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# The comparison of two dental techniques used to treat upper front adult teeth which have become infected or painful following accidental damage

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04/09/2012

**Recruitment status**
No longer recruiting

**Registration date**
16/10/2012

**Overall study status**
Completed

**Last Edited**
15/10/2020

**Condition category**
Oral Health

[_] Prospectively registered
[_] Protocol
[_] Statistical analysis plan
[_] Results
[_] Individual participant data
[_] Record updated in last year

## Plain English summary of protocol

Background and study aims
Children often damage their front teeth. In approximately 6% of cases, the nerve inside the affected tooth dies (becomes non-vital) and natural root development stops. In these cases, the tooth requires a root canal treatment in order to prevent problems such as pain and dental abscesses from arising. However, because the roots of these young teeth are not fully formed, they are weaker and prone to fracture. In addition, root canal treatment is difficult because a root canal filling cannot be placed in a tooth which is not yet fully formed, due to the fact that the root has an open end. To enable root canal treatment to be carried out, a barrier must be placed at the end of the open root. This can be done using materials called Calcium Hydroxide or Mineral Trioxide Aggregate (MTA). These materials are placed inside the root and sealed into the tooth. However, although they help to provide a barrier, they do not help to strengthen the walls of the root. Treatment with these materials requires multiple visits to the dentist, over a period of up to 18 months. There is evidence to suggest that an alternative treatment involving revascularisation (recovery of the blood supply to the tooth) and the use of a triple antibiotic paste allows natural root growth to restart, and also strengthens the walls of the root. Treatment can often be carried out in just two visits. The aim of this study is to discover whether there is a difference between one of two methods of treating non-vital teeth with open ends.

Who can participate?
All children presenting to Liverpool University Dental Hospital aged between 7 and 25 years of age and who are medically well and happy to receive prolonged treatment in the dental chair are eligible to take part in the study if they have a damaged upper front adult tooth in which the nerve has died and the root is open ended. Unfortunately, if there is dental decay in the tooth or a fracture of the root then these teeth are not suitable for this study. In some cases following damage to a tooth, the root of the tooth starts to dissolve and unfortunately these teeth are also not suitable for this study.

What does the study involve?
Children with teeth that fall into this category and require root canal treatment will be given one of two treatments, both of which aim to treat infection, close the root end and to allow healing to take place.
Teeth will receive one of the following methods of root treatment:
1. Revascularisation (recovery of the natural blood supply to the tooth) following placement of an antibiotic paste into the tooth root. The aim of this treatment is to allow natural root growth to restart. Root growth will allow the tooth to form at barrier at the end of the root. No root canal filling will then be necessary.
2. Closure of the open root end by placement of an artificial barrier at the end of the root so that a root canal filling can then be placed. This will be done with a dental material called Mineral Trioxide Aggregate (MTA). Non-vital teeth with an open end are routinely treated in this way at Liverpool Dental Hospital. Participants will be randomly allocated in to one of the above treatment groups. This means that neither the participants, nor the researchers, will be able to choose which group a participant will be allocated to. In both techniques, in order to learn about the bacteria involved in non-vital teeth, we will take samples of the bacteria within the root canal.

What are the possible benefits and risks of participating?
The outcomes of the study will provide us with further information about root growth, the bacteria involved in infection of non-vital teeth and the success of the different treatment methods that are available. This information will enable us to increase our understanding of the treatment of non-vital teeth with an open end and help us to explain our treatments to future patients. There are no risks or disadvantages to taking part in this study. However, if the tooth does not respond to treatment, or if symptoms of infection arise, then alternative treatment methods may be initiated as necessary.

Where is the study run from?
The study is run from Paediatric Dentistry Clinic at Liverpool University Dental Hospital.

When is the study starting and how long is it expected to run for?
October 2010 to October 2015

Who is funding the study?
The study is being funded by the Royal Liverpool and Broadgreen University Hospitals NHS Trust and by the University of Liverpool (UK)

Who is the main contact?
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# Contact information

### Type(s)
Scientific

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# Additional identifiers

### ClinicalTrials.gov (NCT)
NCT01817413

### Protocol serial number
R&D 3968, UoL000590

# Study information

### Scientific Title
Revascularisation versus mineral trioxide aggregate in the Management of non-vital immature permanent incisors in a young population: a randomised controlled trial (pilot study)

### Study objectives
Research Question:
Is pulp revascularisation superior to Mineral Trioxide Aggregate (MTA) in achieving root end closure in the management of non-vital immature permanent incisors?

Null Hypothesis:
The null hypothesis is that there will be no difference in the success rate of revascularisation versus MTA in achieving root end closure in the management of non-vital immature permanent incisors.

Background
The Endodontic Management of Non-Vital Immature Incisors
The most recent United Kingdom survey of Child Dental Health (2003) reported the prevalence of accidental damage to permanent incisors in children as 5% at age 8 years, increasing to 11% at age 12. Around 6% of these teeth are expected to become non-vital and thus require root canal treatment.

The endodontic management of non-vital immature incisors in young children can be challenging. If accidental damage or caries renders immature permanent teeth non-vital, then root development is arrested. Such teeth have an open, and often divergent, apical morphology. They also have thin dentinal walls, which are prone to fracture. An open apex complicates root canal treatment, as a root end closure technique must be carried out that aims to form a barrier at the apex, in order that gutta percha can then be condensed without extruding excess filling material though the apex into the periapical tissues.

Current treatment protocols involve preserving non-vital anterior teeth in young patients; at least until alternative definitive treatment options can be considered. More definitive treatment options such as placement of fixed prostheses or implant-supported crowns are not suitable

until craniofacial growth is complete. Anterior teeth that are aesthetically poor may have detrimental effects on nutrition and social development.

## Apexification with Calcium Hydroxide

Traditionally, treatment for non-vital immature teeth involves creating a calcified barrier using Calcium Hydroxide [Ca(OH) 2]. Calcium hydroxide is placed into the root canal at three-monthly intervals in order to promote formation of a hard apical barrier at the open apex. This technique has a number of disadvantages including prolonged treatment time of up to 18 months, multiple treatment visits, and variable outcomes. Treatment with calcium hydroxide does not increase the thickness of the immature dentinal walls. Long-term dressings with calcium hydroxide have a significant negative effect on the strength of the root2. These factors render teeth treated with calcium hydroxide more likely to suffer a cervical root fracture during, or following, root canal treatment.

## Use of Mineral Trioxide Aggregate as an Apical Plug

Mineral Trioxide Aggregate (MTA) has been recommended as an alternative to prolonged treatment with calcium hydroxide. An apical plug of MTA can be placed at the apical portion of an immature root in order to provide a barrier against which conventional root canal treatment can be carried out.

The main clinical advantage of MTA is a reduction in the duration of the calcium hydroxide dressing and overall treatment time, as an immediate artificial barrier is created. MTA is a biocompatible material that has a good sealing ability and evidence antibacterial activity. Observational studies of MTA apexification have demonstrated comparable healing outcome to calcium hydroxide apexification. However, although the open apex might become closed, this does not promote the continued development of the immature root, which has thin dentinal walls that render it prone to fracture. Post-endodontic restorations may have great significance in preventing root fracture in these thin-walled immature anterior teeth. In vitro studies demonstrate that composite resin restorations following endodontic treatment with extension of the composite resin into the root canal can substantially enhance the strength of MTA-filled immature teeth6.

Drawbacks of MTA include the potential for tooth discolouration, presence of toxic elements in the material composition, long setting time, high material cost and the absence of a known solvent to aid its removal.

## Revascularisation of Pulp Tissue Technique

Lately, the concept of revascularisation of necrotic pulps has been reported as an alternative conservative treatment option for non-vital permanent teeth with immature roots. Previously, the regenerative potential of pulp has been considered to be extremely limited. However, our improved understanding of pulpal inflammation and repair and the availability of improved dental materials have lead to revascularisation becoming a viable alternative to root canal treatment. The potential to regenerate an injured or necrotic pulp would provide the opportunity for the best root filling possible, as by definition, the presence of a vital non-infected pulp prevents apical periodontitis.

It has been shown that revascularisation can be achieved in immature teeth that have been traumatically avulsed and therefore contain a necrotic but uninfected pulp. Skoglund et al demonstrated that in extracted dog teeth, pulpal revascularisation began immediately following reimplantation and was complete at approximately 45 days. The ischaemically necrotic pulp tissue in an avulsion injury acts as a scaffold for the in-growth of new tissue. The usually intact crown in these cases slows bacterial penetration to the pulp.

Revascularisation of the pulp in a necrotic and infected tooth with apical periodontitis was attempted in the 1960s but was mostly unsuccessful. This is most likely because the materials and instruments available at this time were probably not sufficient to create the avulsion-like environment that is necessary for revascularisation to be successful. This environment includes a root canal that is free from the presence of bacteria, which contains a scaffold for the in-growth of new tissue and which is resistant to further bacterial penetration (has a good coronal seal).

Several more recent case reports have reported success following revascularisation of necrotic and infected pulps in immature teeth when a triple antibiotic paste is used. The antibacterial effectiveness of the triple antibiotic paste was initially demonstrated in 1996 and has since been confirmed in an animal study. It has been suggested that the presence of a blood clot within the disinfected canal is essential in acting as a scaffold. It is not yet known whether the necessary factors present in the blood clot can be isolated and perhaps then incorporated into a synthetic scaffold for use in the future.
There is some discussion in the literature about the nature of the new tissue which revascularises the pulp space. It is possible that the tissue is more similar to periodontal ligament than pulp tissue. A recent evaluation of the histological characterisation of tissue present in the pulp space following revascularisation has suggested that the correct term for the procedure may be revitalisation rather than revascularisation. This is based on the finding that although pulp tissue may survive the infection and recover, the procedure of revascularisation allows in-growth of vital tissue consisting of tissues resembling cementum, PDL and bone, but not pulp parenchymal tissue. These tissues do not function like a pulp tissue and therefore, the authors conclude that revitalisation is not tissue regeneration but wound repair. Future research will need to be performed to stimulate pulp regeneration from the pluripotential cells in the periapical region.

Revascularisation aims to promote continued root development, including thickening of dentinal walls. The technique also aims to deliver an improvement in the long-term prognosis and outcomes of the tooth compared to that achieved with either calcium hydroxide or with an apical plug of MTA.

The Presence and Pathogenesis of the Bacteria Involved in Infected Immature Teeth Treated with a Triple Antibiotic Paste

The presence of pathogenic bacteria within the root canal is of interest to the endodontist. Administration of a triple antibiotic paste has been found to be effective in successful cases of revascularisation, yet there are no reports in the literature of microbiological sampling which has taken place during this novel treatment technique in order to determine the pathogens present prior to, or following, use of a triple antibiotic paste.

According to our present knowledge, apical periodontitis is mainly associated with Fusobacterium nucleatum, Streptococcus spp. Prevotella intermedia, Peptostreptococcus micros, Peptostreptococcus anaerobius or Eubacterium lentum.

The effectiveness of antimicrobial measures is often monitored in clinical research studies by microbiological root canal sampling (MRS). This is often also used as a predictor for healing. The extent to which canal sampling techniques accurately reflect the bacterial status of the canal space must be taken into account, as false positive and false negative cultures may adversely affect the performance of MRS . MRS is a passive sample of the main root canal space, which does not include inaccessible areas such as accessory canals. MRS is analysed via anaerobic sampling and cultivation techniques, which may be unable to reproduce the growth conditions

required by fastidious bacteria. It is estimated that between 50% of oral bacteria are uncultivable . Among the cultivable, many bacterial species are difficult to grow and differentiate between following culturing.

Recent advances in molecular techniques based on direct amplification of genes extracted from bacteria in clinical samples, followed by cloning and sequencing of the genes, have allowed bacteria to be characterised without the biases of culture. These molecular-based identification methods are designed to detect microbial DNA rather than living microorganisms. The 16S rDNA directed polymerase chain reaction (PCR) provides an alternative method of detecting and differentiating between bacteria in necrotic root canal samples . The 16S rDNA gene is well-known to be useful for the taxonomy of bacteria . The use of this technique has shown that the microflora associated with endodontic infections is far more diverse than has been previously shown by cultural studies alone. A limitation of PCR is that it cannot distinguish between DNA from viable or dead cells, and therefore it is unclear whether this method represents the living endodontic flora or whether it actually provides a historical record of organisms which have been previously present within the canal.

There are no previously reported randomised control trials comparing the use of pulp revascularisation versus MTA in achieving root end closure in the management of non-vital immature permanent incisors.

Aims
The aims of this pilot study are to develop the optimum study design, sample size and outcome measures required to compare the success rate of revascularisation versus MTA in the management of non-vital immature maxillary central incisors in a young population.

Specific Objectives
1. Compare the number of visits and time taken to complete treatment using revascularisation in comparison to MTA.
2. Obtain pilot study data to allow sample size calculations for a future randomised-controlled clinical trial.
3. Compare the clinical outcomes for revascularisation and for MTA.
4. Compare the microbiological outcome for revascularisation and for MTA.
5. Compare the microbiological outcome of treatment with the clinical outcome of treatment and the extent to which positive or negative cultures at the time of obturation adequately predict treatment outcome.
6. Determine the diversity of bacteria associated with endodontic infections by means of a combined cultural and molecular approach.

## Ethics approval required
Old ethics approval format

## Ethics approval(s)
NHS National Research Ethics Service, August 2010, ref: REC10/H1014/50

## Study design
Randomised single-centre trial

## Primary study design
Interventional

## Study type(s)
Treatment

## Health condition(s) or problem(s) studied
Non-vital immature permanent central incisors (upper front adult teeth)

## Interventions
Participants are randomised into 2 groups via computer generated random allocation. Allocation concealment by sealed serially numbered opaque envelopes.

Pre-operative Assessment
1. All teeth will be subject to a pre-operative radiograph using a paralleling technique.
2. Following confirmation of the inclusion criteria, an information leaflet will be given to patients and their parents and they will be invited to participate in the study.
3. Informed written participant assent and parental consent will be obtained before randomisation.
4. Patients will be randomised into two groups using computer generated random allocation.
5. Allocation concealment will be by sealed serially numbered opaque envelopes and treatment allocation will be determined by opening the next envelope in the sequence.
6. An appropriate treatment appointment will then be arranged.

Participants in both treatment groups will undergo dental treatment in two visits which are two weeks apart. Participants in both treatment groups will be required to attend for four reviews in the subsequent 12 month period.

Further detail of the study method follows below:

Clinical Technique - Group 1 (Revascularisation)

First Visit
1. Local anaesthetic without vasoconstrictor will be administered via a labial infiltration technique.
2. The tooth will be isolated with rubber dam.
3. A conventional access cavity will be prepared.
4. Working length determination will be carried out using an apex locator.
5. Length determination will then be confirmed with a radiograph.
6. The tooth will be irrigated carefully with 5% sodium hypochlorite, followed by 17% EDTA for one minute, followed by a final irrigation of 2.0% chlorhexidine gluconate.
7. The root canal will be dried using absorbent paper points.
8. A 20G needle will be set to 2mm short of the working length and used to introduce the triple antibiotic paste (Ciprofloxacin 500mg, Metronidazole 500mg, Minocycline 500mg). This will be mixed by the hospitals pharmacy on the day of treatment. It will be placed into the canal using a backfill approach up to the level of the cemento-enamel junction (CEJ).
9. The tooth will then be temporarily sealed with a cotton pellet and Intermediate Restorative Material (IRM, Denstply).

Second Visit
1. Local anaesthetic without vasoconstrictor will be administered followed by isolation of the tooth with rubber dam.
2. Following access to the root canal, an endodontic file will be taken and a rubber stop will be placed 2mm beyond the working length. The file will then be pushed past the confines of the

canal into the periapical tissue in order to induce bleeding.
3. When blood is seen to reach the level of the CEJ, haemostasis will be achieved with a cotton wool pellet at a depth of 3-4mm into the canal so that a blood clot can form and provide a scaffold for the in growth of new tissue.
4. MTA will be placed in the cervical portion of the root canal in order to provide a seal.
5. A post-operative periapical radiograph will be taken.
6. The tooth will then be restored with a bonded resin coronal restoration.

Follow Up
1. Treated teeth will be reviewed clinically and radiographically on a 3 monthly basis for 12 months.
2. Two examiners will review all radiographs in which failure is suspected in order to ensure that there is agreement. Inter- and intra-examiner agreement will be assessed before the study begins and throughout its duration.
3. If signs or symptoms that indicate failure of treatment arise, then the patient will be informed and conventional treatment will be provided.

Clinical Technique - Group 2 (MTA)

First Visit
1. Local anaesthetic with vasoconstrictor will be administered (as per normal practice for treatment with MTA).
2. The tooth will be isolated with rubber dam.
3. A conventional access cavity will be prepared.
4. Working length determination will be carried out using an apex locator.
5. Length determination will then be confirmed with a radiograph.
6. The tooth will be irrigated carefully with 5% sodium hypochlorite, followed by 17% EDTA for one minute, followed by a final irrigation of 2.0% chlorhexidine gluconate.
7. The root canal will be dried using absorbent paper points.
8. Ca(OH)2 will be introduced into the canal using the dispenser provided by the manufacturer. Ca(OH)2 will be placed from the working length up to the CEJ.
9. The tooth will then be temporarily sealed with a cotton pellet and Intermediate Restorative Material (IRM, Denstply).

Second Visit
1. Local anaesthetic with vasoconstrictor will be administered followed by isolation of the tooth with rubber dam.
2. Following access to the root canal, a 5mm MTA apical plug will be placed at the apex with an appropriately sized endodontic plugger, via the aid of the endodontic operating microscope.
3. Radiographic examination of the apical plug will be carried out followed by any necessary adjustments.
4.. Filling of the whole canal will be completed using thermoplastic gutta-percha and a resin-based sealer.
5. A periapical radiograph will be taken to confirm the quality of the obturation.
6. The tooth will then be restored with a bonded resin coronal restoration.

Follow Up
1. Treated teeth will be reviewed clinically and radiographically on a 3 monthly basis for 12 months.
2. Two examiners will review all radiographs in which failure is suspected in order to ensure that there is agreement. Inter- and intra-examiner agreement will be assessed before the study begins and throughout its duration.

3. If signs or symptoms that indicate failure of treatment arise, then the patient will be informed and alternative treatment options will be discussed.

Microbiological Sampling
Sampling of the organisms present within the root canal will be carried out at the following three stages:

Group 1 (Revascularisation)

At the first visit:
1.Immediately following access to the root canal
2.Following irrigation and drying of the canal, prior to administration of the triple antibiotic paste

At the second visit:
Following removal of the triple antibiotic paste, prior to initiating bleeding into the root canal

Group 2 (MTA)

At the first visit:
1.Immediately following access to the root canal
2.Following irrigation and drying of the canal, prior to administration of Ca(OH)2

At the second visit:
3.Following removal of Ca(OH)2,prior to obturation of the root canal

Sampling Technique

Sterile nuclease free water will be introduced into the root canal and will be aspirated after gentle irrigation. Samples will also be taken using sterile paper points that will be transported immediately to the microbiology laboratory inside an anaerobic workstation for culture dependant or independent analyses.

Culture Dependant Analysis
The fastidious anaerobic agar (FAA) and blood agar plates will be prepared by dissolving recommended amount of the media powder into distilled water. This solution was then autoclaved at 1210C (15p.s.i) for 15 minutes. The molten agar was then cooled to 470C in a water bath. At 470C the molten agar will be supplemented with 5% horse blood and mixed by swirling. Twenty five mL of molten agar was poured into sterile Petri dishes (Sterilin, UK) to ensure a depth of 4mm ±0.5mm. The agar plates were then left in an air flow chamber to solidify and dry. Once dry the plates will be stored at 4-80C in sealed plastic bags. Plates will be removed from storage 1 hour prior to usage to ensure they were dry.

Dilutions of the samples for culture dependant analysis will be performed immediately in reduced transport medium and will be plated onto FAA and blood agar medium in an anaerobic cabinet. A set of samples will be incubated in air+5% $CO_2$. Selected bacterial colonies will be identified using biochemical methods and the 16S rRNA gene amplification. The results will be compared with bacteria identified using purely on 16S rRNA gene amplification analysis

Culture Independent Analysis

16S rRNA gene amplification and DGGE

Total DNA will be isolated from both culture dependant and independent samples using MasterPure™ DNA Purification Kit and the 16s bacterial rRNA gene will be amplified by PCR (5 min at 95°C followed by 40 cycles of 1 min at 95°C, 1 min at 64°C and 1.5 min at 72°C followed by a 15 min extension step at 72°C). Using the universal primers (27F, 5-AGAGTTTGATCMTGGCTCAG-3 and 1525R, 5-AAGGAGGTGATCCAGCC-3) DNA isolated from samples and their 16S rRNA gene amplicons will be assessed by agarose gel electrophoresis. PCR product will then be analysed using Denaturing gel gradient electrophoresis (DGEE). Specific regions of the 16s rRNA gene locus that are amplified by PCR, will be run on a denaturing gel that separates amplicons according to nucleotide composition. Different amplicons are then displayed as bands with different migration distances to yield a distinctive fingerprint representing each of the various bacterial species. DNA isolated from standard isolates such as Enterococcus faecalis, Porphyromonas gingivalis ATCC33277 and Streptococcus gordonii DL1 will be used as controls for the molecular identification of bacteria from samples. Specific bands of interest will be excised from gels and their 16s rRNA fragments can then be sequenced and compared with known sequences in an rRNA database.

### Intervention Type
Procedure/Surgery

### Primary outcome(s)
1. A satisfactorily restored tooth
2. An asymptomatic tooth
3. Absence of signs of failure of treatment (e.g. sinus, swelling, mobility)
4. Evidence of periapical healing
5. Absence of root resorption
6. Patient satisfaction

### Key secondary outcome(s))
1. Development and maturation of the root following pulp revascularisation
2. Reduction in the size of the periapical radiolucency between pre- and post-treatment radiographs
3. In the case of pulp revascularisation - a positive response to vitality testing

### Completion date
01/10/2015

## Eligibility

### Key inclusion criteria
1. Between 7 and 25 years of age
2. Have no significant medical history
3. Cooperative in the dental chair
4. Able to commit to the recall schedules prescribed by the study
5. Have one or more traumatised non-vital permanent maxillary central incisors with incomplete root development

### Participant type(s)
Patient

### Healthy volunteers allowed
No

### Age group
Other

### Sex
All

### Key exclusion criteria
1. Have a medical history that may complicate treatment
2. Have a medical history for which the study procedures may place the patient at increased risk
3. Have a diagnosis of avulsion or severe intrusion following dental trauma
4. Permanent maxillary central incisors for treatment must not:
4.1. Be less than half formed
4.2. Have anatomical complexity (such as dens invaginatus)
4.3. Have horizontal or vertical root fractures present
4.4. Have evidence of root resorption

### Date of first enrolment
01/10/2010

### Date of final enrolment
01/10/2015

## Locations

### Countries of recruitment
United Kingdom

England

### Study participating centre
**University of Liverpool**
Liverpool
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L3 5PS

## Sponsor information

### Organisation
The Royal Liverpool and Broadgreen University Hospitals NHS Trust (UK)

### ROR
https://ror.org/009sa0g06

# Funder(s)

**Funder type**
University/education

**Funder Name**
University Of Liverpool

**Alternative Name(s)**
The University of Liverpool, , Universidad de Liverpool, UoL

**Funding Body Type**
Government organisation

**Funding Body Subtype**
Universities (academic only)

**Location**
United Kingdom

# Results and Publications

**Individual participant data (IPD) sharing plan**

**IPD sharing plan summary**
Not provided at time of registration