMO solvent (Sigma-Aldrich, Germany) was used to dissolve the insoluble, crystalline formazan. An ELISA microplate reader (Anthos, 2020, Salzburg, Austria) was used to measure the spectrophotometric absorbance at 570 nm wavelength and areference filter at 620 nm.

2.3. Preparation of titanium and zirconia specimens

A total of 96 experimental discs (48 zirconia (Y-TZP, Zr) and 48 titanium (TiAl6V4) discs) were prepared with final dimensions of 9 mm diameter and 2 mm height (after sintering and the subsequent shrinkage of the zirconia specimens). Zirconia cylinders (10 × 25 mm) were cut into disc-shaped specimens (10 × 2.5 mm) with the DORSA cutting machine and were sintered in a Vita Zircomat T furnace. Titanium discs were cut from a titanium sheet (1 m × 4 cm × 2mm) using laser cutting with a nitrogen generator. The zirconia and titanium discs were polished with a similar sequence of mullets and polishing felt (FGM diamond felt disc).

2.4. Surface roughness evaluation

To prevent movement of the discs during the measurement, theywere fixed in wax molds. A Contact surface profilometer (TR200, SaluTron GmbH) with a crosshead diameter of 5 µm and a speed of 0.25 mm/s was used to measure the Ra (roughness average) and Rz (average maximum height of profile) parameters in three different areas of each disc without overlapping.

2.5. Cell culture on discs

Following sterilization (drowning in 70% isopropyl alcohol, three washes with PBS, autoclaving in high-pressure steam at 120 ˚C for 20 min), discs were placed in 48-well culture plates. A total of 10×10^4 HGFs in 500 µL DMEM supplemented with 10% FBS, 1% penicillin-streptomycin, and various concentrations of glucose (5.5, 11, 22, 33 mM) were seeded on each disc (every five wells had a similar concentration). A total of 104 cells (passage number = 5) were seeded on each disc. Cultures with 5.5 mM glucose concentration are considered the control based on our previous studies (16,17). As standard cell seeding, 48-well disc-free polystyrene culture plates were seeded with 10×10^4 HGFs, 500 µL DMEM (FBS 10%, penicillin-streptomycin 1%), and glucose at a defined concentration. Standard and experimental culture plates were incubated at 37 ˚C with 95% humidity and 5% CO2 for 24 and 72 h.

2.6. Cell viability and proliferation assay

After 24 and 72 h, the cultures were removed from the incubators, and the media were fully aspirated. 500 µL of fresh DMEM (without FBS and antibiotics) containing 10% MTT was added to each well. Then the plates were incubated for 3 h at 37 ˚C and 5% CO2 until the formation of formazan crystals. Afterward, the medium was substituted with 200 µL DESMO solvent. 100 µL of the colored solution of each well was transferred to a 96-well culture plate. The spectrophotometric absorbance (optical density, OD) at 570 nm was read using an ELISA microplate reader (Anthos 2020, Salzburg, Austria). The reference filter was 620 nm.

2.7. Cell attachment and morphology assay

At the end of each incubation period, the samples' culture medium was aspirated, and the cells were washed twice with PBS. 2.5% glutaraldehyde (Sigma-Aldrich, Germany) was used for fixation. After 24 h, a graded series of ethanol (30%, 50%, 70%, 80%, 90%, and 100%) was used to dehydrate the cells (10 min for each solution). Followingethanol removal , the specimens were air-dried. The cell morphology was evaluated by scanning electron microscopy (SEM) (Arya Electron Optics Co., Tehran, Iran) with ×20, ×50, and ×100 magnifications. A sputtered gold coating was applied to the specimens before capturing images.

2.8. Statistical analysis

Statistical analyses were performed by the GraphPad Prism software (version 9) using a one-way analysis of variance (one-way ANOVA) followed by Tukey's post-hoc test for pairwise comparisons. A P-value < 0.05 was considered statistically significant.

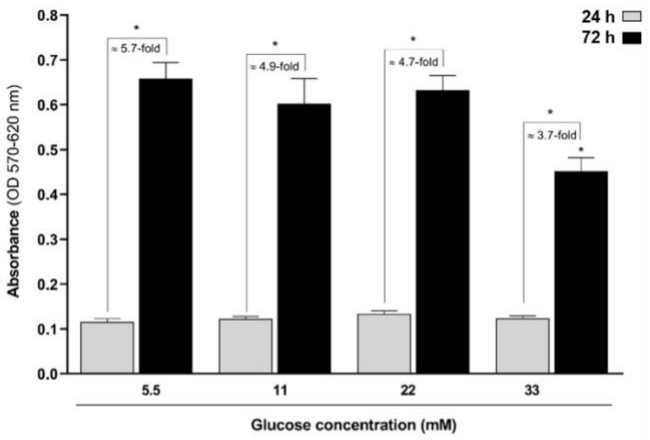
3. Results

3.1. Surface roughness

Ra and Rz parameters (Mean ± SEM) were 0.27 ± 0.06 and 1.54 ± 0.27 in titanium and 0.43 ± 0.07 and 2.21 ± 0.31 in zirconia specimens. There was no statistically significant difference between the specimens' surface roughness (P > 0.05)

3.2. Cell morphology and proliferation in standard cultures

After 24 h of incubation, there was no significant difference between cultures with various glucose concentrations in the number of viable cells, while after 72 h, the high-glucose medium (33 mM glucose) had significantly fewer viable cells than the control group (P < 0.05). Cells significantly proliferated after 72 h of incubation in all mediums (P < 0.05); however, the proliferation rate was significantly lower in a high-glucose medium (33 mM glucose) than in other concentrations (Figure 1). The morphology of the cells after 24 and 72 h of incubation was similar to that of the control groups, indicating no cellular toxicity at any of the glucose concentrations tested (Figure 2).



	HGF Cell Attachment/Proliferation *(Mean±SEM)* on **Polystyrene surface** *in the presence of* **Glucose**			
	5.5 mM	11 mM	22 mM	33 mM
24 h	0.12±0.01	0.12±0.01	0.13±0.01	0.12±0.01
72 h	0.66±0.04	0.60±0.06	0.63±0.03	0.45±0.03

Figure 1. HGFs' attachment and proliferation on standard cultures (polystyrene surface) after exposure to 5.5, 11, 22, and 33 mM glucose concentration, incubated for 24 h (white columns) and 72 h (black columns). The rate of proliferation is calculated and compared between two time points.

3.3. Cell morphology, attachment, and proliferation on titanium surfaces

Regarding viable attached cells, there was no significant difference among specimens with various glucose concentrations incubated for either 24 or 72 h. From 24 to 72 h, the cells significantly proliferated in the control group (5.5 mM glucose) and specimens with 11 mM glucose (1.7 and 1.9 folds, respectively) (Figure 3).

3.4. Cell morphology, attachment, and proliferation on zirconia surfaces

The difference in the attached cell numbers was not statistically significant between the experimental and control groups (P > 0.05). Cells treated with all glucose concentrations significantly proliferated from 24 to 72 h of incubation (P < 0.05) (Figure 4).

3.5. Cell morphology, attachment, and proliferation in Standard, zirconia, and titanium cultures

Cell viability and attachment were not significantly different between polystyrene, titanium and zirconia surfaces after 24 h when the glucose concentrations were 5.5, 22, and 33 mM.

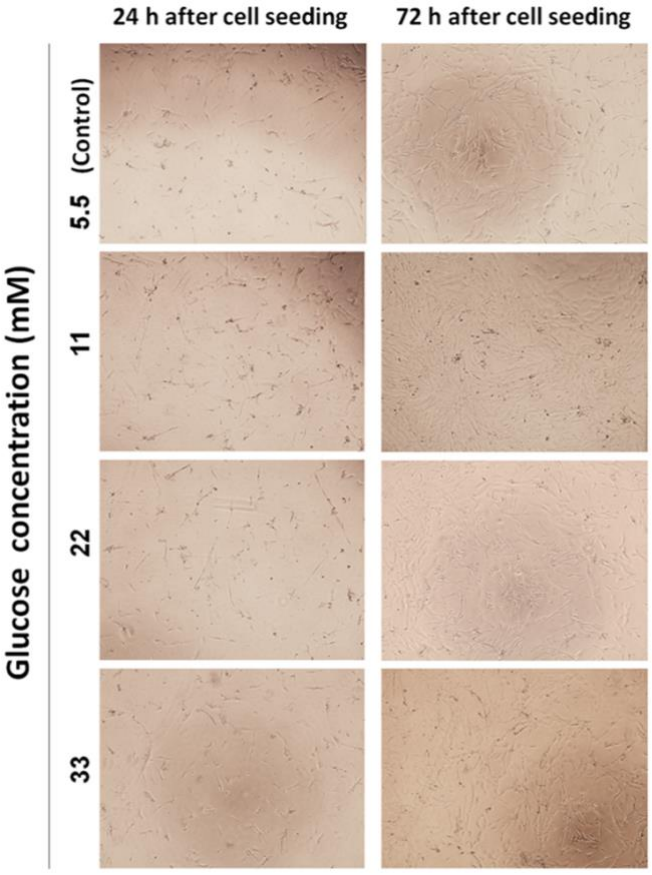
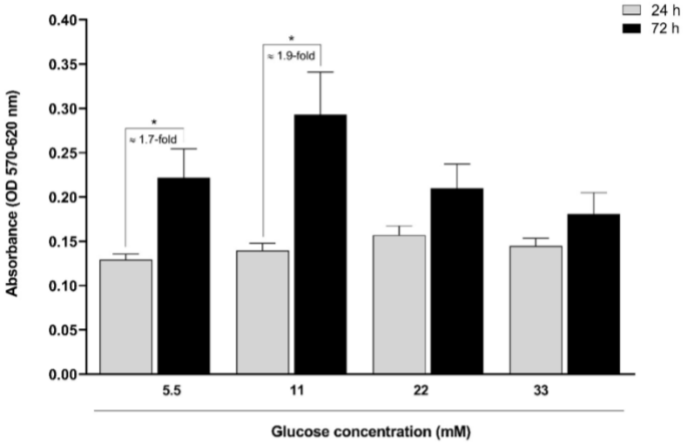
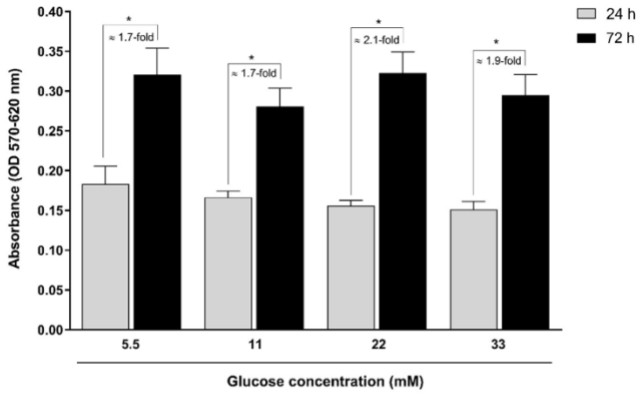


Figure 2. Inverted microscope images (10x magnification) of attached HGF cells on standard cultures, 24 and 72 h after treatment with various concentrations of glucose.



	HGF Cell Attachment/Proliferation *(Mean±SEM)* on **Titanium disk** *in the presence of* **Glucose**			
	5.5 mM	**11 mM**	**22 mM**	**33 mM**
24 h	0.13±0.01	0.13±0.00	0.16±0.01	0.14±0.01
72 h	0.22±0.03	0.25±0.01	0.21±0.03	0.18±0.02

Figure 3. HGFs' attachment and proliferation on titanium surfaces after exposure to 5.5, 11, 22, and 33 mM glucose concentration, incubated for 24 h(white columns) and 72 h (black columns). The rate of proliferation is calculated and compared between two time points.



	HGF Cell Attachment/Proliferation *(Mean±SEM)* on **Zirconia disk** *in the presence of* **Glucose**			
	5.5 mM	**11 mM**	**22 mM**	**33 mM**
24 h	0.18±0.01	0.17±0.00	0.16±0.01	0.15±0.01
72 h	0.32±0.03	0.28±0.02	0.32±0.03	0.30±0.03

Figure 4. HGFs' attachment and proliferation on zirconia surfaces after exposure to 5.5, 11, 22, and 33 mM glucose concentration, incubated for 24 h (white columns) and 72 h (black columns). The rate of proliferation is calculated and compared between two time points.

However, cells treated with 11 mM glucose had significantly better proliferation and attachment on zirconia surfaces compared to titanium and polystyrene ($P < 0.05$) (Figure 5). After 72 h, at all glucose concentrations, cells had significantly higher proliferation and attachment to polystyrene surfaces than titanium and zirconia ($P < 0.05$). The difference between zirconia and titanium was not statistically significant in the presence of 5.5 and 11 mM glucose, wherease at higher glucose concentrations (22 and 33 mM glucose), cells had significantly more proliferation and attachment to zirconia discs ($P < 0.05$). SEM observations showed cell proliferation from 24 to 72 h of incubation on both titanium and zirconia surfaces. High-glucose medium (33 mM glucose) had significantly lower cell proliferation than control specimens. In addition, in high-glucose media, zirconia surfaces had better cell-surface affinity and proliferation than titanium (Figure 6).

4. Discussion

Delayed wound healing (1,2), deficiency in collagen synthesis (3), impairment of fibroblast proliferation, and connective tissue repair (4) are complications caused by diabetes that may adversely affect gingival attachment to the abutment surface. The present *in vitro* study aimed to evaluate differences in HGFs' viability, proliferation, and attachment to titanium, zirconia, and polystyrene surfaces treated in a medium with various glucose concentrations (normal, 2, 4, and 6 times higher than normal concentrations).

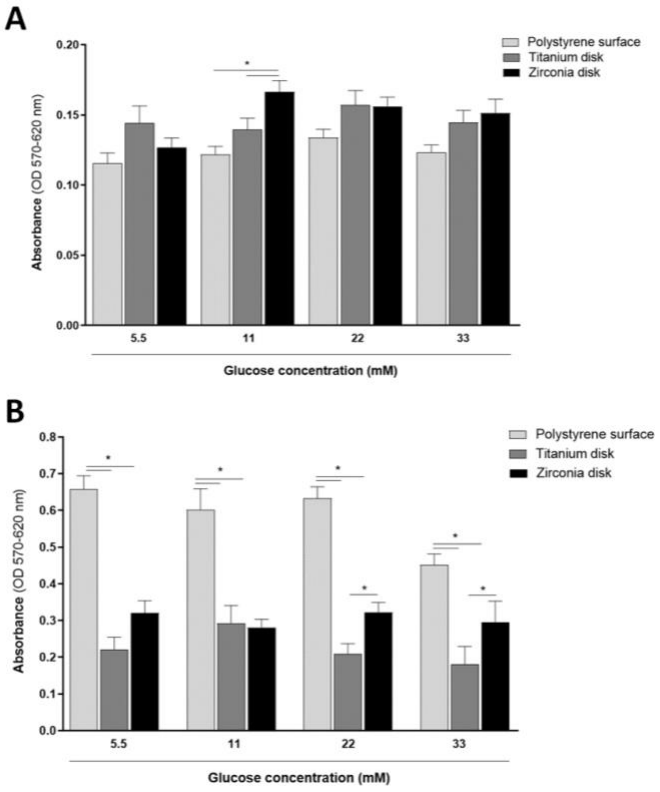


Figure 5. Comparison of Cell adhesion and proliferation on polystyrene, zirconia, and titanium surfaces after 24 (upper) and 72 (lower) hours of evaluation in various glucose concentrations (*$P < 0.05$).

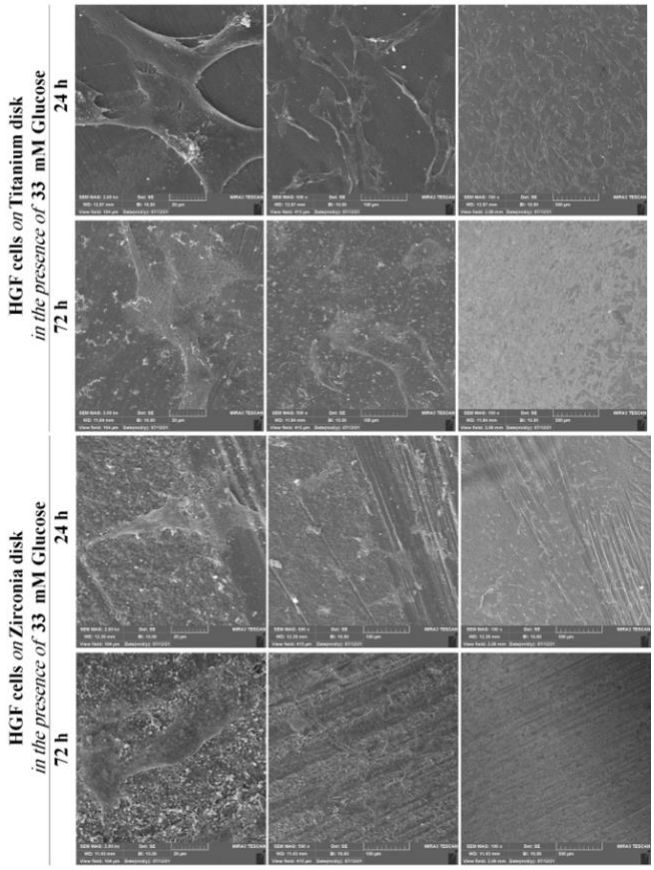


Figure 6. SEM images (20, 100, and 500 µm) of attached HGF cells on titanium (upper) and zirconia (lower) surfaces 24 and 72 h after exposure to 33 mM concentration of glucose.

In the current study, the surface roughness of zirconia and titanium discs was not significantly different. According to our findings in normal glucose concentration, zirconia and titanium surfaces similarly induced cell proliferation and had the same levels of cell-surface affinity. This was in line with the study by Esfahanizadeh et al. (18), where HGFs had similar patterns of gene expression, proliferation, and surface adhesion on zirconia and titanium discs. However, in contrast to our study, they found significantly higher surface roughness on zirconia than on titanium discs (0.40 ± 0.05 VS, 0.20 ± 0.08 µm). The proliferation and attachment of HGFs on machined, polished, and sandblasted zirconia and titanium discs after 24 and 72 h of incubation were evaluated by Nothdurft et al. (19). The mean Ra values in polished, machined, and sandblasted titanium alloys were 10.5 ± 1.5 nm, 69 ± 13.7 nm, and 1.514 ± 0.045 µm. Zirconia Ra values were 198.8 nm ± 15.7 nm, and 1.021 µm ± 0.043 µm in machined and sandblasted surfaces. The Rz values in machined and sandblasted titanium alloy were 428.7 nm ± 98.7 nm, and 11.436 ± 0.730 µm. Whereas the Rz values in zirconia surfaces were 1.570 µm ± 0.200 µm, and 6.339 µm ± 0.507 µm, respectively. As can be seen from these values, the machining process caused more roughness on zirconia surfaces than on titanium surfaces, whereas sandblasting caused more roughness on titanium alloy surfaces. In evaluating the effect of surface roughness on proliferation, the zirconia disc with a rough surface and the titanium disc with a machined surface were the two materials with the highest proliferation rates. When comparing materials with similar surface roughness, zirconia surfaces were found to have more attached vital cells, which is contrary to what we found; although zirconia had higher cell affinity in high-glucose medium, the difference was not statistically significant in normal glucose medium. According to Kim et al. (20), zirconia and titanium surfaces had a comparable proliferation rate after 72 h of incubation, yet after seven days of incubation, zirconia exhibited significantly higher proliferation. The anodic oxidation of zirconia surfaces might have enhanced cell proliferation. SEM evaluations conducted by Pandoleon et al. (21) showed variations in cells' morphology on titanium and zirconia surfaces. On titanium surfaces, most cells had a flattened phenotype, whereas on zirconia surfaces, most cells were elongated. Both morphologies indicate favorable cell-surface attachment. They stated that while there was no difference in HGF viability when in contact with titanium and non-aged zirconia, aging zirconia significantly decreased cells viability. Conversely, in another *in vitro* study, MTT and SEM evaluations from day 1 to 21 showed that cell growth and proliferation in zirconia groups were lower than those in

titanium groups, and that surface roughness did not affect the cells' biological behaviors. Cellular responses may have been influenced by the low surface roughness of titanium specimens, as well as other factors affecting the interaction between cells and dental materials (22).

Results from evaluating the effect of elevated glucose levels showed that, rather than 24 h of incubation, a glucose concentration of 33 mM significantly reduced cell proliferation in all cultures after 72 h. This negative effect of high-glucose concentration on cell proliferation was also reported by Liu et al. (23). Based on their evaluations, in contrast to low-glucose mediums (with 5.5 and 15 mM glucose), high-glucose concentrations (35 and 45 mM) significantly reduced the periodontal ligament fibroblast proliferation, total protein synthesis, alkaline phosphatase (ALP) activity, and collagen I and osteocalcin (OCN) secretion in a dose-dependent manner after 24 h of incubation. When the standard (polystyrene) and experimental (zirconia and titanium) surfaces were compared at increased glucose levels, significantly higher initial cell attachment (at 24 h of incubation) was found in zirconia specimens at an 11 mM glucose concentration. While the difference between zirconia, titanium, and polystyrene surfaces was not different in other concentrations. In contrast, after 72 h of incubation, the polystyrene surface showed a better proliferation rate and cell attachment than the experimental groups at all concentrations. In addition, zirconia discs outranked titanium once in high-glucose mediums (22 and 33 mM). The extracellular matrix proteins consist of hydrophobic and hydrophilic domains. They exhibit different surface interactions and conformations based on the chemistry of the surface material. Cell-surface adhesion is dependent on the presence of adsorbed proteins. In addition, cell adhesion molecules (CAMs) play an essential role in mediating this process. Differences in the mechanical and chemical characteristics of zirconia, titanium, and polystyrene (e.g., surface topography and microstructures, wettability, reactive surfaces, and impurities) might have caused differences in the nature and organization of extracellular matrix proteins and molecules, thus resulting in cell attachment level that variy (24).

5. Limitations

Several limitations of this study must be acknowledged. First, as an *in-vitro* investigation, it cannot fully replicate the complex *in-vivo* oral environment, which involves dynamic factors such as saliva, mechanical masticatory forces, the oral microbiome, and host immune responses. Second, the evaluation of cell attachment and proliferation was limited to short-term observation periods, leaving long-term cellular behavior and survival unexplored. Third, the experiments utilized static glucose concentrations, which do not reflect the continuous glycemic fluctuations experienced by diabetic patients. Finally, this study focused solely on human gingival fibroblasts (HGFs), whereas peri-implant wound healing and junctional epithelium formation are multicellular processes involving epithelial and immune cells. Therefore, future in vivo studies and clinical trials are necessary to validate these findings.

6. Conclusion

In light of the current study's findings, increasing the concentration of glucose did not affect HGF viability/proliferation or initial cell attachment/adhesion to zirconia or titanium surfaces. However, long-term culture of cells in high-glucose media (22 and 33 mM) significantly reduced cell-surface affinity on titanium discs. Nevertheless, it seems that zirconia and titanium abutments are not superior to each other in healthy patients or those with mild diabetes, while in severely diabetic patients, zirconia abutments are the better choice. However, due to the small number of specimens in terms of surface roughness, the short periods of assessment, the absence of the oral environment, and confounding factors such as saliva, thermal changes, immune system response, and zirconia aging, additional *in-vitro* and *in-vivo* studies are required to confirm these findings.

Acknowledgements
None.

Ethics
This study was approved by the ethics committee of School of Dentistry, Shahid Beheshti University of Medical Sciences, Tehran, Iran (IR.SBMU.DRC.REC.1400.080).

Using artificial intelligence (AI)
It is declared by the authors of this manuscript that no generative artificial intelligence (AI) or AI-assisted technologies were used to generate content, ideas, or theories during the writing process of this work.

Author contribution
Sadaf Keshavarz: Investigation, Writing - Original draft
Maryam Torshabi: Conceptualization, Methodology, Data curation, Validation, Resources, Writing - Review & editing
Amirhossein Zamanian: Conceptualization, Supervision, Writing - Review & editing
Zeinab Rezaei Esfahrood: Conceptualization, Writing - Review & editing

Conflict of interest
The authors declare no conflict of interest.

Funding
This research was founded by the School of Dentistry of Shahid Beheshti University of Medical Sciences (Grant number: 29499)

References
1. Pavlovic MD, Milenkovic T, Dinic M, Misovic M, Dakovic D, Todorović S, et al. The prevalence of cutaneous manifestations in young patients with type 1 diabetes. Diabetes care. 2007;30(8):1964-7.
2. Greenhalgh DG. Wound healing and diabetes mellitus. Clinics in plastic surgery. 2003;30(1):37-45.
3. Kern P, Moczar M, Robert L. Biosynthesis of skin

collagens in normal and diabetic mice. Biochemical Journal. 1979;182(2):337-45.
4. Franzén LE, Roberg K. Impaired connective tissue repair in streptozotocin-induced diabetes shows ultrastructural signs of impaired contraction. Journal of Surgical Research. 1995;58(4):407-14.
5. Brown DL, Kane CD, Chernausek SD, Greenhalgh DG. Differential expression and localization of insulin-like growth factors I and II in cutaneous wounds of diabetic and nondiabetic mice. The American journal of pathology. 1997;151(3):715.
6. Loots MA, Lamme EN, Mekkes JR, Bos JD, Middelkoop E. Cultured fibroblasts from chronic diabetic wounds on the lower extremity (non-insulin-dependent diabetes mellitus) show disturbed proliferation. Archives of dermatological research. 1999;291:93-9.
7. Ardlin BI. Transformation-toughened zirconia for dental inlays, crowns and bridges: chemical stability and effect of low-temperature aging on flexural strength and surface structure. Dental Materials. 2002;18(8):590-5.
8. Christel P, Meunier A, Heller M, Torre J, Peille C. Mechanical properties and short-term in vivo evaluation of yttrium-oxide-partially-stabilized zirconia. Journal of biomedical materials research. 1989;23(1):45-61.
9. Denry I, Kelly JR. State of the art of zirconia for dental applications. Dental materials. 2008;24(3):299-307.
10. Aldafeeri HR, Al-Zordk W, Ghazy MH. Marginal Accuracy of Machinable Monolithic Zirconia Laminate Veneers. Journal of Dental and Medical Sciences. 2019.
11. Park JB, Bronzino J. The biomedical engineering handbook. Boca Raton, FL: CRC Press. 2000;4:1-8.
12. Ishizawa H, Ogino M. Formation and characterization of anodic titanium oxide films containing Ca and P. Journal of biomedical materials research. 1995;29(1):65-72.
13. Cate AT, Deporter D. The degradative role of the fibroblast in the remodelling and turnover of collagen in soft connective tissue. The Anatomical Record. 1975;182(1):1-13.
14. Deveci M, Gilmont RR, Dunham W, Mudge B, Smith D, Marcelo CL. Glutathione enhances fibroblast collagen contraction and protects keratinocytes from apoptosis in hyperglycaemic culture. British Journal of Dermatology. 2005;152(2):217-24.
15. Benazzoug Y, Borchiellini C, Labat-Robert J, Robert L, Kern P. Effect of high-glucose concentrations on the expression of collagens and fibronectin by fibroblasts in culture. Experimental gerontology. 1998;33(5):445-55.
16. Torshabi M, Rezaei Esfahrood Z, Jamshidi M, Mansuri Torshizi A, Sotoudeh S. Efficacy of vitamins E and C for reversing the cytotoxic effects of nicotine and cotinine. European Journal of Oral Sciences. 2017 Dec;125(6):426-37.
17. Torshabi M, Esfahrood ZR, Gholamin P, Karami E. Effects of nicotine in the presence and absence of vitamin E on morphology, viability and osteogenic gene expression in MG-63 osteoblast-like cells. Journal of basic and clinical physiology and pharmacology. 2016 Nov 1;27(6):595-602.
18. Esfahanizadeh N, Motalebi S, Daneshparvar N, Akhoundi N, Bonakdar S. Morphology, proliferation, and gene expression of gingival fibroblasts on Laser-Lok, titanium, and zirconia surfaces. Lasers in medical science. 2016;31(5):863-73.
19. Nothdurft FP, Fontana D, Ruppenthal S, May A, Aktas C, Mehraein Y, et al. Differential behavior of fibroblasts and epithelial cells on structured implant abutment materials: A comparison of materials and surface topographies. Clinical implant dentistry and related research. 2015;17(6):1237-49.
20. Kim Y-S, Ko Y, Kye S-B, Yang S-M. Human gingival fibroblast (HGF-1) attachment and proliferation on several abutment materials with various colors. Int J Oral Maxillofac Implants. 2014;29(4):969-75.
21. Pandoleon P, Bakopoulou A, Papadopoulou L, Koidis P. Evaluation of the biological behaviour of various dental implant abutment materials on attachment and viability of human gingival fibroblasts. Dental Materials. 2019;35(7):1053-63.
22. Jung S, Moser MM, Kleinheinz J, Happe A. Biocompatibility of lithium disilicate and zirconium oxide ceramics with different surface topographies for dental implant abutments. International Journal of Molecular Sciences. 2021;22(14):7700.
23. Jia-qiang L, Hong-chen L, Yi W, Yuan F, Hui G. The biological effect of high glucose on human periodontal ligament fibroblast. Shanghai Journal of Stomatology. 2011;20(3).
24. Lavenus S, Pilet P, Guicheux J, Weiss P, Louarn G, Layrolle P. Behaviour of mesenchymal stem cells, fibroblasts and osteoblasts on smooth surfaces. Acta biomaterialia. 2011;7(4):1525-34.x]