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Storage Media for An Avulsed Tooth: Nature to the Rescue

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ABSTRACT

Tooth avulsion or exarticulation is a traumatic injury of dental tissue characterised by complete displacement of the tooth out of its socket. Successful treatment outcome of such an injury is dependent on the survival of the viable periodontal ligament cells attached to the tooth root surface. The viability of the periodontal ligament cells is best preserved either when the tooth is immediately replanted into its socket or if it is stored in an appropriate storage /transport medium till a time, the tooth can be replanted into its socket. A number of storage /transport medium are available both of natural origin and laboratory prepared solutions. This review discusses various storage/ transport media for avulsed tooth obtained from natural sources. The knowledge of availability and accessibility of such natural storage media would increase the awareness among the healthcare providers and would therefore improve the prognosis of the treatment protocol by reducing extra-oral dry time of the avulsed tooth.

Keywords: Trauma, Storage media, Transport media, Avulsion

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INTRODUCTION

Tooth avulsion or exarticulation is a traumatic injury of dental tissue characterised by complete displacement of the tooth out of its socket. This injury is comparable to losing one's organ in an accident, such as a finger and therefore has severe psychological implications¹. Children of age group of 7 to 9 years are more likely to suffer these injuries particularly involving avulsion of single maxillary incisors (0.5%–16% of all traumatic injuries of permanent anterior teeth).¹ Ideal management of tooth avulsion is prompt treatment of socket and tooth, with reinsertion of the tooth into its socket. Reinsertion of tooth back within 5 minutes has the best outcome in which there is least loss of viability of periodontal ligament cells and hence reestablishment of periodontal fibres occurs, regaining the functionality of tooth.² However, this may not always be possible owing to the various associated factors such as the person's conscious state, lack of first aid knowledge, informed consent issues and lack of confidence in strangers gathered at the scene of accident.³ Clinically a variety of factors such as age of the individual, width and length of the root canal, stage of root development, mechanical damage during trauma and replantation, type of splinting, mastication, treatment of the socket, endodontic treatment, antibiotics, time of replantation, macroscopic contamination, storage media and storage period are important and can influence the clinical success of replantation⁴. When a tooth is completely displaced out of the socket, the PDL ruptures with some part attached to the alveolar bone and some part attached to the tooth root. When the tooth is immediately replanted the healing responses starts. The viable fibroblasts are critical for repair as they account for almost a quarter of the PDL cells. During healing there is an increase in the formation and differentiation of fibroblasts.⁵ The progenitor cells responsible for producing fibroblast phenotypes are located adjacent to vascular channels of PDL cells and alveolar bone.⁶ These cells are closely related to the preservation of a viable mixed population of PDL cells that adhere to the roots of avulsed teeth. Therefore it is essential to maintain the viability of the PDL cells and its progenitor cells attached to the roots in an avulsed tooth. This is accomplished by soaking the avulsed tooth in an appropriate storage media or transport media. A storage medium may be defined as a physiological solution that closely replicates the oral environment to help preserve the viability of PDL cells following avulsion⁷. The ideal requirements for a storage medium are⁸

- It should have antimicrobial characteristics
- It should maintain the viability of periodontal fibres for an acceptable period of time
- It should favour proliferative capacity of the cells (clonogenic and mitogenic capacity)

- It should have the same osmolarity as that of body fluids (290-300 mosmol/ kg) and pH balanced (7.2 – 7.4)
- It should be unreactive with body fluids
- It should not produce any antigen-antibody reactions
- It should reduce the risk of post-replantation root resorption or ankylosis
- It should have a good shelf life
- It should be effective in different climates and under different conditions
- It should wash off extraneous materials and toxic waste products
- It should aid in reconstitution of depleted cellular metabolites.

Use of such a storage media has been associated with favourable healing outcomes.⁹ A number of solutions have been tested and tried to meet the ideal requirements of storage media, but none satisfy all the requirements. The storage media can be classified as Laboratory prepared and Natural source. (Table 1)

Table 1: Classification of Storage/ Transport media for avulsed tooth

Laboratory prepared	Natural source
Hank's Balanced Salt Solution	Milk
Normal saline	Saliva
ViaSpan	Propolis
Eagle's medium	Coconut water
Custodiol	Egg white
Dubelco's storage	Emdogain
Tooth rescue box	Morusrubra
Conditioned medium	Salvia officinalis extract
Gatorade	Honey milk
Contact lens solution	Tap water
Growth factors	
Ascorbic acid	
L-DOPA	
Cryoprotective agents	
Catalase supplementation	

Hanks' Balanced Salt Solution (HBSS) was introduced by John H Hanks in 1975 as a solution for preservation of tissue culture¹⁰. Among all the storage medium HBSS is considered as the gold standard and is often used as a comparison reference medium to deduce the clinical efficacy of other media. The American Academy of Endodontics has accepted HBSS as an acceptable medium for avulsed teeth because of its capability to maintain vitality and proliferative capacity of PDL for an extended period of time (up to 48 hours)¹¹. It contains the sodium chloride, D-glucose, potassium chloride, sodium bicarbonate, potassium phosphate (monobasic), calcium chloride and magnesium sulphate anhydrous. It can preserve cells and tissues for 24 h and both the pH (7.4) and the osmolarity (280 mosmol

kg-1) are ideal¹². It can maintain the viability of PDL cells for several hours with a success rate of 90% reported when degenerated PDL cells were stored in HBSS for less than 30 min.¹³ The vitality, clonogenic and mitogenic capacity of PDL cells using this medium are excellent. HBSS is the only medium that can replenish metabolites in depleted PDL cells.¹¹ It has been reported that when an avulsed tooth that has been kept dry for 15–60 min is replanted after being soaked in HBSS for 30 min, less root resorption occurred.¹⁴ It is recommended that avulsed teeth should be soaked in HBSS for 30 min before replantation even if they have been stored in a suitable physiologic medium because of the replenishing ability of HBSS.¹⁵ HBSS does not need to be refrigerated. Commercially available form of Hanks' Balanced Salt Solution (HBSS) is marketed as Save-A-Tooth (Save-A-Tooth; Phoenix Lazerus Inc., Pottstown, PA, USA), to maintain periodontal ligament cell viability¹⁶. It is a special kit available in some countries, which has been designed for the public to use for the emergency management of avulsed teeth. This kit contains a small basket in which the avulsed tooth is suspended and submerged in HBSS. Gentle agitation can remove debris from the PDL during storage and lost nutrients can be replenished by the HBSS before replantation. Unfortunately, HBSS is not widely used in India, because it is not readily available. Hence, an alternative medium that is not only comparable in performance to HBSS, but is also readily available and inexpensive is required.

Milk

Milk as a storage medium is the most practical transport medium for the storage of avulsed teeth because of its ready availability in almost all situations. Milk, which contains amino acids and vitamins, is capable of inactivating enzymes harmful to the PDL cells.¹⁷ Milk is a compatible short-term storage medium for teeth if they were placed in it within 15 to 20 min of being avulsed¹¹. Milk has a pH of 6.5 to 7.2 and osmolarity of 270 mosmol kg-1, which is similar to extracellular fluid. Milk can potentially maintain PDL cell viability for up to 2 hours¹⁸. The clonogenic capacity can be maintained at the same level for an additional 45 min by keeping the milk chilled with an ice pack or in the refrigerator. Milk maintains the viability of PDL cells at a clinically significant level for up to 1 h of extra-oral time.¹⁹ Milk can usually be obtained on short notice. The disadvantages are that milk needs to be fresh and kept refrigerated, it does not replace depleted cell metabolites, and it does not facilitate cell mitosis. It prevents cell death, but does not restore the cells' normal morphology and ability to differentiate and undergo mitosis²⁰. At a cellular level, milk is ranked equal to HBSS as a storage medium although it loses its effectiveness after 2 h.¹⁹ Milk is superior to saliva with regard to the number of viable cells and the ability of the cells to recover and heal wounds. Powdered milk is one of the presentation forms of bovine milk and is considered as a feasible

medium in cases of delayed tooth replantation. It has shown similar results to long shelf-life whole milk in relation to the healing process after delayed replantation of avulsed teeth.²¹ However, the powdered form is more effective than pasteurised milk as a medium only up to four hours, following which these substitutes perform worse than whole milk.²²

Saliva

Saliva can also be used as an immediate interim storage medium. Though very readily available, avulsed teeth should not be stored for longer than 30 min in saliva. Saliva contains potentially harmful substances, such as enzymes, bacteria and their by-products.¹² Patients' own saliva is the best immediate transport medium for an avulsed tooth.²³ It is also an immediately available storage medium at all the accident sites. After trauma, several ml of saliva can easily be collected in a cup and the tooth dropped into this, or the tooth can be placed in the patient's mouth under the tongue. In an animal study, Andreasen showed that saline and saliva were suitable storage medium for protection against root resorption for short extra-alveolar periods.²⁴ Thus saliva can be considered to be an acceptable short-term storage medium (less than 30 min) and its use should be limited to cases where the extra-alveolar duration is less and other superior storage media are not available.

Propolis

Propolis is a natural product used by bees in the construction and maintenance of their hives. It has anti-inflammatory, antibacterial, anti-oxidant, anti-fungal and has tissue regenerative actions. It can inhibit the late stages of osteoclast maturation so it may be useful as an intra-canal medicament to reduce resorption of traumatised teeth²⁵. A recent study showed that propolis could be used for a vulsed teeth and that a 6-h period of storage was more appropriate than 60 min of storage²⁶. As with most substances for natural therapies, propolis has no standard recommended weight per volume of solution. The major disadvantage of propolis is that it is not readily available to the public and therefore of little value as a storage medium for avulsed teeth.

Coconut water

Biologically pure tender coconut water, which aids in replenishing the fluids, electrolytes and sugars lost from the body during heavy physical work, has been suggested as a promising storage medium for avulsed teeth²⁷. It was observed that coconut water was superior to HBSS, milk or propolis in maintaining the viability of PDL cells²⁸. Coconut water was found to have a pH of 4.1 which is deleterious to cell metabolism. Coconut water by itself is less effective than coconut water buffered with sodium bicarbonate.²⁹

Egg white

Egg white has a pH of 8.6–9.3 and its osmolarity is 258 mosmol/ kg. Egg white was found to

be more suitable storage media because there was no significant difference between egg white and milk at storage times of 1, 2, 4, 8 and 12 h in cell viability.¹² It has shown better and significantly higher incidence of PDL healing as compared to milk and equivalent cell viability as HBSS.³⁰ Egg albumin is considered as a good choice because of its high protein content, vitamins, water, lack of microbial contamination and easy accessibility. Egg white as a storage medium suffers the setback of impracticality and among those with food preference.

Emdogain

Emdogain (Biora, Malmo, Sweden) is a commercial Enamel Matrix Derivative (EMD) extracted from developing embryonic enamel of porcine origin and contains several matrix proteins³¹. It can influence the migration, attachment, proliferative capacity and biosynthetic activity of PDL cells³². It has also been used in anti-resorptive-regenerative therapy along with topical glucocorticoids and systemic doxycycline. Use of emdogain has been shown to increase the incidence of healed PDL when this gel was applied to root surface of the avulsed tooth and /or inserted directly into alveolar socket before implantation. It appeared to aid in preventing or retarding root resorption and ankylosis¹. Thus, it is a recommended therapeutic agent for the management of avulsed permanent teeth. However, no firm conclusion regarding the efficacy of EMD application on healing of replanted³³ and auto transplanted permanent teeth can be drawn because of a lack of randomised controlled trials and clinical controlled trials.³⁴

Morus rubra

Juice of the leaves of fruit of *Morus rubra* (red mulberry) has been recommended as a suitable transport medium for avulsed teeth. They are rich in flavonoids, alkaloids and polysaccharides components which are the most potent active constituents.³⁵ At 4% concentration, *M. rubra* was found to be more effective than HBSS up to 12 hours, in maintaining the PDL cells' viability.³⁶

Salvia officinalis extract

Salvia officinalis belongs to the family of Labiatae and is native to the Mediterranean region. Essential oil obtained from it has some antimicrobial and antioxidant properties. The major constituents are α - and β -thujone (>50%) and camphor (<20%)³⁷. Other components include manool, ledene, viridiflorol, 1-8 cineole, limonen, and trans-caryophyllene. The oxygenated component represents more than 80% in the essential oils. PDL cells' viability at 1-3 hours was found similar for 2.5% *S. officinalis* and HBSS, whereas at 24 hours, the efficacy of 2.5% *S. officinalis* is significantly better than HBSS³⁸. Thus, *S. officinalis* can also be recommended as a suitable transport medium for avulsed teeth.

Honey milk³⁹

Recently long-shelf life honey milk has been studied as a storage medium. Honey milk has 8% non-fat solid milk, 3gr protein, 11gr carbohydrate, 0.1gr calcium, 0.6gr minerals and 0.12gr phosphorous and natural honey (5%). This product has extended storage capability of at least 6 months without the need for refrigeration. Long-shelf life honey milk may be considered as appropriate storage media which are comparable to HBSS and better than fresh milk medium.³⁹

CONCLUSION

Currently there is no national data on incidence of avulsion injuries in the Indian population. This may be one of the reasons for no demand of Save A Tooth kit and hence high cost if at all available. Therefore in such situations the best storage media would be natural products with easy accessibility to general public and dentists as well. Chilled low fat/skimmed milk is the best available storage medium. Newer research is still required to find that easily available, cheap and low maintenance storage medium which can maintain the viability of PDL cell and regenerate them as well.

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