## *In vitro* cytotoxicity of acrylic resins with different polymerization methods

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The polymethylmethacrylate used in Dentistry <sup>(1)</sup> is produced synthetically <sup>(2)</sup> by the transformation of monomers into polymer - polymerization <sup>(3)</sup>. Activation of this reaction can be done by several methods <sup>(3)</sup>, which seems to influence the amount of monomer that does not integrate the polymer chain (residual monomer). When released into the oral cavity, it can lead to local adverse reactions <sup>(4)</sup>. For this reason, its toxicity <sup>(5)</sup> has been investigated, starting with in vitro cell studies <sup>(6)</sup>.

**Objective:** To compare, in vitro, cell viability (CV) of two acrylic resins, one with conventional thermal polymerization [ProBase@Hot (PBH)] and another with injection [IvoBase@System (IBS)].

## Materials & Methods:

 PBH and IBS disks (Ø=5mm x h=2mm) (n=4) were incubated (37°C, 5% CO<sub>2</sub>) in 7 mL of DMEM cell culture medium (figure 1) during the study times: 30 min (TO), 24 hours (T1), 7 days (T7), 14 days (T14) e 1 month (T30).



Figure 1 - Specimens



 200 µl of the resin extracts were placed in contact with 2x10<sup>4</sup> 3T3 mouse fibroblastic cells (figure 2) and the cell viability assessed by the MTT assay, measuring the absorbance at 595 nm on a Model 680 microplate reader (Bio-Rad, California).

Figure 2 - TA = 400x

3. Three independent assays were performed for each resin / time with 8 replicates and positive controls (cells not exposed) and negative (DMSO-exposed cells) in each assay (figure 3).

100						20
					XX	4.94.99
2	1	$\sim$	1	$\sim$	$\approx$	XX
-	2	<u>44</u>	10	1	$\sim$	20
1		101				44
1100	ere	101	00		No. Con	23
	e i c	101	00	00	20	100
UP (C)	10 10	0	6		20	44 (9)

Figure 3 - Cell culture 96 well plates.

- 4. The measured absorbance values were converted into percentages of cell viability through the ratio between the values under study and positive control.
- 5. The results of this in vitro prospective longitudinal experimental study were analyzed using the Kolmogorov-

## Results & Discussion:

From the analysis of the cell viability values (graph 1) there was no statistically significant difference between the two resins if we considered the results of the incubation times all together (p=0,249). This may indicate that there are no differences in the residual monomers release, being in agreement with some published studies (7,8).

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For each incubation time there were differences between the two resins only in T14 and T30 where IBS promoted higher (p=0,015) and lower (p<0,05) cell viability than PBH, respectively.

Analyzing the evolution of cell viability within the different incubation times for each resin we verified statistically significant differences. For PBH, in T7 the value of cell viability was significantly higher (p<0,05) than the other times and the lowest in T14 (p<0,05). These differences found among times for the PBH resin don't seem to have a predictive pattern.

For IBS, in the TO and T30 the cell viability values were

Smirnov and Levene test. Having verified the normality of the distribution and homogeneity of the variance, we used parametric comparative tests of one-way ANOVA and T-Student for a significance level of 5%.

significantly lower (p=0,012) than in the other times. From a clinical point of view this result may indicate that local adverse reactions may be more likely to occur within the first 30 minutes or after 30 days of prothesis placement.

**Conclusions:** The type of polymerization has not showed significant effects on cell viability and it is not expected to produce different local adverse reactions. According to ISO 10993-5: 2009 <sup>(9)</sup>, where the cell viability limit is set at 70% <sup>(9)</sup>, the studied resins are not considered cytotoxic. In order to evaluate if there is a tendency to decrease cellular viability of the IBS it may be important, in the future, to extend the incubation time.

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