Clinical & Histologic Study of Human Pulpal Response to Calcium Hydroxide, Resorcinol & Zinc Oxide Eugenol as a Direct Pulp Capping Agent

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The healthy pulp has the inherent potential for healing provided the exposure site is kept bacteria free. Various materials have been tried as direct pulp capping (DPC) agents. In this study, calcium hydroxide, Resorcinol (Resinifiying Agent) and zinc oxide eugenol were used as DPC agents. Under aseptic conditions, Class I cavity was prepared on premolar teeth and small exposure was made at pulp horn and experimental materials were placed over the exposure site and remaining cavity filled with quick setting zinc oxide eugenol cement. After treatment, the teeth were extracted and processed for routine histologic evaluation.

Teeth with calcium hydroxide show resolution of inflammatory response and reparative dentin formation after 90 days. Teeth with resorcinol showed increase fibrosis and widened predentin layer whereas zinc oxide eugenol shows only chronic inflammation and no evidence of reparative dentin formation. By evaluating clinical and histological response with these materials, calcium hydroxide and resorcinol can be used as a direct pulp capping agent but long term study required.

Key words: Direct pulp capping agents, calcium hydroxide, resorcinol

A constant concern in the practice of dentistry is the maintenance of tooth vitality. The objective of pulpal therapy is the conservation of tooth in a healthy state A non-vital tooth requires 2.5 times more of a load to register a proprioceptive response than vital tooth.[1]

Exposure of dental pulp can happen as a result of caries, trauma or cavity preparation .The accepted line of treatment is the direct pulp capping. Direct pulp capping is defined as a procedure in which the exposed pulp covered with suitable material in an attempt to preserve its vitality. A wide array of materials like calcium hydroxide,zinc oxide eugenol cement,dentinchips, glass ionomer cement, MTA, biodentin,and many more materials had been tried as pulp capping agents.[2] One of the first material to show promising result was calcium hydroxide. Numerous subsequent studies have demonstrated dentinal bridge formation in about 50 to 80 % of cases treated with Ca(OH)2 formulation.[3,4,5] Recently introduced MTA had been used in pulp capping procedures demonstrating remarkable success than calcium hydroxide.[5,6,7] But it is very expensive material.

Resorcinol (resinifying agent) had been successfully tried in the past as a direct pulp capping agent and it shows promising results.[9] Resorcinol offers significantly economical incentives.

Present study was carried out to investigate human pulpal response to calcium hydroxide, resoecinol and zinc oxide eugenol cement as a direct pulp capping agents. zinc oxide eugenol cement used as a negative control.

AIMS & OBJECTIVE

The purpose of this study was to find out clinically and histologically the effectiveness of calcium hydroxide, Resorcinol (Resinifying Agent) zinc oxide

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eugenol cement as a direct pulp capping agent on human dental pulp.

MATERIALS & METHODS

The study was carried out on 90 non-carious, sound premolar teeth to be extracted for orthodontic reason from 28 young patients of both sexes in the age group 14 to 25 years. The inform consent of human subjects who participated in this study were obtained after the nature of procedure and possible discomfort and risk had been fully explained.

1) **Group I** – Self cured radio-opaque calcium hydroxide cement.
2) **Group II** – Resorcinol (Resinifying agent)
3) **Group III** – Resin bonded zinc oxide eugenol cement was used as control material.

The selected tooth was anaesthetized by using 2% liagnocaine hydrochloride. Tooth was isolated with rubber dam which was swabbed with 5% tincture of iodine and ligated with dental floss to avoid leakage. Class I cavities were prepared with suitable tungsten carbide bur in an air turbine with water spray without perforating the pulp chamber. Pin point exposure of the pulp horns were attempted with round bur rotating at slow speed. The cavities were washed with sterile cotton pellet moistened in sterile saline solution to the exposure point to control any hemorrhage. (Fig. 1) The capping materials were placed on the exposure site and entire cavity floor. The remaining cavity was sealed with quick setting zinc oxide eugenol cement.

All patients were checked for 1-3 days, after the procedure then after 1 week, 1 month and at intervals usually monthly up to 3 months. The teeth were extracted under local anesthesia. Extractions were performed over period of 7 days, 30 days, and 90 days after treatment of pulp exposures.

After extractions, immediately the apical third of anatomic root, or roots of tooth was removed by means of water coolant diamond wheel to facilitate the fixing of the pulp.

The remainder of the tooth was placed in 10% formalin solution for fixation, followed by decalcification in 8% nitric acid. The teeth were then processed and embedded in paraffin wax and serial sections of 6 microns thickness were obtained and stained with hematoxylin and eosin stain

OBSERVATION & RESULT

Patients were recalled for clinical examination.

- Questioning the patient about any postoperative pain
- Clinical examination of the condition of the filling.
- Mobility, vitality & percussion test of treated tooth.

Specimen subjected to histologic evaluation at different intervals of 7, 30, and 90 days and graded accordingly.

- Inflammatory cell response
- Fibrosis
- Odontoblastic differentiation
- Reparative dentin formation

The number of teeth and data recorded in clinical as well as histological groups are summarized in table 1 and table 2 respectively.

DISCUSSION

Armstrong and Hoffman evaluated the success of the clinical cases by the absence of pain and pathologic changes, vitality and their normal physiological performance in the mouth, whether secondary dentin is formed or not.[10]

The specimen treated in this study can be considered successful according to Armstrong and Hoffman.[11] As clinically, there was no significant difference between each group over a period of 7 days, 30 days and 90 days. Hence all three materials counted as successful pulp capping agent (Table 1).

The formation of a reparative dentine barrier is essential for the health of the pulp tissues if the integrity of the pulp is taken into consideration. The absence of inflammatory cells adjacent to the barrier, the good staining properties of treated pulps, and the retaining of the pulp structure with the functioning vas-

cular system are the signs of healing of pulpal lesions.[11]

The results of present study indicate that after 7 days, almost in all cases pulp tissues immediately in contact with calcium hydroxide is usually completely dearranged and distorted because of the caustic effect of the drug. This superficial coagulated necrotic zone consists of the dentin, dentinal fragments, haemorrhage, blood clot, dilated blood vessels and particles of calcium hydroxide (Fig.2)

At the end of 30 days, inflammation is reduced, odontoblatic differention seen and widened predentin layer shows early signs of pulp healing (Fig 3). At the end of 90 days, reparative dentin formation was seen covering the exposure site. (Fig 4)

The healing response of Calcium hydroxide was found similar to Heys and colleagues. Cvek, Glass, Zandar & Sazak who demonstrated healing and bridge formation after 5 to 8 weeks after capping.[2,12] In contrast Fitzgerald, Cox & Mjor & Tronstad showed that repartive dentin formation was seen after short periods of 7, 10 and 14 days. However the general concept about this condition was that fibrosis increases with collagen synthesis beginning from 15th day. After 30 days, incomplete hard tissue bridge formation was seen in fibrous areas. In present study, similar findings were seen.

When calcium hydroxide is applied directly to pulp tissue, there is necrosis of the adjacent pulp tissue and an inflammation of the contiguous tissue. Dentin bridge formation occurs at the junction of necrotic tissue and the vital inflamed pulp tissue.

Although calcium hydroxide works effectively, the exact mechanism is not well understood. Compounds of similar alkaline (pH = 11) cause liquefaction necrosis when applied to pulp tissue. Calcium hydroxide maintains a local state of alkalinity that is necessary for bone or dentin formation. Beneath the region of coagulation necrosis, cells of underlying pulp tissue differentiate into odontoblasts and elaboratre dentin marix in about 4 weeks. In the alkaline medium, enzyme phosphatase is active in releasing inorganic phosphate from blood. Calcium phosphate is then precipitated.

Occasionally in spite of successful bridge formation, the pulp remains chronically inflamed or become necrotic. Internal resorption and dystrophic calcification sometimes occurred with calcium hydroxide.[11]

In present study, resorcinol (resinifying agent) used as direct pulp capping agent. Resinifying therapy was suggested by Wang Mane in 1957 as new method of treating pulpoperiapical disease and it has been widely used in patients since 1960, in China, as an obturating material.[12] The principle of this therapy is that when liquid phenol aldehyde resin when inserted into root canal becomes solid after polymerization. The pulp remains in the canal get resinified.

Dr. Vimal Sikri, tried resorcinol (Resinifying therapy) as a direct pulp capping agent. The clinical, radiologic and histologic evaluation of the material clearly support the evidence that resorcinol can be used as best direct pulp capping agent. It also overcomes the drawbacks of calcium hydroxide.[9,13]

In the present study after 7 & 30 days, remnants of resorcinol material, is seen in the coronal part of the pulp tissue in all cases (Fig.5 & Fig. 6). Middle part of the pulp tissue showed marked fibrosis (Fig. 7). Odontoblastic differentiation was seen and incomplete reparative dentin formation was seen along the lateral wall which indicate that pulp healing would take place. (Fig 8)

The Resorcinol resin when set over the exposure site, prevent its link with the environment. The fibroblastic activities and incomplete reparative dentin formation in 6 cases is suggestive of the view that pulpal tissue would heal. The exposed pulp has good healing potential if symptomless and kept bacteria free. Resorcinol resin has the potential to keep exposure site bacteria free and thus therapy promotes healing.[14]

Hence, it may be used as dierect pulp capping agent. However long term histological studies should be undertaken to ascertain the ultimate success of pulp capping with Resorcinol.

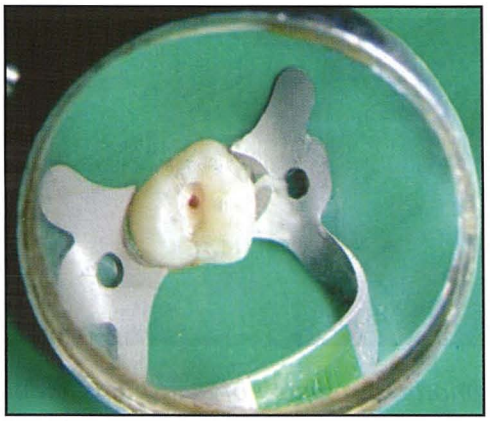


Fig. 1: Photograph Showing Class I cavity preparation with pin point exposure in mandibular premolar tooth

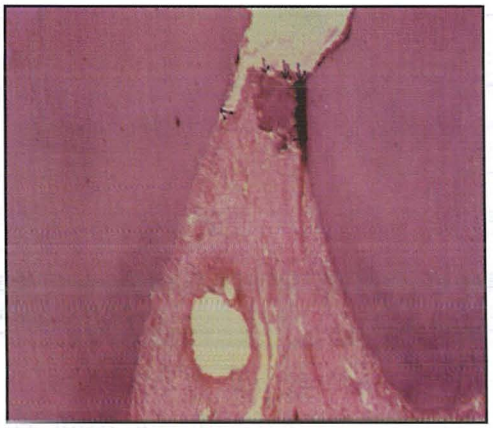


Fig. 2. Photomicrograph of pulp of one of the teeth treated with calcium hydroxide showing haemorrhagic mass (↓↓) & distruption of odontoblastic layer at the exposure siteat observation period of 7 days.(←) [H & E stain x40 magnification

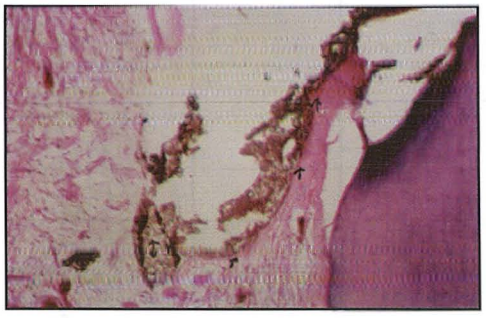


Fig. 3: Photomicrograph of pulp of tooth treated with calcium hydroxide showing increased odontoblastic activity and initial stages of reparative dentin formationat 30 days observstion period.

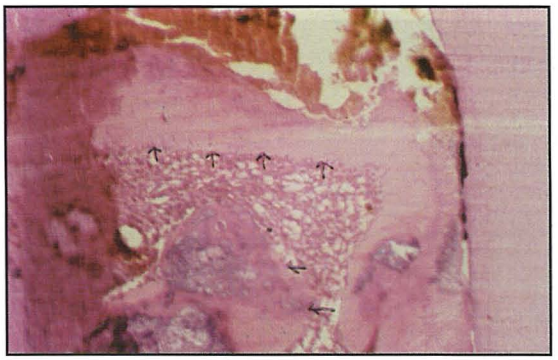


Fig. 4: Photomicrograph ofpulp of one of the tooth treated with calcium hudroxide showing reparative dentin formation covers the exposure site completelyat observation period of 90 days.

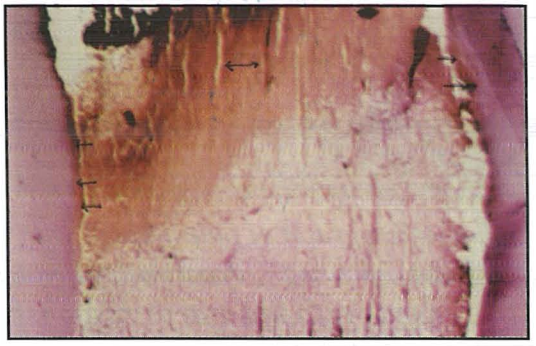


Fig. 5: Photomicrograph of pulp of tooth treated with resorcinol showing ramnents of experimental material at observation period of 7 days.

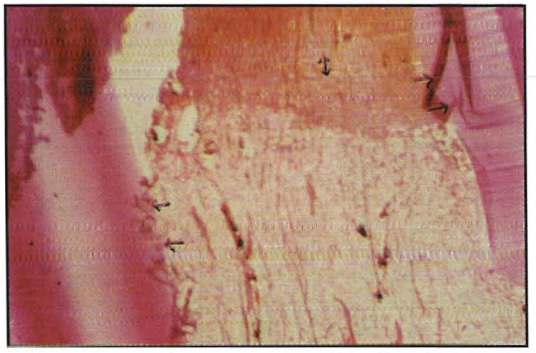


Fig. 6: photomicrograph of pulp treated with resorcinol shows inflammationat a postoperative period of 30 days.

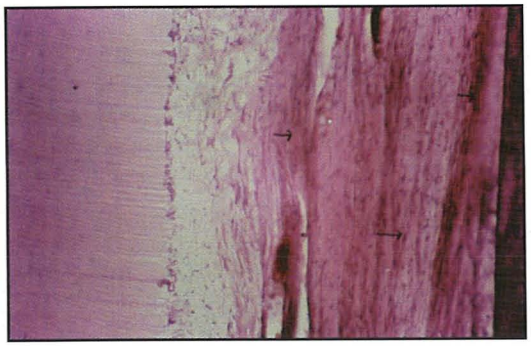


Fig. 7: Photomicrograph dentin and pulp treated with resorcinol showed increased fibrosis



Fig 8. photomicrograph of dentin and pulpof one of the teeth treated with resorcinol showed widened predentin layer and increased odontoblastic activity at a post operative period of 90 days.

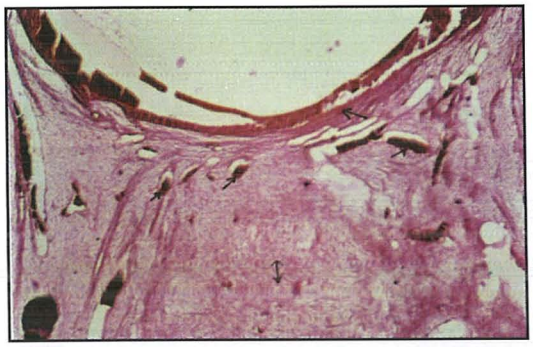


Fig 9. photomicrograph of pulp treated with zinc oxide eugenol showing experimental material and increased inflammatory response at observation period of 7 days.

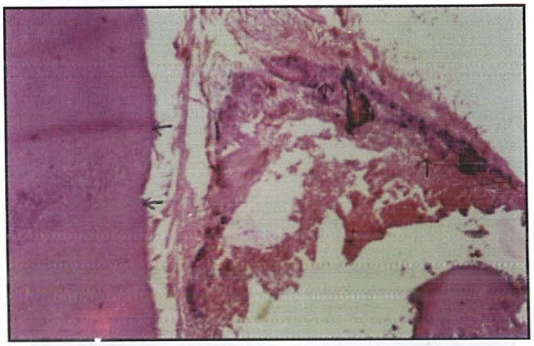


Fig 10. photomicrograph of dentin and pulp of one of the teeth treated with zinc oxide eugenol cement showed chronic inflammatory response.

Table 1

Clinical Evaluation of the patients treated with Calcimol, Resorcinol and Kalzinol

Material	Days	No. of teeth	Pain/ discomfort	Sensitivity to hot or cold	Tenderness	Swelling	Mobility	Normal pulp response
Calcimol	7	10	-	-	-	-	-	10
$Ca(OH)_2$	30	10	1	-	-	-	-	9
	90	10	-	-	-	-	-	10
Resorcinol	7	10	-	-	-	-	-	10
	30	10	1	-	-	-	-	9
	90	10	1	-	-	-	-	8
Kalzinol	7	6	-	-	-	-	-	5
(ZOE)	30	7	1	1	-	-	-	6
	30	7	1	1				6

Zinc oxide eugenol cement when comes in direct contact with pulp tissue, there is little tendency for calcific bridging of the exposure, and the pulp although probably symptomless. It remains chronically inflamed after 7, 30 and 90 days. (Fig.9) Only in 2 cases odontoblastic differentiation was seen and with only one case after 90 days, there was evidence of incomplete bridge formation, whereas chronic inflammation seen after 7, 30, 90 days in almost every case (Fig 10). histologic observations reported in this study were similar to those of Glass & Zander, Nixon & Hannah[15], Watt[16] who stated that pulp response beneath zinc oxide eugenol was unfavourable in pulp capping procedure.

These results disagree with those of Bermann & Massler,[17] who observed over the same time period that zinc oxide eugenol did not cause necrosis of superficial pulp tissues and there was delay but eventful formation of permanent dentinal bridge. Sveen, Rowe reported 87% success with zinc oxide eugenol in capping of primary teeth. Transtad & Mjor found zinc oxide eugenol more beneficial for inflamed exposed pulp and felt that production of calcific bridges is not necessary, if pulp is free of inflammation after treatment.

Table 2

Histological evaluation of direct pulp capping with calcimol, resorcinol & kalzinol.

Days	Material	Total No. of teeth	Inflammation					Fibrosis					Odontoblastic Differentiation				Reparative Dentine Formation			
			0	1	2	3	%	0	1	2	3	%	0	1	2	%	1	2	3	%
7	Calcimol $Ca(OH)_2$	10	-	9	-	1	100	6	4	-	-	40	5	5	-	50	-	-	-	-
	Resorcinol	10	7	3	-	-	30	-	3	2	5	100	10	-	-	0	-	-	-	-
	Kalzinol (ZOE)	5	-	3	2	-	100	5	-	-	-	0	5	-	-	0	-	-	-	-
30	Calcimol $Ca(OH)_2$	10	8	2	-	-	20	1	7	2	-	90	4	3	3	60	4	4	2	60
	Resorcinol	10	5	5	-	-	50	-	7	3	-	100	10	-	-	0	10	-	-	0
	Kalzinol (ZOE)	5	-	-	1	4	100	5	-	-	-	0	5	-	-	0	5	-	-	0
90	Calcimol $Ca(OH)_2$	10	8	1	1	-	20	4	4	2	-	60	-	5	5	100	-	2	8	100
	Resorcinol	10	8	1	1	-	20	-	6	4	-	100	4	6	-	60	4	6	-	60
	Kalzinol (ZOE)	5	-	4	1	-	100	-	4	1	-	100	3	2	-	40	4	1	-	20

Flammatory Cell Response: 0 - No infiltration of inflammatory cells. **1.** Infiltrated by few PMNL; round cells and/or macrophages. **2.** Infiltrated by moderate number of PMNL, round cells and/or macrophages. **3.** Massive infiltration of PMNL, round cells and/or macrophages.

Fibrosis: 0. None or slight. **1.** Mild fibrosis. **2.** Moderate fibrosis. **3.** Severe fibrosis.

Odontoblasic differentiation: 0. No identifiable matrix forming cells across or beneath the exposure. **1.** Early or incomplete differntion of matrix forming cells associated with irregular or regular fibrillar matrix. **2.** Highly differentiated cells associated with irregular tubular dentin.

Reparative dentin formation: 1. Absent – No evidence of reparative dentin formation. **2. Incomplete** – Reparative dentin formation at some distance form the capping agent. Calcific matrix did not occlude the exposures. **3. Complete** – Reparative dentin formation directly against the capping agent. Exposure site was completely occluded by the calcific matrix.

Bridging of the exposure site under zinc oxide eugenol is inhibited. This is due to that Eugenol, a phenol derivative is known to be toxic and it is capable of producing thrombosis of blood vessels when applied directly to pulp tissue.[18]

In group 1, only one tooth from 30 days experienced mild pain. None of the remaining teeth showed adverse signs or symptoms. Histologically after 7 days all teeth showed mild inflammation. After 30 days odontoblastic proliferation and incomplete dentin formation was seen. After 90 days odontoblastic proliferation and complete dentin bridge formation was seen in almost all cases. Hence, Calcium hydroxide clinically as well as histologically appears to be acceptable.

In group 2, only one tooth each from 30 and 90 days experienced mild pain. Histologically 3 teeth showed mild inflammation after 7 days. Mild to moderate fibrosis was seen in almost all cases. After 90 days 6 teeth showed incomplete dentin formation, widened predentin layer with odontoblastic proliferation which suggest that pulp would heal with resinifying agent at longer duration period but more research is needed.

In group 3, only one tooth each from 30 & 90 days complained about mild pain and sensitivity to thermal stimulation. Although zinc oxide eugenol cement clinically appears to be acceptable, but histologically almost in all cases, chronic inflammation with no evidence of calcific bridging were seen. Hence, histologically Zinc oxide eugenol cement is unsuccessful as pulp capping agent.

CONCLUSION

Calcium hydroxide can be used successfully as direct pulp capping agent. Resorcinol may be used as direct pulp capping agent but long term study should be undertaken

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