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Review Article

Tooth Saving Solutions- Best Transport Media for Avulsed Tooth? A Review

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Abstract:

Avulsion is considered one of the most serious types of trauma to teeth as it can displace the tooth from its bone socket and also causes injury to its surrounding structures. The neurovascular supply is greatly hampered by the complexity of this damage, which typically leads to pulp vitality loss. The treatment for avulsed tooth is immediate replantation but, in several situations, it is not possible. The maintenance of viability of PDL cells on the root surface of avulsed tooth is absolutely necessary till the tooth is placed back in the socket to prevent ankylosis and replacement resorption.

Research has led to the development of various transport media for avulsed teeth that can produce conditions that closely resembles the environment of alveolar socket with adequate osmolality, pH, nutrition & metabolites.

This review sheds light on various transport media used for avulsed teeth, its ideal requirements and newer research on such solutions.

Keywords: Transport media, Avulsion, Avulsed tooth

INTRODUCTION

Clinical surveys indicate that traumatic dental injuries in children and adolescents are a common problem. Studies have shown that the prevalence of these injuries are increasing. Avulsion injury, one of the most severe forms of dental trauma, is characterized by complete displacement of the tooth from its alveolar socket. Due to the complexity of this injury, the neurovascular supply is severely compromised and usually results in loss of pulp vitality.[1]

When a tooth is avulsed, attachment damage and pulp necrosis occur. The tooth is 'separated' from the socket, mainly due to the tearing of the periodontal ligament which leaves viable periodontal ligament cells on most of the root surface.[2]

The status of the PDL cells is critical for the healing of replanted teeth. It is suggested that when the tooth is stored in a dry environment for longer than one hour, almost all the PDL cells attached to the root cannot survive.[4]

The best treatment protocol is the immediate replantation of the involved tooth at the accident site however; this may not be possible to occur. [5]

The ability of a storage/transport medium to support cell viability of the avulsed tooth before treatment can be more important than the extra oral time, for preventing ankylosis and replacement resorption.[6]

Various solutions have been investigated as storage media, such as Hank's balanced salt solution (HBSS), milk, aloe vera extract, propolis, coconut water, and egg white. The International Association of Dental Traumatology suggests that if there is access to special storage or transport media at the place of the accident, then media such as tissue culture/transport medium, HBSS, or milk can preferably be used. [2,5]

Ideal properties of a Transport medium
Should have antimicrobial characteristics
Should be able to maintain the viability of periodontal fibres for an acceptable period of time
It should have the same osmolarity as that of body fluids
It should not produce any antigen-antibody reaction
It should have a good shelf life
It should wash off extraneous materials and toxic waste products
It should aid in the reconstitution of depleted cellular metabolites

CLASSIFICATION OF TRANSPORT MEDIA

Laboratory prepared	Naturally available
Saline	Coconut water
HBSS	Milk
Viaspan	Egg white
Gatorade	Propolis
Contact lens solution	Aloe vera
Culture medium	Green tea extract
Emdogain	Pomegranate juice
Histidine-tryptophan-ketoglutarate (HTK)	Turmeric
Tooth rescue box	Neem
ORS	Salvia officinalis
Levodopa	Dragon's blood sap
Ascorbic acid	
Placenta derived mesenchymal stem cells – conditioned medium	

VARIOUS TRANSPORT MEDIA

Saline

Saline offers an osmolality of 280 mOsm/kg, and it is compatible with PDL cells. But it is deficient of essential nutrients such as magnesium, calcium, and glucose, required for normal metabolism of PDL.[3]

Saliva

Saliva was found to be more effective than tap water, but also has the potential for bacterial contamination. The osmolarity of saliva (60–80 Osm/l) was found to be much lower than the normal range (230–400 Osm/l) required for cell growth. Saliva is a hypotonic solution, causing periodontal cells to swell and burst.[4]

Coconut water

Natural coconut water is sterile and has a 93% water and 5% sugar composition, which gives it a high osmolality. It is rich in proteins, vitamins and minerals such as potassium, calcium and magnesium. According to Gopikrishna et al. (2008), this product was better than HBSS and milk in maintaining human fibroblast viability.[3]

HBSS (Hank's Balanced Salt Solution)

Hank's balanced salt solution (HBSS) is recommended as storage media of choice by AAE. It has appropriate pH and osmolality to maintain PDL cell viability. It is non-toxic, pH-balanced, and contains many essential nutrients. HBSS has an osmolality that ranges from 270 to 320 mOsm. It has been shown that solutions with osmolality in a range of 230–400 mOsm are good for cell growth, and optimal growth will occur in a range of 290– 330 mOsm.[10]

It is composed of 8 g/L sodium chloride, 0.4 g/L of D-glucose, 0.4 g/L potassium chloride, 0.35 g/L sodium bicarbonate, 0.09 g/L sodium phosphate, 0.14 g/L potassium phosphate, 0.14 g/L calcium chloride, 0.1 g/L magnesium chloride, and 0.1 g/L magnesium sulphate. These ingredients can sustain and reconstitute the depleted cellular components of the PDL cells.[11] HBSS is recommended by the International Association of Dental Traumatology as a storage medium for avulsed teeth. The pH and osmolality values of HBSS are suitable for cell growth and, hence, for preserving PDL cell viability.[7]

Sigalas E et al 2004 compared the viability of PDL cells in HBSS, culture medium, Gatorade, water, ice and contact lens solution. The results of their study showed that HBSS is an optimal storage medium for avulsed tooth when compared with other tested solutions.[5]

Ashkenazi et al showed that HBSS was the most effective medium for preserving viability, mitogenicity, and clonogenic capacities of PDL cells for up to 24 hours at 4°C, when compared with culture medium (EM supplemented with 15% foetal calf serum and antibiotic solution containing 100 UI/mL penicillin, 50 μg/mL gentamicin, and 0.3 μg/mL fungizone), EM, milk, ViaSpan, and conditioned medium.[11]

Recent studies have evaluated the use of 0.9% isotonic saline, milk, HBSS, and Viaspan as storage media for the preservation of cell viability. HBSS was the most effective although milk and saline were suitable, provided the extraoral time did not exceed 2 hours.[13]

Milk

Milk has been seen to be a compatible short-term storage medium of avulsed teeth, when the teeth are placed in it within 15 to 20 minutes. Milk only prevents cell death rather than restoring normal morphology and ability to differentiate and mitose. Gamsen *et al.* showed that milk is able to maintain the osmotic pressure for periodontal ligament cells but it does not have the ability to reconstitute depleted cell metabolites and restore viability. The easy availability of milk makes it one of the suitable medium for transport of avulsed tooth.

There is evidence that low fat milk maybe more appropriate in maintaining cell viability than high fat milk.[20]

Viaspan

The ViaSpan is a cold transplant organ storage medium and it has been very effective for storing avulsed teeth. It has an osmolality of 320 mOsm/kg and its pH is around 7.4 at room temperature, which is ideal for the cellular growth.

It has peculiar characteristics that allow maintaining cell viability, minimize damages to PDL cells and provide conditions for cell proliferation, therefore can be indicated for use as storage media for avulsed teeth.[9]

Egg white

The osmolarity of egg white is 300mosm/kg. It has shown better cell viability and significantly higher incidence of PDL healing. It is considered as a good choice because of its high protein content, water, vitamins, lack of microbial contamination, and easy accessibility.[4]

Propolis

Propolis, derived from honeybee extract, is a powerful agent with antimicrobial, antioxidant, anti-inflammatory, antibacterial, antifungal, antiviral, and tissue regenerative properties. Typically, propolis consists of 50% resin and vegetable balsam, 30% wax, 10% essential and aromatic oils, 5% pollen, and 5% various other substances, including organic debris.[12] It is nontoxic and inhibits osteoclastic resorption of teeth. It is not easily available. Ozan et al. from their study found that propolis was superior to HBSS or milk in maintaining PDL cell viability after storage of avulsed tooth. Similarly, efficacy of propolis in maintaining PDL cell was observed in many studies.[13]

Aloe Vera

Aloe vera contains 99% water and over 75 nutrients, which include 20 minerals, 19 amino acids and 12 vitamins. Aloe vera also contains allantoin, which has been found to stimulate fibroblast activity and collagen proliferation.[14]

The osmolarity of aloe vera was found to range from 280-300 mOsm/L and as normal cell growth occurs at an osmolarity range of 230 to 400 mOsm/L, the possibility of maintaining cell viability is high with aloe vera.[13] The presence of catalase enzyme, an antioxidant enzyme that converts hydrogen peroxide (H2O2) to water and oxygen and suppression of the generation of these free radicals may improve the effectiveness of cell preservation and prevent lipid peroxidation. Hence, presence of antioxidants in storage media is necessary for inhibiting the generation of free radicals thereby minimizing cell damage.[15]

Gatorade

Gatorade can be considered for the storage of the avulsed tooth, as it is a common sports drink used for rehydration and readily available at sports centres where most of the injuries take place. The pH of Gatorade is 2.91, and osmolality 407 is mOsm/kg. Because of its unfavourable pH and osmolality, cell growth was impossible and studies showed that the remaining cells were damaged when stored for a longer period.[9]

Contact lens solution

Contact lens solutions were initially thought to be of possible benefit as a storage solution for avulsed teeth because they are essentially saline solutions. They are comprised of a fatty acid monoester and a cationic antimicrobial component. The presence of preservatives in its formula was harmful to the cells of the PDL and therefore, they are not recommended.[11]

Culture medium

Culture media such as Eagle's medium, alpha-Minimum Essential Media (MEM) and αMEM-S (supplemented with foetal calf serum and antibiotic) have been shown to maintain the viability and proliferative activity of PDL cells for an extended period of time (48-53 hours) with a reduced rate of inflammatory resorption. This can be attributed to the availability in the culture medium of all the required essential nutrients for the growth and proliferation of PDL cells.[20]

Additionally, it consists of growth factors (platelet derived growth factor, insulin-like growth factor, epidermal growth factor, recombinant human platelet-derived factor-AB, natural human platelet-derived growth factor), in a culture medium has also been shown to increase the mitogenic and clonogenic capacity of PDL cells for as long as 24 hours.[2]

Emdogain

According to Ashkenazi and Shaked Emdogain diminishes the percentage of fibroblasts of the periodontal ligament with capability of forming colonies and that lowers the capability for the fibroblasts to repopulate the dental radicular surface after dental avulsion. Emdogain can delay, but not stop the development of replacement resorption, one of the worst complications of dental trauma.[14]

Green tea extract

Green tea extract (GTE) has been described to have extraordinary antioxidant, anti-

inflammatory, and anticarcinogenic efficacy and to prolong allograft survivals properties. Hwang et al from their study found that green tea is equally effective as HBSS and better than milk for preservation of periodontal ligament cells of avulsed tooth.[17]

The tooth rescue box

The tooth rescue box introduced by Dentosafe; Germany contains a tissue culture medium similar to a medium used during islet cell transplantation. Besides different salts the medium also contains amino acids, vitamins, and glucose. The medium was shown to maintain vitality and proliferative capacity of PDL cells for up to 48 h at room temperature.[20]

Histidine-tryptophan-ketoglutarate (HTK)

The osmolality and physiological pH of Histidine-tryptophan-ketoglutarate (HTK) are known to enhance cell proliferation., mannitol as an oxygen-free radical scavenger and ketoglutarate as an energy source. HTK solution is highly feasible because it is easy to store.[17]

Pomegranate juice

Emadi F et al in their study compared Pomegranate juice with HBSS and tap water which resulted in three times more cell viability in PJ compared to HBSS after 24 hours. Punicalagin is the major antioxidant polyphenol ingredient in PJ is assumed to be effective for maintaining PDL cell viability. It is known that pomegranate flavonoids have anti-inflammatory and antibacterial properties, while pomegranate polyphenols have antioxidant and antiviral properties which may result in higher viability of PDL cells.[18]

ORS (Oral rehydrating solution)

ORS solutions are used to combat dehydration in cases such as diarrhoea. They consist of essential cell nutrients, such as glucose and vital salts, in 77concentrations deemed adequate for the cell metabolism to remain unhindered. ORS solutions are readily available to the common man over the counter at any pharmacy and are economically feasible. Rajendran P et al. observed in their study that ORS preserved vitality of the PDL cells equal to HBSS than milk.[26]

RESEARCH ON NEWER TRANSPORT MEDIA

Levodopa

Levodopa (L-dopa) a precursor of central nervous system catecholamines is a drug with possible mitogenic effects. Mandona et al., observed the effect of levodopa on human PDL fibroblasts and concluded that L-dopa due to its mitogenic activity can be used for preserving viable cells.[19]

Ascorbic acid

Ishikawa et al studied the effect of ascorbic acid on PDL cells and observed that ascorbic acid increased the ALP activity, which is required for the binding of PDL cells to type I collagen via 2 beta 1 integrin, whose expression is again increased by ascorbic acid. As type I collagen production is considered to be an initial process in differentiation of PDL cells, it may serve as a potential storage medium.[11]

Turmeric

Mandrol *et al*. investigated the *in vitro* cytotoxicity of curcumin against primary dental pulp fibroblasts by MTT assay. No cytotoxicity was detected and the results revealed that the viability of primary dental pulp fibroblasts increased with an increasing concentration of curcumin. Curcumin promotes cell viability and induces proliferation of dental pulp fibroblasts and thus can be used as a suitable natural storage medium.[21]

Neem

A study conducted by Dhimole P and Bhayya DP et al showed that neem (*A. indica*) serves as a good storage medium as majority of the fibroblasts were viable. The result can be attributed to its several active constituents such as nimbidin, nimbin, nimbolide, azadirachtin, and cyclic trisulfide which are responsible for its antibacterial action. The Phenolics in Neem exhibit antioxidant activity by inactivating lipid-

free radicals or preventing decomposition of hydroperoxides into free radicals.[25]

Salvia officinalis

It is well known the antioxidant activities of Salvia species are due to the beneficial properties of their phenolic constituents. Ozan F & Polat ZA et al. in their study compared S. officinalis with HBSS, PBS & tap water. The results of their investigation showed that 2.5% S. officinalis can preserve PDL cell viability & was better than the other tested solutions at 24hrs.[27]

Dragon's blood sap

Martins CM, Hamanaka EF et al in their study showed that the dragon's blood sap preserved cell viability up to 24 h and they concluded that the dragon's blood sap was an efficient storage medium for avulsed teeth since it preserved membrane integrity of different cell types and maintained functional viability of periodontal ligament cells.[21]

Placenta derived mesenchymal stem cells – conditioned medium

Placenta-derived mesenchymal stem cells (PMSCs), which originate from placental tissue, can secrete numerous cytokines, including vascular endothelial growth factor (VEGF), platelet-derived growth factor (PDGF), transforming growth factor (TGF), epidermal growth factor (EGF), basic fibroblast growth factor (bFGF), and insulin-like growth factor (IGF). Ji LL, Song G in their in vitro study suggested that PMSC-CM could better maintain the viability of PDLSCs during a long period of time (48 h) compared with HBSS. Additionally, PMSC-CM inhibited the apoptosis of PDL cells, without any effect on cell cycle.[5]

CONCLUSION

Up to now, there is not a single product or solution that possesses all the characteristics required to be indicated as the ideal storage/ transport medium for avulsed teeth, that is, be capable of preserving the vitality of the PDL and pulp cells, while presenting compatible physiological pH and osmolality, clonogenic capacity, antioxidant property, no or minimal microbial contamination, high availability, ready accessibility at accident sites, homes, schools, hospitals and dental offices, and low cost.

The literature suggests milk as the most viable option in terms of PDL cell viability, ease of availability and cost effectiveness followed by HBSS. Among the natural products other than milk, propolis and coconut water were frequently recommended.

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