Handling Property Improvement of Nano Calcium Hydroxide from Indonesian Limestone with Different Solvent Vehicles

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Abstract

As intracanal endodontic medicament, calcium hydroxide [Ca(OH)2] paste needs to have good handling to reach the root canal system's complexity and penetrate the dentinal tubule. This study aimed to evaluate the handling of synthesized nano Ca(OH)2 from natural limestone (CaCO3) of Palimanan, Indonesia, formulated with different solvents; distilled water, propylene glycol (PG), and polyethylene glycol (PEG). The handling property of the nano Ca(OH)2 prototype was evaluated compared to the conventional micro-sized Ca(OH)2 particle paste. Both Ca(OH)2 powder were mixed with three different solvents: water, PG, and PEG. Flowability was evaluated by measuring the spread area following ISO1566 for zinc phosphate cement. Injectability test was done by measuring paste quantity extruded through a syringe after applying 300N forced at a cross-head speed of 0.25mm/sec in the 30s in a Universal Testing Machine. Data were analyzed by ANOVA (P<0.05) and T-test as a post hoc multiple comparison tests. The handling of the nanosize Ca(OH)2 prototype was significantly higher than the conventional Ca(OH)2. PG with nanosize Ca(OH)2 prototype demonstrated the best performance in flowability, while the water solvent had the best injectability compared to other solvents. The injectability and flowability of Ca(OH)2 paste is related to its particle size and solvent, which were improved by nanosized Ca(OH)2.

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Introduction

Calcium Hydroxide [$Ca(OH)_2$] is widely used as endodontic material. The endodontic material $Ca(OH)_2$ is available commercially in various forms for pulp capping, intracanal dressing, root canal sealer, apexogenesis, apexification, and root resorption treatment.[1-4] It has been reported that the application of $Ca(OH)_2$ eliminates microorganisms effectively. The antimicrobial effect of $Ca(OH)_2$ in the dentinal tubules may or may not effective depending on its handling property. [5-7]

Endodontic medicaments must penetrate dentinal tubules and kill bacteria because bacteria located inside dentinal tubules are protected from host defense cells, systemic antibiotics, and chemo-mechanical preparation.[8-10] It should have the ability to be in contact with bacterial products deeply inside the dentinal tubules to insult the bacterial activity effectively. It has been suggested to insert the $Ca(OH)_2$ particle into dentinal tubules to act as a direct resource of dissociated $Ca(OH)_2$. Applying $Ca(OH)_2$ powder alone is difficult; therefore, it needs a liquid as a vehicle. The limitation of the flowability and the $Ca(OH)_2$ paste diffusion may lead to the failure or prolonged endodontic treatment.[11-14] $Ca(OH)_2$ in an injectable syringe is clinically more preferable because it can deliver the material more conveniently in a more significant time. It is also more suitable for a root canal with limited accessibility. $Ca(OH)_2$ paste in suitable injectability and flowability, can reach small and narrow areas in the root canal system.[15-16]

As a cement, the powder's characteristic will determine cement paste performance, including its handling characteristic.[17-18]

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Modifying the particle size of cement into a smaller size will change the paste's property or set cement. The larger specific surface area of cement particles allows the higher flowability of cement paste and produce smaller porosity void.[19-20] Furthermore, a smaller particle size presents high doses of material to achieve the maximum level of ions release to exert the antimicrobial effect and hard tissue stimulation.[21]

Kobayashi et al. reported that commercial $Ca(OH)_2$ has microparticles size around 0.5-2.5μm while the average diameter of dentinal tubules is 2-5μm^2.[17] Inside the dentinal tubules, the bacterial product is penetrated deeply from 50-100 μm. Recently, applying nanotechnology gives a new approach to reducing particle size into nanosized to improve the characteristic of the various material to achieve more highly active and effective materials. The nanotechnology of materials could modify the material at the molecular or atomic level to change the chemical and physical properties of materials.[21] Nanosize of $Ca(OH)_2$ increases the particles' possibility of being inserted deeply inside the dentinal tubules in high doses. In this situation, the effectiveness of $Ca(OH)_2$ medication is expected to be improved.

In the previous study, $Ca(OH)_2$ has been successfully obtained from the Indonesian natural limestone from the Palimanan area in West Java. The synthesized $Ca(OH)_2$ prototype has the potential to be developed as an endodontic materials.[22] In this study, the obtained $Ca(OH)_2$ prototype was formulated in nanosized with various solvent, and the handling property, including flowability and injectability, was investigated.

Materials and methods

Preparation of nanosized $Ca(OH)_2$ prototype. $Ca(OH)_2$ powder from the natural source of limestone ($CaCO_3$) from Palimanan, Cirebon, Indonesia, obtained from the previous study.[22] Obtained powder $Ca(OH)_2$ then followed crushed in the planetary ball mill with PG solution as media for 2h to obtained nanosize of $Ca(OH)_2$. The obtained sample then dried in the oven at 60°C for 8d. Dried nano $Ca(OH)_2$ was kept in the desiccators before evaluation. Conventional $Ca(OH)_2$ powder (EMSURE, Germany) was used as a comparison. All samples were subjected to a particle size analyzer (Horiba, SZ-100).

Cement Paste. Nano $Ca(OH)_2$ powder obtained, and the conventional $Ca(OH)_2$ was mixed with each solvent at the ratio liquid to powder (L/P) 0.5, 0.75, and 1.0 with a spatula on a glass slab for 1m to form a paste consistency. The solvents used in this study were distilled water, PG (USP EP, China), and PEG 400 (batch J0195/18e, PT. BRATACO, Indonesia).

Flowability measurement. The cement flowability index was evaluated by measuring its spread area according to the international standard ISO1566 for dental zinc phosphate cement. In the present study, a glass plate weighing 2kg was placed on top of 0.2mL cement paste for 3cm to spread out the cement paste on the glass surface. The L/P of cement paste was represented at 1.0. Then, the spread area from each sample group was scanned and measured.

Injectability measurement. The cement paste was packed in a 3mL syringe (of inner diameter 8.5mm and plunger travel length of 5cm) with a No.18 gauge (0.85mm ID, and 1.45mm OD). The 2cc of cement paste was filled in the syringe and packed by pressing the plunger. The syringe was fitted vertically in a fixture and put in a platen of a 300N force. The average of manual thumb force at cross-head speed 0.25mm/sec in 30s was apply using a universal testing machine, and the quantity of extruded paste was evaluated in gram.[23-29] The ratio of cement paste for injectability measurement was L/P 0.5-1.0.

Statistical analysis. Results were expresses as mean±standard deviation (SD). Data were statistically analyzed by ANOVA at a significance level of 5% ($P<0.05$) and T-test method as a post hoc multiple comparison tests were performed to evaluate significant differences between groups.

Results

Table 1 showed particle size distribution of $Ca(OH)_2$ powder of (a) conventional $Ca(OH)_2$ obtained commercially as a control group, (b) prototype $Ca(OH)_2$, and (c) nanosized prototype. The particle size of nano $Ca(OH)_2$ prototype was 75% smaller than the conventional one and 95% smaller than before milling.

	$Ca(OH)_2$ prototype before milling	$Ca(OH)_2$ prototype after milling 2h	$Ca(OH)_2$ conventional
Mean	5195.8nm	279.4nm	1101.1nm
SD	2439.7nm	17.0nm	57.6nm
Mode	6401.1nm	278.7nm	1111.0nm

Table 1. Particle size distribution of $Ca(OH)_2$ conventional, and prototype before and after ball miling for 2h.

Figure 1 showed the flowability of cement paste obtained by the spread area test. The spread area of conventional $Ca(OH)_2$ paste with distilled water, PG and PEG were significantly smaller (*P<0.05*) than the nanosize $Ca(OH)_2$ prototype. In the solvents comparison, nano $Ca(OH)_2$ manipulated with distilled water solvent demonstrated a significantly smaller spread area than PG and PEG. No significant differences were observed for PEG as the solvent vehicle for conventional and nano $Ca(OH)_2$ prototype. PG and PEG improved the spread area showing a good flowability index for both conventional and nano $Ca(OH)_2$ paste compare with distilled water solvent.

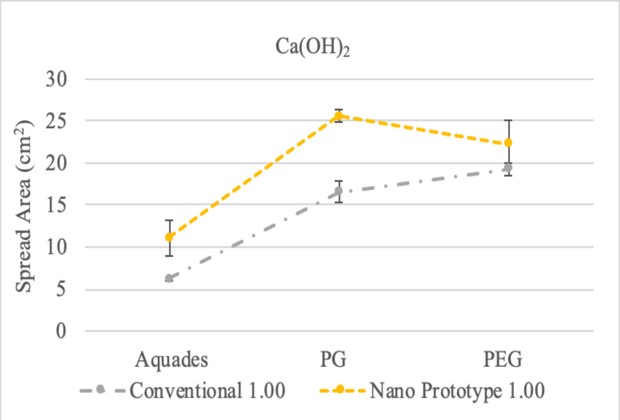

Figure 1. Spread area measurement as flowability index of conventional $Ca(OH)_2$ and nano $Ca(OH)_2$ prototype at the ratio L/P 1.0.

The injectability of conventional and nano $Ca(OH)_2$ prototypes at the L/P ratio of 0.5-1.0 is shown in Figure 2. Increasing the L/P ratio resulted in improvement of injectability, whereas at the same L/P ratio, the injectability of nano $Ca(OH)_2$ prototype was significantly higher than conventional. At a lower L/P ratio of 0.5, conventional $Ca(OH)_2$ paste mixed with PG and PEG could not be extruded out of the cannula. Contrary to the nano $Ca(OH)_2$ prototype, there were no significant differences for all solvent, except at the higher L/P ratio of 0.75

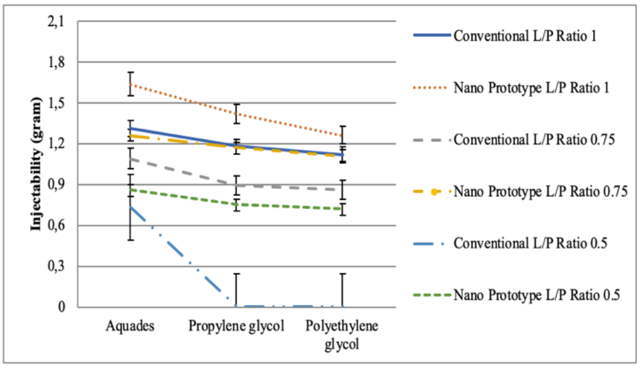

Figure 2. Paste extruded mass as injectability index of conventional $Ca(OH)_2$ and nano $Ca(OH)_2$ prototype at the ratio L/P 0.5, 0.75 and 1.0.

Discussion

The results of this study provide some insights that certain aspects, particle size, L/P ratio, and solvents contribute to the handling properties of $Ca(OH)_2$ paste. The particle size of $Ca(OH)_2$ synthesized from Indonesian limestone can be minimized to nanosize after 2h of milling. Even though it was limited to 279.4±17nm, respectively, increasing milling time more than 2h might be more decreasing the particle size, if desired. According to the study of Kobayashi et al., particle size was no impaired to the bacteriostatic level of $Ca(OH)_2$.[17] Therefore, the nanosize $Ca(OH)_2$ might be improving other characteristics of $Ca(OH)_2$ as medicament intracanal without altering the bacteriostatic effect of $Ca(OH)_2$.

Paste flowability can be measured either with a rheometer or measurement of a spread are paste film between two glass slabs under load.[28] In the present study, the spread area of cement paste between two glass slabs under 2kg (ISO1566) was used. This method is suitable to evaluate $Ca(OH)_2$ paste when the thicker paste is employed or when the paste was manipulated by mixing the paste and liquid prior to application. The spread area in figure 1 showed that the flowability of conventional $Ca(OH)_2$ paste was significantly smaller than nanosized $Ca(OH)_2$ paste. The nanosized of $Ca(OH)_2$ paste at all solvent variables showed significantly higher flowability than conventional. The nanosized particles are more easily moved and spread out than a conventional $Ca(OH)_2$ paste with a larger particle size. The results support the fact that decreasing the $Ca(OH)_2$ particle size could improve the flowability of $Ca(OH)_2$ paste. Nanoparticles can accommodate better particle dispersion in a solvent.

Since the 1970's PEG and PG have been used as a solvent for $Ca(OH)_2$. Both are non-aqueous solvent and contribute to the improvement of chemical and physical properties of endodontic paste such as dissolution rate, hydroxyl ion release, and the clinical handling.[30-35] Incorporating PG and PEG significantly improves the flowability of $Ca(OH)_2$ paste (Figure 1). Teoh et al. reported that $Ca(OH)_2$ paste with PEG as vehicle and thickener had more significant pH change and more bio-available hydroxyl ion release in the dentinal tubules than those containing a mixture of PEG-water, water, or saline.[32] These findings promise better bioactivity of nano $Ca(OH)_2$ prototypes with PEG as the solvent.

$Ca(OH)_2$ paste in an injectable syringe is clinically more preferable because it can deliver the paste more conveniently in a more significant time. In other words, it has excellent handling property, and it is very suitable for root canal medicament with limited accessibility. As shown in Figure 2, the injectability of $Ca(OH)_2$ varied depending on the solvent, L/P ratio, and particle size. A non-aqueous solvent like PG and PEG is thicker than water, therefore resulting in the decreasing the injectability of the $Ca(OH)_2$ paste.[34-35] It might be related to the canula's backpressure in the syringe, which was prevented the injectability of $Ca(OH)_2$ paste with PG and PEG. Another problem related to the injectability is the filter pressed phenomena where the solvent and powder particle were separated during the filtering. In this case, the paste's density extruded, and the excess paste that remains in the syringe might be different.[26] The higher injectability of $Ca(OH)_2$ paste with water solvent can be related to its higher hygroscopic property and lower viscosity of water compared to PG and PEG. As expected, the nanosized $Ca(OH)_2$ paste with all solvent demonstrated significantly higher injectability than conventional $Ca(OH)_2$ paste with the same solvent. This supports the fact that smaller particle size could be dispersed more efficiently in their solvent vehicle and are easier to move through the syringe cannula. Smaller particle size also improved the particle dissolution and minimize residual unreacted particle, which also help $Ca(OH)_2$ to penetrate deeper to the dentinal tubules and improve it antimicrobial activity.[36] Considering the side effect of the limited injectability of the non-aqueous solvent in this study, the L/P ratio can be adjusted to achieve excellent handling and bio-activity of nano $Ca(OH)_2$ prototype.

Conclusions

Nano-sized $Ca(OH)_2$ prototype paste synthesized from Palimanan limestone has significant advantages over the conventional micron-sized particles. Handling property was improved by nano $Ca(OH)_2$ particle size paste with distilled water, PG, and PEG solvent. The results of this study gave valuable insights for further development of nano $Ca(OH)_2$ paste formulation from the natural limestone of Palimanan Indonesia to be suitable for clinical application.

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Declaration of Interest

The author declared no conflicts of interest

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