Enhancement of fluoride release in glass ionomer cements modified with titanium dioxide nanoparticles

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Abstract
Background: Several efforts have been made to improve the glass ionomer cements (GICs) properties with nanotechnology. Fluoride release in once of most beneficial properties of GICs. The purpose of this study was to evaluate the fluoride release, recharge, and cytotoxicity in GICs reinforced with titanium dioxide nanoparticles (TiO_N).

Objective: Evaluate the fluoride release, recharge, and cytotoxicity in GICs reinforced with TiO_N.

Methods: Four GICs, FUJI IX EXTRA (G1c), KETAC MOLAR (G2c), IONOFILL MOLAR (G3c), and FUJI IX (G4c) were combined with TiO_N (G1e, G2e, G3e, and G4e) and divided into blocks of 5-mm width and 1-mm thickness 10 each. A total of 80 samples were arranged as follows: GICs alone as negative control (n = 40) and GICs + TiO_N as experimental groups (n = 40). The fluoride release was determined for periods of 1, 2, 6, 10, 31, 90, 180, 240, and 300 days. On days 30 and 179, samples were recharged by submerging in 1 mL of 20,000 ppm sodium fluoride gel. Cytotoxic activity was carried out with gingival fibroblasts, using 3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyltetrazolium bromide cell viability assay.

Results: The experimental groups obtained the highest and more constant fluoride released when compared to control groups. After the first recharge, experimental groups (G1e, G3e, and G4e) showed statistically significant results (P = .001, 0.010, and 0.001 respectively) enhancing their recharge ability regarding control groups. The second recharge showed better results in G1e concerning the rest of the groups. No cytotoxic activity was observed in all experimental groups, although significant differences were observed in G3e and G4e regarding control group.

Conclusion: The incorporation of TiO_N enhance the fluoride release in glass ionomers with a noncytotoxic effect on human gingival fibroblasts.

Abbreviations: GICs = glass ionomer cements, G1c = Glass ionomer FUJI IX EXTRA, G2c = Glass ionomer KETAC MOLAR, G3c = Glass ionomer IONOFILL MOLAR, G4c = Glass ionomer IONOFILL MOLAR, G1e = Glass ionomer FUJI IX EXTRA modified with titanium dioxide nanoparticles, G2e = Glass ionomer KETAC MOLAR modified with titanium dioxide nanoparticles, G3e = Glass ionomer IONOFILL MOLAR modified with titanium dioxide nanoparticles, G4e = Glass ionomer IONOFILL MOLAR modified with titanium dioxide nanoparticles, HGF = human gingival fibroblasts, PDL = population doubled level, TiO_2 = titanium dioxide, TISAB = total ionic strength adjustment buffer solution, TiO_N = titanium dioxide nanoparticles.

Keywords: Fluoride, glass ionomer cements, nanoparticles, nanotechnology

1. Introduction

In recent years, the concept of minimal intervention has been promoted in the clinical management of dental caries, the principle of this therapy is the remineralization for non-cavitated carious lesions.[1,2] This method has proven to be an economical and effective alternative to control the development of caries, to achieve less discomfort and anxiety to patients than the conventional rotary instruments. When surgical interference is required, only the part of the tooth surface which was irreversibly broken down will be replaced, therefore, demineralized tooth structure adjacent to the removed tissue should not be removed as it can be remineralized and healed.[3] Glass ionomer cements (GICs) have become the most used material for the therapy of minimal intervention, but it is properly cited. The work cannot be used commercially without permission from the journal.

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they can release fluoride which is taken up by the tooth structure. Fluoride is considered the gold standard of the different remineralization protocols and plays an important role in caries prevention, stabilizing the mineral apatite, and favoring the remineralization of enamel, which increases acid resistance of the dental surface and prevents caries. Fluoride has been added to different materials with the objective of reduce the prevalence of caries, which according to the World Health Organization affects 60% to 90% of school children and approximately 36% of global population suffers from this disease in permanent teeth.

Another advantage of GICs is their ability to be recharged, which contributes to the long-term inhibitory effect on enamel demineralization because these materials can capture available fluoride ions around the environment. This capability is determined by several factors involved in the process, such as concentration, application frequency of fluoride agent, and permeability of the material. The recharge capacity is very important when GICs have been used as a restorative materials. A constant fluoride release in restorative materials is focused on replacing the concentration, application frequency of fluoride agent, and permeability of the material. Another advantage of GICs is their ability to be recharged, which contributes to the long-term inhibitory effect on enamel demineralization because these materials can capture available fluoride ions around the environment. This capability is determined by several factors involved in the process, such as concentration, application frequency of fluoride agent, and permeability of the material. The recharge capacity is very important when GICs have been used as a restorative materials. A constant fluoride release in restorative materials is focused on replacing the nanoparticles of different materials as silver, magnesium oxide, hydroxyapatite etc. have been added to GICs for enhancing their antibacterial and/or mechanical properties without taking into consideration its crucial role of fluoride releasing, considered one of the most important properties of GICs. Titanium dioxide (TiO2), is an inorganic additive, that has many promising properties because it is chemically stable and biocompatible; it has been proposed as reinforcing fillers, and recently was reported that the incorporation of titanium dioxide nanoparticles (TiO2N) to GICs at 3%, significantly enhanced the hardness and showed antibacterial activity, without showed fluoride release and recharge capability of this materials. This study aimed to evaluate the fluoride release, recharge, and cytotoxicity in glass ionomer cements reinforced with TiO2N.

2. Materials and Methods

An in vitro study was performed to determine the effects of TiO2N on fluoride release from glass ionomer cement. All protocols were approved by the bioethics committee of the Center for Research and Advanced Studies in Dentistry. This study has followed the Checklist for Reporting in vitro Studies guidelines for in vitro studies as discussed in the 2014 concept note.

2.1. Sample preparation

Four glass ionomer cements were used for the control groups of 10 samples each. G1c: Fuji IX EXTRA (GC, Kyoto, Japan), G2c: KETAC MOLAR (3M ESPE, MN), G3c: IONOFILL MOLAR (Voco, Cuxhaven, Germany), and G4c: Fuji IX (GC, Kyoto, Japan) and for the experimental group TiO2 NPs were added to the same cements (G1e, G2e, G3e, and G4e).

TiO2 N were weighted by using an analytical balance (Hanna Instruments, Ann Arbor, MI) and adjusted to a final concentration of 3%, which were incorporated into the powder component of the cements before mixing according to the manufacturer’s instructions.

The samples consisted of blocks of 5-mm width and 1-mm thickness, that were placed in cavities with similar measures in a Teflon matrix. A person independent of the experiment selected the samples with the necessary size and thickness using a dental caliper. The presence of TiO2N was corroborated through an energy dispersive spectroscopy analysis, using a scanning electron microscope.

2.2. Fluoride release

Samples were placed in plastic containers, which had tight-fitting lids to prevent evaporation of the solution. Subsequently, these were stored in deionized water (5 mL) at a temperature of 37°C for 1, 2, 6, 10, 31, 90, 180, 240, and 300 days for all materials.

The potentiometer was calibrated by using standard solutions of sodium fluoride with 1, 10, 100, and 1000 ppm, and the Total Ionic Strength Adjustment Buffer solution (Hanna Instruments) was added to obtain a constant ionic strength background. The fluoride release was determined by using a fluoride ion-selective electrode (model 1011; Hanna Instruments). Results were expressed in ppm.

2.3. Fluoride recharge

On days 30 and 179, samples were recharged by immersing in 1 mL of 2% (20,000 ppm) sodium fluoride gel (Ionite Borgatta, Mexico) for 4 minutes, and subsequently rinsed with 2 mL of deionized water. The fluoride released was determined 24 hours after the recharge as previously described on days 31 and 180.

Figure 1. (A) Represents an image of SEM with TiO2 nanoparticles dispersed in the crystals of the glass ionomer. (B) EDS image representing the presence of % TiO2. EDS = energy dispersive X-ray spectroscopy, SEM = scanning electron microscope.
2.4. Cell culture
Oral cells, human gingival fibroblasts (HGFs), were obtained from patients between 15 and 18 years old through biopsy of gingival tissue after extraction of a dental organ indicated for orthodontic reasons, with the previous signature of the informed consent of the patient’s parent or guardian and the authorization of the bioethics committee (CE_16/004_SN). The obtained tissue was suspended in Dulbecco’s Modified Eagle Medium (Gibco, Carlsbad, CA) supplemented with 20% heat-inactivated fetal bovine serum (Gibco), supplemented with 100 IU/mL of penicillin G, and 100 μg/mL of streptomycin sulfate (Gibco). The tissue was sectioned in small portions using a #15 scalpel blade and placed in 100 millimeters culture dishes and incubated for its exponential growth at 37°C with 5% CO2 and 95% air for 2 weeks.

Cell growth was used as a primary culture with a population doubling level (PD) zero. Cell subcultures were maintained at a concentration of 1:4 and the culture medium was replaced every week. HGF has an in vitro lifetime (cumulative number of PD) of 47 PD, regardless of the culture medium used. The cells were detached from the bottom of the culture dish with 0.25% trypsin and 0.025% Ethylene Diamine Tetraacetic Acid Disodium Salt (EDTA-2Na) in phosphate buffered saline free from calcium and magnesium ions.[29,30]

2.5. Evaluation of indirect cytotoxicity activity
HGF cells (2 × 10^4 cells/mL) were inoculated into 24-well culture dishes and incubated for 48 hours to achieve complete adhesion. The culture medium was replaced by fresh culture for 30 minutes at 37°C in 5% CO2 to stabilize the pH and temperature of the culture medium. The GICs samples were immersed in DMEM supplemented with 10% of fetal bovine serum. The samples were incubated at 37°C with continuous agitation at 250 revolutions per minute for 24 hours. The medium was inoculated at 100% and incubated for 24 hours. The negative control consisted of a well with a fresh culture medium without cells. The number of viable cells with metabolic activity were determined using MTT method (3-[4,5-dimethylthiazol-2-yl]-2,5-di-phenyltetrazolium bromide), Sigma-Aldrich, St. Louis, MO). In brief, the culture medium was replaced with 0.2 mg/mL of MTT reagent, (Sigma-Aldrich) and incubated for 7 hours at 37°C. After removing the culture medium, the formazan was dissolved with dimethyl sulfoxide (Karal, Guanajuato, Mexico) and the absorbance was analyzed at 570 nanometers wavelength, to determine the mitochondrial activity, using a micro plate reader (Multiskant go, Thermoscientific, Finland).[29,30]

2.6. Statistical analysis
The data were analyzed for statistical differences using Student’s t test, to compare the fluoride release between the control and experimental groups (P ≤ .05) after 24 hours and 240 days and the recharge ability of experimental groups regarding control groups. Kruskal–Wallis test and Mann–Whitney U test were used for evaluating the cytotoxic activity in experimental groups (P ≤ .05), using Statistical Package for the Social Sciences Statistics program (v.21, IBM).

### Table 1
Descriptive analysis of fluoride release in experimental groups.

| Groups | 1 d (F-) | 2 d (F-) | 6 d (F-) | 10 d (F-) | 31 d (F-) | 90 d (F-) | 180 d (F-) | 240 d (F-) | 300 d (F-) |
|--------|---------|---------|---------|---------|---------|---------|---------|---------|---------|
| G1c    | 17.80 ± 8.16 | 10.0 ± 5.60 | 7.74 ± 7.13 | 5.60 ± 7.29 | 2.40 ± .28 | 13.30 ± 1.26 | 11.90 ± .25 | 9.70 ± .48 | 6.00 ± .75 |
| G1e    | 28.86 ± 2.48 | 25.03 ± 1.54 | 15.47 ± .62 | 11.46 ± 40 | 5.81 ± .27 | 20.12 ± .54 | 23.18 ± .28 | 4.17 ± .11 | 1.19 ± .09 |
| G2c    | 13.43 ± 1.18 | 5.90 ± 2.81 | 2.30 ± .38 | 1.50 ± 18 | 1.20 ± .16 | 2.20 ± .18 | 1.50 ± .19 | 24 ± .19 | 0 ± 0 |
| G2e    | 17.47 ± 80 | 8.66 ± 1.46 | 2.85 ± 57 | 1.76 ± 11 | 1.33 ± 14 | 2.24 ± 43 | 4.26 ± 22 | 70 ± 08 | 0 ± 0 |
| G3c    | 32.80 ± 2.46 | 22.00 ± 1.47 | 4.70 ± 47 | 4.20 ± 33 | 11.90 ± 25 | 14.40 ± 96 | 4.30 ± 28 | 2.40 ± 10 | 12 ± 05 |
| G3e    | 32.8 ± 1.7 | 30.3 ± 3.8 | 14.43 ± 80 | 5.59 ± 26 | 12.25 ± 23 | 16.48 ± 20 | 4.42 ± 41 | 3.64 ± 09 | 1.34 ± 05 |
| G4c    | 10.00 ± 2.08 | 3.20 ± 20 | 2.50 ± 28 | 2.20 ± 21 | 1.90 ± 26 | 1.19 ± 94 | 1.88 ± 21 | 2.90 ± 08 | 0 ± 0 |
| G4e    | 34.99 ± 4.05 | 23.29 ± 20 | 12.63 ± 38 | 11.35 ± 46 | 7.84 ± 30 | 6.14 ± 11 | 4.73 ± 14 | 3.95 ± 40 | 1.43 ± .34 |

F: fluoride release in parts per million, G1: Fuji IX Extra, G2: Ketac Molar, G3: Ionomifil Molar, G4: Fuji IX, c: control group e: experimental group, mean ± standard deviation.

3. Results

3.1. Initial fluoride release
Fluoride release of each conventional glass ionomer cement and the experimental group reinforced with TiO2N (control and experimental groups, respectively) were evaluated from days 1 to 300 (Table 1).

All tested materials released measurable amounts of fluoride throughout evaluated periods. Regarding control groups, the highest amount of fluoride was released during the first 24 hours mainly in G3c and G1c, with values of 32.80 ± 2.46 and 17.80 ± 8.16 ppm, whereas G4c released the lowest levels of fluoride with a value of 10.00 ± 2.08 ppm. After 48 hours, a considerable decrease was observed in all conventional glass ionomer cements, showed in Figure 2, during this period, the G3c released 22.00 ± 1.47 ppm and the G4c released 3.20 ± 0.20 ppm.

On the other hand, the glass ionomer cements reinforced with TiO2N enhanced the fluoride release in all groups. This improvement was more remarkable for the G4e which presented values of 34.99 ± 4.45 ppm, showing a 3-fold-increase than that in the conventional glass ionomer cement.

The G1e and G2e showed values of 28.86 ± 2.48 ppm and 17.47 ± 0.80 ppm, respectively; the G3e did not show significant differences during the first 24 hours. Nevertheless, this experimental group (G3e) obtained a more constant fluoride release with 30.3 ± 3.8 on the second day with a slight decrease of 2 ppm approximately. During this period, a decrease was observed in the rest of the experimental groups, which was more remarkable in the G2e and G4e (8.66 ± 1.46 and 23.29 ± 0.20 ppm, respectively).

After day 6, experimental groups G1e and G4e presented the highest fluoride amount (15.47 and 14.43, respectively) until day 10 of study. For day 90, G1e and G3e released the highest amount of fluoride (13.30 ± 1.26, 14.40 ± .36, respectively) until the end of the study. The G2e and G2e stopped releasing fluoride on day 240, whereas the rest of the study groups maintained releasing fluoride up to day 300, with exception of G3c and G4c.

Significant differences can be observed (Table 2) when comparing experimental and control groups through the Student’s t test after 24 hours, exception for G3e (Ionomifil molar) which did not show significant differences during the highest release of GICs. However, in the last period when all GICs released measurable amounts of fluoride another comparison was performed and can be observed that G3e showed a significant difference like the rest of the experimental groups after 240 days.

3.2. Fluoride recharge
A higher concentration of fluoride release was observed after recharging with the fluoride gel on day 31 (Table 3), mainly in experimental groups: G1e, G3e, and G4e (5.81 ± .27, 12.25 ± .23,
7.84 ± .30, respectively). This fluoride release was sustained until the second recharge. Significant differences in fluoride release between control and experimental groups after the first recharge were observed through a Student’s t test in G1e-G1c, G3e-G3c, and G4e-G4c (P values: .001, .010, and .001 respectively). The second recharge, evaluated on day 180 of the study, showed better results in G1e concerning the rest of the groups, with a mean of 23.18 ± .47 on day 180, demonstrating significant differences when compared to the control group (P = .001). The G2e showed significant differences in this period with respect to the control group (P = .001). Nevertheless, the amount of fluoride released by this group was inferior with respect G1e (4.26 and 23.18 ppm, respectively). The most of experimental groups demonstrated a great ability to be recharged (Table 3).

### 3.3. Cytotoxicity activity

Gingival fibroblast cells did not show any cytotoxicity regarding G1e and G2e groups when compared to control groups (P = .51), G3e and G4e demonstrated a significant results vs control group (P = .05) showed in Tables 4 and 5. Significant results were observed between experimental groups when compared with the control group. The Mann–Whitney U test allowed us to identify differences for G3e and G4e groups compared to the control group in the cytotoxic activity (Table 5).

### 4. Discussion

Fluoride release is considered one of the most important advantages of glass ionomer cements. Some authors mention that fluoride release shows a pattern of initial rapid release, followed by a marked decrease.[27] Nigam et al.[31] reported that among the fluoride releases from various dental materials during the first 24 hours, glass ionomer cement is the material that released the highest amount of fluoride with 57.97 ppm. Likewise, this author mentions that a marked decrease occurs after the first day of evaluation. Similar results were obtained in our research during the first 24 hours, with a subsequent and remarkable decrease for the second day of the experiment.[31]

The glass ionomer cement used as control, that released the highest fluoride amount after 24 hours, was G3c (Ionofill Molar) with 32.80 ppm, followed by G1c (Fuji IX Extra) which released 17.80 ppm. Some authors evaluated Fuji IX Extra and reported greater values (33.9 ppm) than those found in our study. This study also reported values of 10.9 for Ketac molar (G2c) and 19.7 ppm for Ionofill molar (G3c). In our study, we observed values of 13.43 and 32.80 ppm, respectively, for these materials.[32]

Several studies have focused on assessing the effect of nanotechnology on the mechanical properties of GICs. In previous investigations, the TiO$_2$N have been used in glass ionomer cements to modify the properties in these materials like fluoride release. Elsaka et al.[33] evaluated for 1 month the fluoride release of glass ionomer cements reinforced with TiO$_2$N at several concentrations (3%, 5%, and 7%) and stated that there are no significant differences when nanoparticles were incorporated. Nevertheless, in our study, statistically significant

![Figure 2. Fluoride release of each glass ionomer during 300 days of study, for control, and experimental groups.](image-url)
**Table 3**
Fluoride recharge ability enhancement of experimental groups after the first and second recharge.

| Study groups | Mean ± of F- after recharge on day 31 | t     | P     | Mean ± of F- after recharge on day 180 | t     | P     |
|--------------|--------------------------------------|-------|-------|----------------------------------------|-------|-------|
| G1c          | 2.40 ± .28                           | −24.141 | .001* | 11.90 ± .25                           | −82.670 | .001* |
| G1e          | 5.81 ± .27                           |         |       | 23.18 ± .28                           |         |       |
| G2c          | 1.20 ± .16                           | −1.662  | .119  | 1.50 ± .19                            | −26.143 | .001* |
| G2e          | 1.33 ± .14                           |         |       | 4.26 ± .22                            |         |       |
| G3c          | 11.90 ± .25                          | −2.824  | .010* | 4.30 ± .28                            | −6.88  | .515  |
| G3e          | 12.25 ± .23                          |         |       | 4.42 ± .41                            |         |       |
| G4c          | 1.90 ± .26                           | −41.451 | .001* | 1.88 ± .21                            | −31.270 | .001* |
| G4e          | 7.84 ± .30                           |         |       | 4.73 ± .14                            |         |       |

G1: Fuji IX Extra, G2: Ketac Molar, G3: Ionofill Molar, G4: Fuji IX. c: control group, e: experimental group, F-: fluoride release in parts per million, ±: standard deviation, t: Student’s t test value, P: statistical significance.

*Statistical significance ≤ .05.

**Table 4**
Cell viability assay of glass ionomer cements modified with TiO2N at 3%.

| Groups      | G1e (%) | G2e (%) | G3e (%) | G4e (%) | Control (%) |
|-------------|---------|---------|---------|---------|-------------|
| Sample 1    | 0.159   | 0.413   | 0.845   | 1.796   | 0.370       |
| Sample 2    | 0.506   | 0.209   | 0.624   | 1.129   | 0.380       |
| Sample 3    | 0.475   | 0.725   | 0.726   | 1.325   | 0.364       |
| Mean        | 0.380   | 0.449   | 0.731   | 1.416   | 0.371       |
| Cell viability | 102.42 | 121.02  | 197.03  | 381.67  | 100         |

Kruskal–Wallis test P = .032 *

G1e: Fuji IX Extra, G2e: Ketac Molar, G3e: Ionofill Molar, G4e: Fuji IX, e: experimental group, TiO2N: Titanium dioxide nanoparticles, P: statistical significance.

*Statistical significance ≤ .05.

**Table 5**
Cell viability individual comparison of each experimental group versus control group using Mann–Whitney U test.

| Groups          | Mann–Whitney U | P value |
|-----------------|----------------|---------|
| G1e-control     | 3.000          | .51     |
| G2e-control     | 3.000          | .51     |
| G3e-control     | .000           | .05*    |
| G4e-control     | .000           | .05*    |

G1e: Fuji IX Extra, G2e: Ketac Molar, G3e: Ionofill Molar, G4e: Fuji IX, e: experimental group, P: statistical significance.

*Statistical significance ≤ .05.

differences were found until 8 months (P = .001) as shown in Table 2. These results agree with those mentioned by other authors who incorporated TiO2 nanoparticles to glass ionomer cements and reported an increase in the amount of fluoride released.\[14\]

Several authors used the mouth rinse for 1 minute at 300 ppm to recharge dental materials, and they did not report any changes in the fluoride concentration.\[15\] Han et al.\[15\] stored the samples inside a mouth rinse for 20 days in concentrations of 450 and 900 ppm, which showed an increase in fluoride concentration, mainly with a concentration of 900 ppm.

In the present study, titanium dioxide nanoparticles enhanced the recharge capacity in all experimental materials, after the first recharge (day 30), G1e (Fuji IX extra + TiO2N), and G4e (Fuji IX + TiO2N) released the 2-fold and 3-fold increase in the amount of fluoride, respectively. Concerning the second recharge (day 180) similarly, G2e (Ketac molar + TiO2N) enhanced their recharge ability by 65% and G1e and G4e showed similar results as the first recharge.

It has been shown that the use of dental materials with NP aggregates, produces a biological impact on oral tissues, for this reason, the present study analyzes the cytotoxic capacity of glass ionomers reinforced with TiO2N through MTT method which is designed for determining cell viability spectrophotometrically by determining the mitochondrial activity in living cells. This method is simple, accurate, and yields reproducible results. When comparing MTT methodology with others, previous studies have demonstrated that MTT prevent over or under-estimation of cell viability during cytotoxicity evaluation, thus considered reliable.\[16\] As well as, MTT has also been used in previous studies to determine cytotoxic effects caused by different dental materials on HGF,\[17\] human periodontal ligament fibroblast cells,\[18\] and in mouse fibroblast cells.\[19\]Likewise, previous studies have evaluated cell viability of TiO2N using MTT method on human cells.\[20\]

Glass ionomer cements are capable of being cytotoxic even without incorporating the metallic elements, which can be produced due to the presence aluminum phosphate and calcium fluoride in the powder. On the other hand, Patel et al.\[21\] exposed the glass ionomer samples with TiO2N in cell cultures and after 24 hours were cytotoxic with a frequency of cellular aberrations at 75 and 125 μM (micromole), while at 25 μM no positive results were observed.

The outcomes of our study did not show relevant cytotoxic effects in the samples of glass ionomer cements reinforced with TiO2N at 3% in all study groups, which agrees with reported by other studies.\[22-20\] The highest cytotoxic effects were observed in G3 and G4. Nevertheless, the groups G1 and G2 showed minimum levels of cytotoxicity.

**5. Limitations of the study**

Authors would consider the need of measuring GICs mechanical properties as compressive strength, flexural strength, bond strength and microhardness, in future studies, so that a complete efficacy of TiO2N incorporated to glass ionomer cements would be fully studied. Although a detailed clinical evaluation would be also necessary for further future studies.

The incorporation of TiO2N at 3.2%, enhance the fluoride release in all glass ionomers evaluated in this study, and the highest and more constant fluoride release was seen in the experimental glass ionomer cements Fuji IX and Fuji IX EXTRA.
These results suggest that incorporation of TiO$_2$N to glass ionomer cements improves the fluoride release and recharge ability of this materials, therefore it will be able to avoid demineralization and preserve the health of the dental surface, without cytotoxic effects on oral tissues.

**Author contributions**

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