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REVIEW ARTICLE

How has Tooth Manipulation been Conducted for Dental Pulp Stem Cells Isolation? A Scoping Review

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ABSTRACT

The aim of this study was to realize a scoping review the literature to verify how tooth manipulation for dental pulp stem cells (DPSCs) isolation has been conducted and if a standard tooth, preparation protocol for DPSCs isolation exists. The electronic search was conducted without initial date restriction up to and including April 2014 in PubMed, Scopus, Scielo, and ISI Web of Knowledge databases to identify studies that described the methodology used for DPSCs isolation. Two hundred and twenty-two articles were included and the information analysis was performed concerning dental manipulation and pulp tissue processing.

Furthermore, the quality of included studies was evaluated through the assessment of the risk of bias. This scoping review established a platform for dental manipulation protocols for DPSCs isolation purposes. Over the past years, many studies have been conducted using DPSCs. However, there is a clear lack of standardization in tooth manipulation before DPSCs isolation. Currently, given a large number of variables in cell isolation techniques and all possible consequences in the in vitro behavior of cells, it is important to reinforce the importance of standard protocols to obtain a uniform cell culture.

Keywords: Dental pulp, Dental pulp stem cell, Manipulation, Scoping review, Stem cell.

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INTRODUCTION

Mesenchymal stem cells (MSCs) have been considered a promising treatment alternative for illnesses such as type 1 diabetes[1] and heart disease,[2] among others. Advances in the treatment of these diseases are closely associated with the growing number of stem cell research; however, most studies are developed in animal models, suggesting that more research is needed to translate results to human applications.[3] For this, a cell source easily obtainable, with rapid *in vitro* expansion and high proliferative rates is mandatory. Among available options with such characteristics are the dental pulp stem cells (DPSCs).[4]

DPSCs are found in small quantities in the human dental pulp.[1,5] Therefore, the simulation of the cellular microenvironment must be the best possible to achieve a sufficient cell proliferation rate and quality for *in vitro* or *in vivo* purposes.[6] However, teeth manipulation and pulp tissue processing for cell isolation is a complex task and can be determinant in the success of stem cell isolation.[7]

Although DPSCs have been widely used in studies with clinical applications, the literature is scarce in relation to tooth preparation before DPSCs isolation. Thus, the aim of this study was to realize a scoping review to verify how tooth manipulation for DPSCs isolation has been conducted and if a standard tooth preparation protocol exists. To the best of our knowledge, this is the first scoping review evaluating tooth manipulation for DPSCs isolation.

MATERIALS AND METHODS

Study Questions

How tooth manipulation for DPSCs isolation has been conducted?
Is there a standardized protocol for tooth manipulation to isolate DPSCs?

Inclusion and Exclusion Criteria

To be included, the study had to describe stem cell isolation from human dental pulp of permanent teeth. Exclusion criteria were studies using cells from a non-human source, stem cells from sources other than the dental pulp, human cells but not stem cells, SHEDs, and studies in which the tooth manipulation and isolation technique were not described. Literature reviews, congress abstracts, patents, book section, hypothesis articles, editorial, letters to the editor, news, protocols, interview, articles which are not written in English and that were not fully available even after attempting to contact the authors were also excluded. Articles were not excluded due to more than one exclusion criterion.

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Search Strategy

The electronic search was conducted without initial date restriction up to and including April 2014 in PubMed, Scopus, Scielo, and ISI Web of Knowledge databases to identify studies that described the methodology used for DPSCs isolation. An initial search was conducted using the following terms: "[(dental pulp stem cell (MeSH)]"; "[dental pulp (MeSH)]" and "[stem cell (MeSH)]"; "["dental pulp stem cell" (MeSH)]". No language and date restrictions were applied in the search.

All references were managed in EndNote X7 software (Thomson Reuters, New York, NY, US). Initially, duplicate references were excluded. Titles, abstracts, and methodologies were screened based on the inclusion and exclusion criteria by two reviewers independently (CPF and CCdoA). Lists were compared, and in case of disagreement, a consensus was reached by discussion. When a consensus was not achieved, a third reviewer decided if the article should be included (FN). This scoping review followed the PRISMA statements[8] with some adjustments (Fig. 1).

Data Extraction

After screening, the following data were collected from articles: name of authors, year of publication, donor's age, donor's gender, tooth type, time and tooth storage methods between the extraction and DPSCs isolation, tooth surface cleaning methods, location and methods of dental section, and methods used to remove the pulp from the dental chamber. Data were extracted and tabulated independently by two reviewers (CPF and RGS) to be submitted to a descriptive analysis. Cases of disagreement were discussed until a consensus was reached. When a consensus was not obtained, a third reviewer participated in the discussion (FN).

Assessment of Risk of Bias

Risk of bias was evaluated according to the following parameters for study quality assessment (a) donor's age, (b) donor's gender, (c) tooth type, (d) tooth storage methods, (e) tooth storage time, (f) dental surface cleaning (g) dental section methods (h) dental section location and (h) pulp removal methods. A parameter was given a "Y" (yes) if it was described in the study, if it was mentioned but not specifically, it was marked with a "U" (unclear); and finally if the parameter was not described, it was given an "N" (no). After the evaluation, the data were imported into Review Manager 5.3 for analysis and graph generation. Studies withup to 30% of "Y" had a high risk of bias, above 30% and lower than 65% had a medium risk, and above 65% low risk of bias.

RESULTS

Descriptive Analysis

The electronic search yield 3.126 articles. From those, 1.539 were duplicated and removed. A total of 1.587 articles were included by title, abstract, and methodology screening. From those, 222 were included for full-text analysis (Fig. 1).

Most of the studies were published in 2011 and 2013 (Supplementary material 1). Figure 2 shows that tooth donorsare predominantly males with an age range from 17.5–30.6 years.

Of the studies included in this review, 84% described the type of tooth used for DPSCs isolation. The second most frequently mentioned item was the donor's age (67.6%). However, the tooth storage time and method were the most neglected ones, described in 11.7% and 15%, respectively (Table 1).

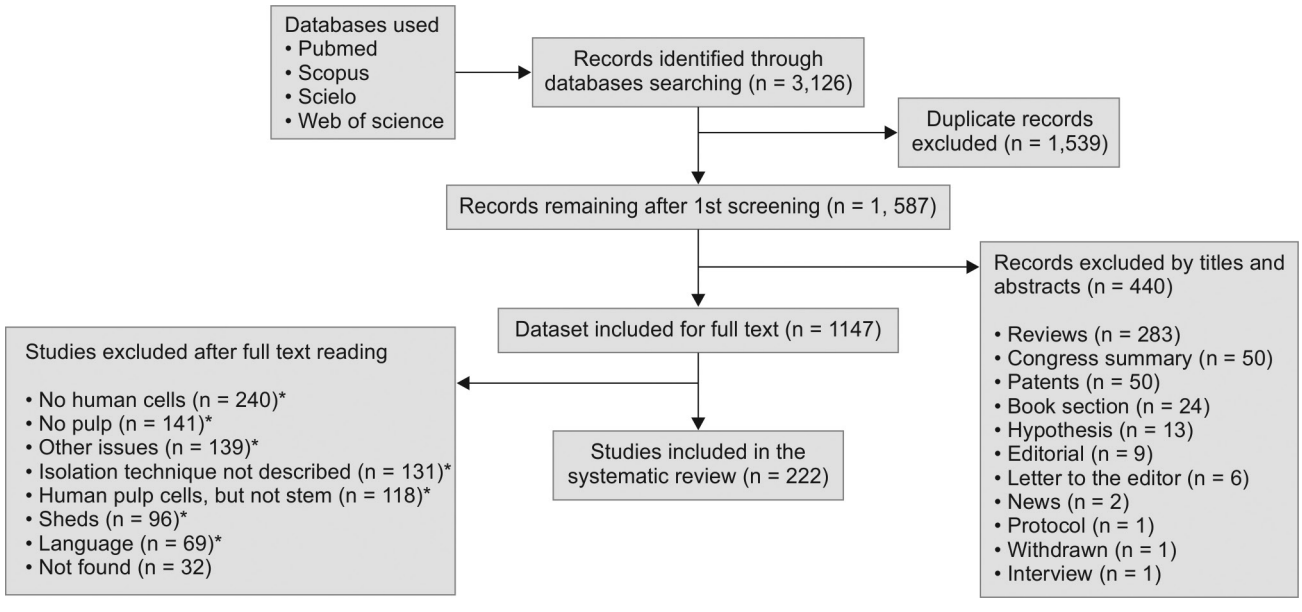

Fig. 1: Flowchart with the studies selection process for inclusion in the systematic review
(Exclusion reasons: a study could fulfill more than one criteria)

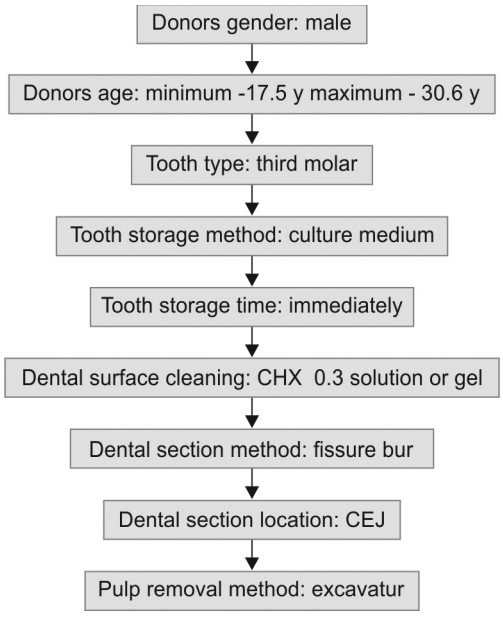

Fig. 2: Flowchart with the most commonly used methods to manipulate tooth prior to DPSCs isolation

Table 1: Distribution according to the presence or absence of important aspects of tooth preparation prior to DPSCs isolation

	Yes		*No*	
	N	*%*	*N*	*%*
Tooth type	187	84.2	35	15.8
Donors age	150	67.6	72	32.4
Donors gender	35	15.8	187	84.2
Tooth storage methods	34	15.3	188	84.7
Tooth storage time	26	11.7	196	88.3
Dental surface cleaning	85	38.3	137	61.7
Dental section method	84	37.8	138	62.2
Dental section location	42	18.9	180	81.1
Pulp removal methods	54	24.3	168	75.7

Concerning the type of teeth used for DPSCs isolation, data shows that the third molar is the most commonly used, at 77.6% or even higher since 9.2% reported just molars without specifying whichone-first, second or third. The second most commonly used teeth are the premolars (10.7%). Supernumerary and incisors teeth represent 2.6%, and the canine was not mentioned in the studies (Table 2).

In relation to the storage method used to transport the newly extracted teeth to the site of DPSCs isolation, seven different types of solutions were cited. The most common was the same medium used subsequently for DPSCs culture (44% of the studies), followed by HBBS (20.6%). Considering the time spent between the extraction and isolation, the majority of authors (55.6%) conducted the isolation of DPSCs immediately after extraction and over 92% within 24 hours (Table 3).

Thirty-eight percent of the articles reported cleaningthe tooth surface before sectioning the dental element to access the pulp chamber (Table 1). Thirteen cleaning solutions were mentioned, used alone or in combination with each other. PBS and CHX were the most cited solutions, each one used in 14.7% of the studies. CHX was used in concentrations of 0.2 and 0.3% as a solution or gel (Table 4).

In the normal procedure, after cleaning, the teeth are sectioned so that the pulp chamber and the pulp tissue can be access. However, only 37.8% of the papers reported the sectioning method (Table 1). The most used tools

Table 2: Distribution according to the types of teeth used to isolate DPCSs.

	N	*%*
Supernumerary		
	2	1.0
Incisor		
Incisor (not described the type)	1	0.5
Left upper central incisor - traumatized	1	0.5
Left upper lateral incisor - traumatized	1	0.5
Incisor total	3	1.5
Premolar		
Premolar (not described the type)	18	9.2
First premolar	2	1.0
Lower premolar	1	0.5
Premolar total	21	10.7
Molar		
Molar (not described the type)	18	9.2%
Third molar		
Third molar (not described the type)	88	44.9
Impacted third molar	57	29.1
Upper third molar	3	1.5
Mandibular third molar	1	0.5
Non-erupted third molar	1	0.5
Semi-erupted third molar	1	0.5
Semi-impacted third molar	1	0.5
Third molar total	152	77.6
Total	196	100.0

Table 3: A) Distribution according to the storage solution used to transport the newly extracted teeth to the site of DPSCs isolation. Abbreviations: Hank's buffered salt solution - HBBS; phosphate buffered saline – PBS; Dulbecco's phosphate buffered saline – DPBS; Fetal bovine serum – FBS. B) Distribution according to the tooth storage time between the extraction and DPSCs isolation

A: Storage solution to transport teeth		
	N	*%*
Culture medium	15	44.1
HBBS	7	20.6
PBS	4	11.8
DPBS	2	5.9
Saline solution	2	5.9
Freezing medium	1	2.9
FBS	1	2.9
Stored (not described)	2	5.9
Total	34	100.0
B: Tooth storage time		
	N	*%*
Immediately	15	55.6
Within 2 h	6	22.2
Within 24 h	4	14.8
6 to 48 h	1	3.70
72 h	1	3.70
Total	27	100.0

Table 4: Distribution according to the tooth surface cleaning method. Abbreviations: clorhexidina - CHX; phosphate buffered saline - PBS; Dulbecco's Phosphate-Buffered Saline - DPBS; povidone-iodine - PVP-I; Hank's Balanced Salt Solution - HBSS.

	N	%
CHX		
0,2%	3	2.6
0,2% solution	1	0.9
0.3%	3	2.6
0,3% solution	7	6.0
0,3% gel	7	6.0
Not described the type	2	1,69
Solution	1	0.9
CHX total	17	14.7
PBS	17	14.7
Professional hygiene	15	12.9
DPBS	9	7.8
Ethanol	9	7.8
PVP-I	8	6.9
Physiological solution	4	3.5
Destilled water	3	2.6
Sodium thiosulfate	2	1.7
Sterile surgical blade	1	0.9
HBSS	1	0.9
Dental burs	1	0.9
Dental scaler	1	0.9
Not described	28	24.1
Total	116	100.0

described were the fissure bur, forceps, and diamond discs, representing 24.7, 18.3, and 11.8%, respectively (Supplementary material 2).

Few studies indicated at which level the teeth were sectioned, of that 88.1 % chose the cementum-enamel junction (CEJ) (Supplementary material 3).

More than 75% of the articles did not mention the methods used to remove the pulp tissue from inside the dental chamber (Table 1). Eight different instruments were used to remove the pulp tissue, the most common being the excavator, used in 59.7% of the studies. The second most used instrument was the forceps (11.7% of the studies, Supplementary material 4).

Risk of Bias

Of the 222 included studies, age and tooth type showed a low risk of bias (>65%). Dental surface cleaning and section methods showed the medium risk of bias (above 30% and lower than 65%). Donor's gender, tooth storage methods, tooth storage time, section site and pulp removal methods presented a high risk of bias (<30%) (Supplementary material).

DISCUSSION

This scoping review demonstrates that DPSCs isolation has been reported since 2000. In the first five years following the discovery of DPSCs, only five articles were published. The number of publications increased in 2006 (5 publications) and continued to increase in the following years reaching its peak in 2013 (Fig. 3). Although great knowledge in DPSCs biology and its applications has been produced during these years, this scoping review shows the lack of standardization towards dental preparation before DPSCs isolation. This is the first time that the vast literature regarding tooth manipulation for DPSCs isolation has been summarized in a rigorous and replicable manner (Fig. 2).

The most commonly used teeth to obtain DPSCs were the third molars. According to Gronthos et al.[4] DPSCs derived from molars have a greater degree of cell proliferation *in vitro*, when compared with bone marrow stem cells (BMSCs), and this behavior is attributed to differences in the development stage of each organ[4]. In addition, third molars are easily accessible and commonly indicated for extraction for orthodontic reasons.[9]

Most articles neglected the description of tooth donors'gender (84%), and when described, males were the most prevalent donors (55.9%) (Table 1). Also, 67.6% of the articles mentioned the donor's age (Supplementary material), with a range of 17.5–30.6 years. It has been reported that the final development of the lower and upper third molars occurs at the age of 21.6 and 22.3

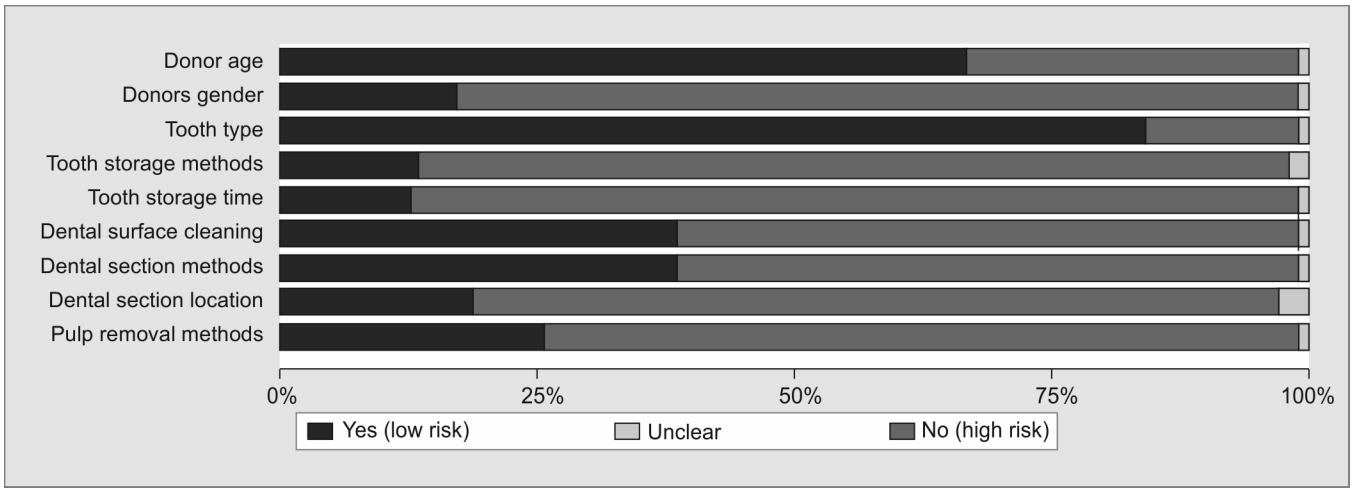

Fig. 3: Risk of bias considering aspects reported in the material and method section

respectively, in women. In men, the lower and upper third molar complete their development at 21.8 and 22.4 years.[10] Age-related issues could decrease cell number or function leading to tissue deterioration.[11] It has been suggested that stem cells from the marrow of younger individuals could have a greater pool size than from older people. The difference is related to the changes in the skeletal dynamics from a modeling mode of skeletal growth and consolidation to a remodeling dynamic of the adult skeleton.[12] The same occurs with DPSCs, in which the most indicated time for stem cell isolation is during teeth's developmental period, as stem cells are still in growth and consolidation phases, rather than in the remodeling phase.[9]

Often, teeth extraction and dental pulp tissue processing occur in different locations and thus the time and conditions in which teeth are transported to one place to the other are important factors.[13,14] However, our results indicated that most studies neglected these aspects (Table 1). Even though the need for teeth conditioning is evident.[14] our results show a high risk of bias for this variable (Fig. 1). The most commonly used media were the same medium used subsequently for DPSCs culture (44.1%), Hank's balanced salt solution (20.7%), and PBS (11.7%).

This review showed that over 88.3% of the studies did not report teeth storage time (Table 1). Among the studies that reported this factor, 92.6% initiated cell dissociation within 24 hours after tooth extraction. The literature suggests that teeth maintained in the cell culture medium, PBS or hypo-thermosol (HTS) can conserve DPSCs viability for up to 120 hours,thus contradicting the hypothesis that DPSCs are viable only when isolated within 48 hours after dental extraction.[14]

Another important issue considering teeth processing before DPSCs isolation is the cleaning procedure of the dental surface. Our review showed that nearly 62% of all studies did not mention the process used for cleaning the tooth surface before the dental section (Table 1). However, this procedure might be critical forthe prevention of cell culture contamination, since more than 700 bacterial species or phylotypes,[15] mycoplasmas,[16] and a wide range of fungi[17]are detected in the oral cavity. The authors who reported this step mainly used substances such as chlorhexidine(15% of the total studies). Chlorhexidine has been shown to present antibacterial and antifungal properties,[18] which justifies its use for surface cleaning before dental section. Chlorhexidinehas bactericidal activity at high concentrations (>0.12%) and bacteriostaticactivity at low concentrations (0.02–0.06%).[19] This could explain why 3.5% of the authors opted to disinfect the teeth surface with chlorhexidineat 0.2%, while 8.6% used 0.3% chlorhexidine. Another group of researchers (2.6%) used mechanical tools to clean the surface of teeth such as dental burs, a dental scaler, and a sterile surgical blade. Some authors (12.9%) suggested professional oral prophylaxis before tooth extraction.

As pioneers in DPSCs isolation, Gronthos et al.[4] provided the basis for many researchers. In agreement to their methodology,[4] 88.1% of the included studies sectioned the tooth at the cementum-enamel junction, and 24.7% used dental fissure burs. To perform this step, a high-speed handpiece coupled to a dental unit is used. However, this equipment is often unavailable in most laboratories and therefore it is commonly performed in the clinic, jeopardizing the sterile environment needed to prevent contamination of the pulp tissue, and consequently the cell culture. In addition, the literature suggests that thermal damage should be avoided since a 5.5°C increase in intrapulpal temperature can cause irreversible damage to the pulp tissue.[20] Subsequently, once the pulp chamber has been accessed, the next step is to remove the pulp tissue. Of all the articles included in this review, 75.7% did not mention this step. According to our findings, the use of excavators is the choice of approximately 60% of authors who mention the method used for dental pulp tissue removal.

After collecting the dental pulp, DPSCs can be isolated using two main techniques: explant[21-23] and enzymatic.[22,23] The association of the enzymatic technique and the use of mechanical devices to intensify cell dissociation is also frequently used.[24]

This scoping review clearly shows the lack of information provided in articles, since a high risk of bias was found in almost all variables evaluated, with exception to donor's age (67.6%) and tooth type (84.2%). We propose that information such as donor's age, type of tooth, storage medium, storage time, tooth surface cleaning method, dental section method, tooth section site, and method for removing the pulp tissue from the chamber should be standardized and provided in all original articles, so that protocols could be well established. Based on the frequency that each step of DPSCs isolation was reported in our scoping review, we developed a platform (Fig. 2). However, it is important to highlight that this platform is based on frequency analysis and not in the effectiveness each step. We recognize that ideally the outcomes of the articles should be assessed instead of the frequency of the methods used.

Nonetheless, negative results are usually not published, and if outcomes were to be considered, they could lead to a great risk of bias. In addition, most studies included in this scoping review were not strictly methodological (evaluating only tooth manipulation) andassessed secondary outcomes. We can speculate, however, that if DPSCs isolation was described in the methodology section of studies evaluating secondary variables, the tooth manipulation method was successful. Therefore, we believe that the frequency of methods would be the most accurate data to evaluate.

CONCLUSION

Over the past 15 years, many studies have been conducted using DPSCs. However, there is a clear lack of standardization in tooth manipulation before DPSCs isolation. Thus, given a large number of variables in cell isolation techniques and its consequences in the *in vitro* behavior of cells, it is important to reinforce the need for standard protocols to obtain a uniform cell culture.

CLINICAL SIGNIFICANCE

Tooth manipulation for DPSCs isolation is a complex task, dependent on aspects such as time and tooth storage methods between the extraction and DPSCs isolation, tooth surface cleaning methods, location and methods of the dental section, and methods used to remove the pulp from the dental chamber, which determine the success of stem cell isolation. The adequate process of cell isolation seems to be fundamental for the use of these cells as therapeutic tools in tissue engineering, cell therapy, and other health areas.

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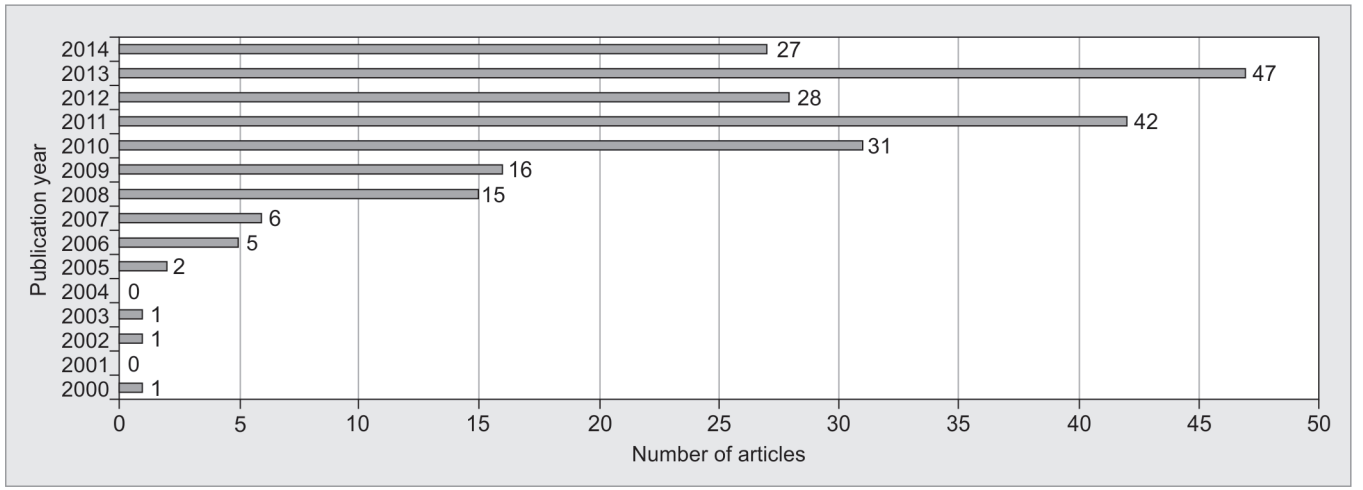
SUPPLEMENTARY MATERIAL

Supplementary material 1. Distribution of articles included in the systematic review according to the year of publication



Supplementary material 2: Relation of the impact factor of the journals in the years of publications articles with the presence of variables of interest.

	Donors age % (n)		*Donors gender* % (n)		*Tooth type* % (n)		*Tooth storage methods* % (n)		*Tooth storage time* % (n)		*Dental surface cleaning* % (n)		*Dental section method* % (n)		*Dental section location* % (n)		*Pulp removal methods* (n)	
Impact factor	*Y*	*N*	*Y*	*N*	*Y*	*N*	*Y*	*N*	*Y*	*N*	*Y*	*N*	*Y*	*N*	*Y*	*N*	*Y*	*N*
Lower than 5	88.2 (150)	11.8 (20)	15.9 (27)	84.1 (143)	100 (170)	0.0 (0)	15.4 (26)	84.6 (143)	11.8 (20)	88.2 (150)	36.5 (62)	63.5 (108)	35.3 (60)	64.7 (110)	16.5 (28)	83.5 (142)	22.4 (38)	77.7 (132)
Higher than 5	0.0 (0)	100 (27)	18.5 (5)	81.5 (22)	63 (17)	37 (10)	3.7 (1)	96.3 (26)	7.4 (2)	92.6 (25)	51.7 (15)	48.3 (14)	55.6 (15)	44.4 (12)	37 (10)	64 (17)	25.9 (7)	74.1 (20)

Supplementary material 3: Distribution according to the method used for dental section

	N	%
Fissure bur	23	24.7
Forceps	17	18.3
Diamond discs	11	11.8
High speed	8	8.6
Other 19 methods	34	36.6
Total	93	100.0

Supplementary material 4: Distribution according to location of the dental section

	N	%
CEJ	37	88.1
Root enamel boundary	4	9.5
Crown-root border	1	2.4
Total	42	100.0

CEJ, Cementum enamel junction

Supplementary material 5: Distribution of data collected from articles to analyze the variables of interest

Author and year of publication	*Tooth*	*Cell type*	*Age at the extraction*	*Sex*	*Storage method for transporting*	*Storage time*	*Hygiene*	*Methods section of tooth*	*Methods for removing the pulp from the interior of the chamber*
Abdullah 2014		DPSCs	10-40 y			Within 24 h	Immerssion in 75% ethanol and soaking in PBS		
Abu Kasim 2012	Third molar	DPSCs	24–35 y			2 h	Root surface cleaned with PVP-I (Sigma-Aldrich, St. Louis, MO, USA)		
Agha-Hosseini 2010	Impacted third molar Semi-impacted third molar	DPSCs	18–28 y				Rinse mouth with CHX before extraction	Cow horn forceps	Small escavator
Ahmed 2011	Third molar	DPSCs	20-29 y		DMEM (Biowhittaker, Gibco, Sigma, USA), penicillin/ streptoycin (Invitrogen Co, USA) and 10% FBS (JRH biosciences, Inc., Lenexa, KS, USA)				Sterile dental probe
Akkouch 2014	Third molar	DPSCs	18–25 y		PBS contaning antibitics, on ice			Cut around the circumference of the teeth using a sterile hand-held high-speed drill at the CEJ level	Endodontic file
Al-Habib 2013	Impacted third molar	hDPSCs	16–24 y		Cell culture medium				
Alongi 2010	Impacted third molar	DPSCs-NPs DPSCs-IPs	14-22 y		Tissue freezing medium (Triangle Biomedical Sciences, Durham, NC, USA) or culture medium				
Armiñán 2009	Third molar	DPSCs	18-21 y						

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
Arthur 2008	Impacted third molar	DPSCs	19-35 y				Cleaned (not described)	Torno (vice)	
Asgary 2014	Third molar	DPSCs					Disinfected by 70% ethanol	Dissected at the crown-root border	
Atari 2011	Third molar	DPMSCs	14-60 y	Male Female		Immediatly	Washed using gauze soaked in 70% ethanol, followed by a wash with sterile destilled water	Hold the tooth with upper incisor fórceps. The incision was in CEJ by using a cylindrical turbine bur	Sterile nerve-puller file 15 and forceps
Atari 2012 (a)	Third molar	DPPSCs DPMSCs	18–27 y	Male Female		Immediatly	Washed using gauze soaked in 70% ethanol, followed by a wash with sterile destilled water	Hold the tooth with upper incisor fórceps. The incision was in JEC by using a cylindrical turbine bur	Sterile nerve-puller file 15 and forceps
Atari 2012 (b)	Third molar	DPPSCs DPMSCs	14–60 y	Male Female		Immediatly	Washed using gauze soaked in 70% ethanol, followed by a wash with sterile destilled water	Hold the tooth with upper incisor fórceps. The incision was in JEC by using a cylindrical turbine bur	Sterile nerve-puller file 15 and forceps
Attar 2014	Molar Third molar	PPSCs DPSCs						Teeth cut around the root enamel boundary using dental fissure bur	Polyps were removed with curettage
Bakopoulou 2011	Impacted third molar	DPSCs	16-18 y		.		Cleaned (not described)	Cut around the CEJ	

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Author and year of publication	*Tooth*	*Cell type*	*Age at the extraction*	*Sex*	*Storage method for transporting*	*Storage time*	*Hygiene*	*Methods section of tooth*	*Methods for removing the pulp from the interior of the chamber*
Batouli 2003	Impacted third molar	DPSCs	19-29 y						
Bonnamain 2013	Non-erupted third molar		15–20 y	Male Female					
Bressan 2012	Molar	DPSCs	16-66 y				Pretreatment for one week with professional dental hygiene. Before extraction dental crown covered with 0.3% CHX gel (Forhans, New York, NY) for 2 min	Mechanical fracturing	Dentinal excavator or a gracey curette
Cai 2011	Impacted third molar	hDPSCs	18-28 y						
Carinci 2008	Molar								Dentinal excavator or a gracey curette
Carvalho 2012	Upper third molar	hDP-DPSCs					Cleaned (not described)	Diamond cutter disc	
Chen 2012	Third molar	DPSCs	19-23 y		α- MEM serum free (Hyclone, Logan, UT, USA)	1 h			
Chen 2013		hDPSCs - liquid nitrogen stored dental pulp tissues hDPSCs - freshly derived dental pulp tissues	Mean of 26.5 y Mean of 23.4 y						Endodontic file

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Author and year of publication	*Tooth*	*Cell type*	*Age at the extraction*	*Sex*	*Storage method for transporting*	*Storage time*	*Hygiene*	*Methods section of tooth*	*Methods for removing the pulp from the interior of the chamber*
Chen 2011		hDPSCs:	Mean of 25.5 y, 6–74 y	Male Female			Cleaned the tooth surface with DPBS	A circumference groove of 0.5–1.0 mm in depth was cut around the entire tooth using an aseptic high speed handpiece. The tooth was split using a chisel.	Endodontic file
		freshly derived dental pulp tissues	Mean of 26.5 y, 6-74 y						
		liq N2-stored dental pulp tissues	Mean of 23.4 y, 6-49 y						
		liq N2-stored intact teeth	Mean of 27.7 y, 8-52 y						
Choi 2012	Molar	DPSCs			HBSS (Welgene, Dae-gu, Korea) with 3% AA (Life Technologies, Carlsbad, CA) at 4°C			Dental high-speed unit	
Chun 2011 (a)	Third molar	DPSCs							
Chun 2011 (b)		DPSCs							
Cmielova 2013	Impacted thrird molar	DPSCs	Mean of 19 y, 12-23 y	Male Female	Stored (not described)		Cleaned (not described)		
Collart-Dutilleul 2014	Impacted third molar	DPSCs	15−18 y				Tooth surfaces were cleaned using 0,2% CHX	Cut around the CEJ	

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
Cui 2014	Third molar	hDPSCs							
Cui 2013	Third molar	hDPSCs							
Dai 2012	Third molar	DPSCs	Mean of 18+ -3.2, 16-25 y						
D'Alimonte 2013		DPSCs	Mean of 17 y	Male Female					
D'Alimonte 2011		DPSCs	Mean of 17 y	Male Female					
D'Aquino 2009	Third molar	DPSCs		Male Female			Professional oral hygiene one week before surgery 0,2% CHX after brushing , twice a day	Surgical drill	
D'Aquino 2007		DPSCs		Male Female			Professional oral hygiene one week before surgery 0,3% CHX after brushing , twice a day	Surgical drill	Dentinal excavator or a gracey curette
de Rosa 2011		DPSCs	21-45 y						
de Souza 2010	Impacted third molar	DPCp	9-15 y				Cleaned (not described)	Longitudinal groove and sterilized diamond discs (KGSorensen, ref.7020, Zenith Dental ApS, AgersKov, Denmark)	Sterile dentinal excavator
Demircan 2011	Molar	hDP-SC	20 y	Male			Immerssion in physiologic solution containing antibiotics	Pliers (bone forceps)	

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
Diomede 2013	Premolar	hDPSCs	18-25 y				Rinsed five times with PBS containing penicillin and streptomycin	Cylindrical diamond bur (314, Ø ISO 014, L.8.0mm Intensiv, Grancia, Switzerland) mounted on a high speed handpiece (Bora L; Bien Air, Bienne, Switzerland) with water spray cooling	Sterile excavator
Dissanayaka 2012	Third molar	DPSCs	18-25 y				Cleaned (not described)	Sterile fissure bur at the CEJ	
Dissanayaka 2011	Third molar	hDPSCs					Cleaned (not described)	Cut at the CEJ by using a sterile fissure bur	
Djouad 2010	Third molar	DP-MPCs	16-26 y						
Dolatshahi-Pirouz 2010	Third molar	DP-MSC	21 y						
Duailibi 2011	Mandibular third molar	DSCs			HBSS (Gibco BRL, Gaithersburg, MD, USA) pre-warmed 37°C				
Ebrahimi 2011		DPSCs			PBS solution containing penicillin and streptomycin on ice		Disinfected in iodine solution, washed with PBS	Cracked using a turbine along the cervical region and with sterilized cowhorn forceps	Gracey curette

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Author and year of publication	*Tooth*	*Cell type*	*Age at the extraction*	*Sex*	*Storage method for transporting*	*Storage time*	*Hygiene*	*Methods section of tooth*	*Methods for removing the pulp from the interior of the chamber*
Egbuniwe 2011	Third molar	tDPSCs					Washed with 70% ethanol and then with HBSS (Invitrogen) p.H 7.4	Horizontal indentations along the cervical margin using a low speed circular diamond circular diamond saw (Agar Scientific Ltd.)	Sterile forceps
		nDPSCs							
Eleutério 2013	Premolar teeth	DPSCs	20-35 y				Professional oral hygiene one week before surgery. Cleaned with PBS containing penicillin and streptomycin	Cylindrical diamond rotary cutting instrument m ounted on a high-speed handpiece with water-spray cooling	Sterile dentinal exacavator
Eslaminejad 2013	Third molar	DPSCs	20-25 y	Male				Cut around the root-enamel boundary using dental fissure bur	
Eslaminejad 2010 (a)	Third molar		20-25 y					Cut around the root-enamel boundary using dental fissure bur	
Eslaminejad 2010 (b)	Third molar		20-25 y						
Eslaminejad 2009	Third molar		20-25 y					Cut around the root-enamel boundary using dental fissure bur	
Eubanks 2014		DPSCs	15–22 y		Sterile saline solution or α-MEM containing 15% FBS	Immediately 24 h at 4°C		Cut above the CEJ	

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
Fang 2013	Third molar	DPSCs	16-30 y		Culture medium containing 100 U/mL penicillin and 100 mg/mL streptomycin		Desinfected with PVP-I for 5 min PBS containing 100 U/mL penicillin and 100 mg/mL streptomycin		
Feng 2013 (a)	Impacted third molar	DPSCs	13-23 y				Cleaned (not described)	Cutting around the CEJ using sterilised dental fissure bur	
Feng 2014	Impacted third molar	DPSCs	13-23 y				Cleaned (not described)	Cutting around the CEJ using sterilised dental fissure bur	
Feng 2013 (b)	Impacted third molar	DPSCs	45-50 y				Dental pulp tissue was washed three times with PBS		
Foudah 2014	Third molar	hDPSCs			PBS	Maximum of 1h		Piezoelectric ultrasonic device (OT7 insert) under abundant irrigation with sterile 0.9% NaCl	MOD.31W hand excavator
Gabanyi 2013	Third molar						Cleaned (not described)		
Gandia 2008	Third molar	DPSCs	18-21 y						
Gay 2014	Third molar	DPSCs			DMEM containing 10% FBS, 1% penicilin and streptomycin	72 h			
Giorgini 2011	Third molar	DPSCs	18-20 y				Cleaned (not described)	Cut around the CEJ by using sterilized dental fissure bur	

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
Govindasamy 2010 (a)	Third molar	DPSCs	24-35 y			2 h	Root surfaces were cleaned with PVP-I (Sigma Aldrich, St Louis, MO)		
Govindasamy 2011	Third molar	DPSCs	24 – 35 y		1 X DMEM-KO, 10% FBS, 2% penicillin,2% streptomycin, 5% GlutaMax, 100 mg/mL ascorbic acid, 1X ITS	2 h	Root surfaces were cleaned with 100% PVP-I (Sigma Aldrich, St Louis, MO)		
Govindasamy 2010 (b)		DPSCs	14–25 y						
Graziano 2008		SBP-DPSC	21–45 y				Petreated for a week with professional dental hygiene. Before extraction, the dental crown crown was covered with a 0,3% CHX (Forhans,New York, NY) for 2 min		Dentinal excavator or gracey curette
Graziano 2007		SBP-DPSC	25-45 y				Petreated for a week with professional dental hygiene. Before extraction, the dental crown crown was covered with a 0,3% CHX (Forhans,New York, NY) for 2 min		Dentinal excavator or gracey curette

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
Gronthos 2000	Impacted third molar	DPSCs	19-29 y				Cleaned (not described)	Cut around the CEJ by using sterilized dental fissure bur	
Han 2008	Third molar	DPSCs					Tooth was washed with PBS containing AA solution for 3 min after washing with 70% ethanol		
Han 2010	Third molar	DPSCs					Tooth was washed with PBS containing AA solution for 3 min after washing with 70% ethanol	Severed with pliers	
Haveleck 2013	Impacted third molar	DPSCs							
He 2008	Impacted third molar	DPSCs	19-29 y						
He 2013	Impacted third molar	hDPSCs	18-22 y						
He 2014	Third molar	hDPSCs	18-22 y						
Hilkens 2013	Third molar	DPSC-EZ DPSC-OG	15-20 y				Cleaned (not described)	Mechanically fractured with forceps	
Hirata 2010	Third molar upper						Cleaned (not described)	Cut around the CEJ by using sterilized dental fissure bur	Barbed broach
Hoss 2013	Impacted third molar	DPSCs					Brushed by using sterilized dental burs		
Huang 2009	Left upper central incisor - traumatized Left upper lateral incisor - traumatized	hDPSCs	41 y					Endodontic file	

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
Huang 2008	Supernumerary tooth (a mesiodens)	DPSCs	20 y	Male	DPBS, on ice (Invitrogen Carlsbad, CA, USA)		Cleaned with DPBS (Invitrogen, Carlsbad, CA, USA)	Sterile hand-held high-speed drill. Cut around the curcumference of the teeth with chinsel.	Endodontic file
Huang 2010 (a)	Third molar	DPSCs	14-22 y						
Huang 2010 (b)	Impacted third molar	DPSCs	16–24 y		Cell culture medium, serum-free				
Ishkitiev 2010	Upper third molar	DPSCs					Cleaned (not described)	Cut around the CEJ by using sterilized dental fissure bur	
Ishkitiev 2012	Third molar	DPSCs					Cleaned (not described)	Cut around the CEJ by using sterilized dental fissure bur	Sterile barbed broach
Jeon 2011	Impacted third molar	DPuSCs	16–18 y		D-PBS, on ice		Cleaning with DPBS containing 1% penicillin (Gibco), streptomycin (Gibco)		
Jin 2013	Third molar	hDPSCs	18-25 y						
Kadar 2009	Impacted third molar	DPSCs	18-26 y				Cleaned (not described)	Cut around the CEJ by using sterilized dental fissure bur sterilized dental fissure bur	
Kanafi 2014		DPSCs	18-40 y						
Kanafi 2013 (a)		DPSCs	5-40 y						
Kanafi 2013 (b)		DPSCs	5 - 40 y						
Kanafi 2013 (c)		DPSCs	5 - 40 y				Washed 2–3 times with DPBS (Invitrogen, Calif., USA)		

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Author and year of publication	*Tooth*	*Cell type*	*Age at the extraction*	*Sex*	*Storage method for transporting*	*Storage time*	*Hygiene*	*Methods section of tooth*	*Methods for removing the pulp from the interior of the chamber*
Kanafi (d)	Third molar	DPSCs	18-30 y						
Karaoz 2011	Third molar		17-25 y				Immerssion in pysiological solution containing antibiotics	Cut around the CEJ by using sterilized dental fissure bur	Sterile excavator
Karbanová 2010	Impacted third molar	DPSCs	17-23 y						
Karbanová 2011	Impacted third molar	DPSCs	17-23 y						
Kawanabe 2012	Premolar	DP	18-27 y					Cut using a dental turbine	
Kellner 2014	Impacted third molar	DPSCs	12-30 y						
Khanna-Jain 2012	Impacted third molar	DPSCs	Mean of 23 ± 2.5 years, 21–26 y						
Kim 2013	Third molar	hDPSCs							
Kim 2011	Premolar	DPSCs	18 and 19 y	Male			Cleaned (not described)	Cut around the CEJ using by sterilized dental disk	
Király 2011	Impacted third molar	DPSCs	19-35 y						
Király 2009	Impacted third molar	DPSCs	19–55 y						
Kolind 2014	Impacted third molar	DPSCs							
Koyama 2009	Impacted third molar		14-35 y				Cleaned (not described)	Cut around the CEJ by using sterilized dental fissure bur	
Kraft 2010	Impacted third molar	PDSC-immature PDSC-mature	21 y 20 y	Male Female					
Laino 2006	Third molar	DPSCs	19-37 y						Dentinal excavator or a gracey curette

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
Laino 2005		DPSCs	30-45 y				Pretreated a week before with professional dental hygiene. Before extraction, the dental crown was covered with 0.3% CHX gel (Forhans) for 2 min		Gracey curette
Lee 2014		DPSCs	18-39 y						
Lee 2008	Molar	hDPSCs					Cleaned (not described)	Cut around the CEJ by using sterilized dental fissure bur	
Lee 2011 (a)	Third molar	hDPSCs							
Lee 2011 (b)	Impacted third molar	hDPSCs	18-22 y					Forceps	
Lee 2011 (c)	Molar	DPSCs			HBBS (Welgene, Dae-gu, Korea) supplemented with 3% AA (Gibco, Grand Island, NY) at 4°C			Dental high-speed unit	
Lee 2010 (a)	Molar	hDPSCs							
Lee 2010 (b)	Third molar	DP-MSCs	17-38 y	Male Female			CHX solution	Hercules cutter	
Lee 2010 (c)	Premolar	DPSCs	18-30 y		Magnetically cryopreserved: Cryopreserved in a program freezer (ABI, Chiba, Japan) supplied with a slight magnetic field. Teeth were transported at4°C and then placed in a freezer at -5°C.	15 min	Cleaned with PBS		

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
					Then cooled at a rate of -0.5°C/min until -32°C. Then transferred to a freezer (MDF-11561; Sanyo, Osaka, Japan) and stored at -152°C. Non-cryopreserved fresh teeth	84 h			
Lee 2012	Incisor	DPSCs	28 y 25 y	Male Female			Cleaned with DPBS		
Lee 2011 (d)	Third molar	DPSCs	18-35 y						
Li 2011		hDPSCs	19-22 y	Male					
Lin 2011	Third molar	DPSCs	25 y 18 y	Male Female			Cleaned with PBS	Diamond burs	Forceps
Lindroos 2008	Impacted third molar	DPSCs	21-31 y						
Liu 2014		DPSCs							
Luo 2014 (a)		hDPSCs	18-25 y						
Luo 2014 (b)		hDPSCs							
Ma 2012	Third molar	DPSCs	18-28 y						

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
		CDPSCs							
Makino 2013	Supernumerary	SNTSCs							
Mangano 2010		DPSCs					Pretreatment for one week with professional dental hygiene. Before extraction dental crown covered with 0.3% CHX gel (Forhans, New York, NY) for 2 min		Dentinal excavator or a gracey curette
Mangano 2011	Third molar	DPSCs					Pretreatment for one week with professional dental hygiene. Before extraction dental crown covered with 0.3% CHX gel (Forhans, New York, NY) for 2 min		Dentinal excavator or a gracey curette
Manikandhan 2010		DPSCs				6-48 h			Brooches
Marchionni 2009	Molar	DP-SCs	Mean of 35 y				Immerssion d in PBS containing AA: 100 U/mL penicillin, 100µg/mL streptomycin, 0.25 µg/mL amphotericin B. Then, immerssion in a 0,2% CHX solution.	Sterile diamond bur	

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
Martens 2012	Third molar	hDPSCs	18-24 y					Mechanically	
Martin 2013	Third molar	hDPSCs	Mean of 22.5 y		DMEM (Sigma-Aldrich, Steinheim, Germany) supplemented with 6% AA : 10,000 U/mL penicillin (Sigma Aldrich) 10 ng/ mL streptomycin (Sigma-Aldrich) and 25 mg/mL amphotericin B (Sigma-Aldrich)	Immediately	Rinsed with PBS (Sigma-Aldrich). Periodontal tissues over the root surface were removed with a sterile surgical blade	Sterilized diamond bur	
Min 2011	Third molar	DPCs	20-25 y						
Mokry 2010	Impacted third molar	DPSCs	18-27 y	Male Female					
Mori 2011	Third molar	DPSCs					Cleaned (not described)	Cut around CEJ by using sterilized dental fissure bur	
Murakami 2013	Third molar	DPSCs	18-29 y						
Murakami 2012	Third molar	DPSCs	18-29 y						
Muthna 2010	Impacted third molar	DPSCs	12-23 y	Male Female					
Nadeem 2013	Third molar	DPSCs							
Nakamura 2009		DPSCs							
Nam 2011	Third molar	hDPSCs	19-25 y						
Navabazam 2013	Third molar	DPSCs	15-32 y						

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
Nawi 2013		DPSCs			HBSS	Immediately	Periodontal and gingival tissues were scrapped off from the tooth surface using sterile surgical blade. Surface was cleaned with iodone and 70% ethanol. Then, washed with DPBS	Cut a the CEJ by using hard tissue cutter (Exact, Düsseldorf, Germany)	Barbed roach (Dentslpy, Germany)
Nesti 2011	Molar	DPSCs	18-35 y						Gracey curette
Neuss 2008	Impacted third molar	DPSCs					Cleaned (not described)	Cut around the CEJ by using sterilized dental bur	
Niu 2014	Third molar	hDPSCs	18-25 y						
Oancea 2013		DPSCs	12-17 y				Pretreatment for a week with professional dental hygiene		Dentinal excavator or a gracey curette
Okamoto 2009	Third molar	DPSCs	22-26 y						
Osathanon 2011	Impacted third molar	DPSCs							
Osathanon 2014	Impacted third molar	DPSCs							
Paino 2010	Molar	DPSCs							
Palumbo 2013	Third molar	hDPSCs							
Pang 2013	Third molar	DPSCs	16-22 y						Barbed broach
Papaccio 2006		SBP-DPSCs	21-45 y				Pretreatment for a week with professional dental hygiene. Before extraction, the dental crown was covered with 0.3% CHX gel (Forhans, NY) for 2 min.		Dentinal excavator or a gracey curette

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Author and year of publication	*Tooth*	*Cell type*	*Age at the extraction*	*Sex*	*Storage method for transporting*	*Storage time*	*Hygiene*	*Methods section of tooth*	*Methods for removing the pulp from the interior of the chamber*
Park 2013	Impactec third molar	hDPSCs	18-35 y						
Patil 2014	Third molar	DPSCs	16-18 y	Male			Rinsed several times with DPBS containing 1% penicillin and 1% streptomycin	Bone forceps	
Pereira 2012 (a)	Molar	N-hDPSCs I-hDPSCs	17-43 y					Sterilized diamond discs and dental surgical elevator	Sterile dentinal excavator
Pereira 2012 (b)		DPSCs-N DPSCs-I	17-43 y					Sterilized diamond discs (KG Sorensen, ref. 7020, Barueri, São Paulo, Brazil) and dental surgical elevator	Sterile dentinal excavator
Perry 2008	Third molar	DPSCs	18-30 y		20mL of one of three collection/ transport solutions: HTS (BioLife Solutions, Bothell, WA) MesenCult basal medium (Stem Cell Technologies, Vancouver, Canada). PBS (Sigma Chemical, St. Louis, MO)	Immediately	Washed with sterile PBS, followed by immersion in 1% PVP-I for 2 min, immersion in 0.1% sodium thiosulfate in PBS for 1 min, and another wash in sterile PBS		
Picchi 2013	Molar								Gracey curette
Pierdomenic 2005	Molar	DP-MSCs	Mean of 40 y	Male Female		Immediately	Immerssion in physiological solution containing antibiotics	Bone forceps	

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Author and year of publication	*Tooth*	*Cell type*	*Age at the extraction*	*Sex*	*Storage method for transporting*	*Storage time*	*Hygiene*	*Methods section of tooth*	*Methods for removing the pulp from the interior of the chamber*
Ponnaiyan 2012	Impacted third molar	DPSCs	18-22 y						
Riccio 2010	Third molar	DPSCs	18-35 y						
Rizk 2013 (a)	Premolar	DPSCs						Bone cutter	
Risk 2013 (b)	Premolar	hDPSCs							
Rodríguez-Lozano 2012	Impacted third molar	DPSCs	Mean of 29 y, 21-45 y	Male Female					
Rodriguez-Lozano 2013	Impacted third molar	DPSCs			DMEM supplemented with 10 % of FCS, 100 U/ mL penicillin and 100 µg/mL streptomycin	24 h			
Ryu 2009	Molar	hDPSCs							
Sakai 2012	Third molar	DPSCs	18-30 y						
Schiraldi 2012	Third molar	DPSCs	21-45 y						
Seifrtova 2012	Impacted third molar	DPSCs							
Seifrtova 2013	Impacted third molar	DPSCs							
Seo 2013	Impacted third molar	DPSCs	20-28 y				Cleaned (not described)	Cut around CEJ by using sterilized diamond stones	
Shafiei 2014	Third molar	DPSCs	20-25 y					Cut around CEJ by using fissure bur	
Shekar 2012	Impacted third molar Premolar	DPSCs			α-MEM supplemented with antibiotics: 100 IU penicillin, antibiotics: 100 IU penicillin, 100 µg/ mL streptomycin, pH 7.27.4	Immadiately	Washing with DPBS	Chisel and mallet	Forceps and a spoon excavator (2 mm diameter)

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
Shi 2002	Impacted third molar	DPSCs							
Sollazzo 2011	Third molar	DPSCs	20-25 y						Gracey curette
Son 2006	Molar	hDPSCs					Cleaned (not described)	Cut around JEC by using sterilized dental fissure bur	
Spath 2010	Third molar	DPSCs	22-35 y				Treatment one week before extraction with professional dental hygiene. Before extraction, dental crowns were covered with a 0.3% CHX gel (Forhans, NY, USA) for 2 min		Dentinal excavator or a gracey curette
Stevens 2008	Premolar	hDPSCs							
Stokowski 2007	Impacted third molar	DPSCs	18-40 y				Cleaned (not described)	Vise	
Struys 2013	Third molar	DPSCs	16-19 y					Fractured mechanically	Forceps
Struys 2011	Third molar	DPSCs						Fractured mechanically	Forceps
Suchanek 2013	Semi-erupted third molar	DPSCs	23 y	Male	HBSS (Gibco, UK)	Immediately		Luer's forceps	Extirpation needle or tweezers
	Third molar		22 y	Female					

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Author and year of publication	*Tooth*	*Cell type*	*Age at the extraction*	*Sex*	*Storage method for transporting*	*Storage time*	*Hygiene*	*Methods section of tooth*	*Methods for removing the pulp from the interior of the chamber*
Suchanek 2007	Impacted third molar Third molar Premolar	DPSCs	15-23 y		HBSS (Gibco, Scotland)	Immediately		Skive or luer's Forceps	Excavator (Henry Schein Inc., UK) and sharp needle
Suchanek 2009	Third molar	DPSCs	12-23 y	Male Female	HBSS (Gibco, Scotland)	Immediately		Luer's Forceps	Extirpation needle or sharp excavator (Henry Schein, UK)
Suh 2014	Third molar	DPSCs	19-40 y					Cut around CEJ by using sterilized dental fissure bur	
Suri 2008	Premolar Molar	HDPCs						Chisel and mallet	Sterile spoon excavator and tweezers
Suzyki 2011	Third molar	DSCs	14 y 28 y	Male Female					
Tamaki 2013	Third molar	DPSCs	16-28 y					Cut around CEJ by using sterilized dental fissure bur	
Tammaro 2014	Impacted third molar	hDPSCs	18-22 y					Forceps	
Tandon 2010	Premolar	DPSCs				30 min	Rinse with 0.2% CHX for 60 sec		Barbed broach
Tirino 2012	Third molar	DPSCs					Treatment with professional dental hygiene. The dental crown is covered with 0.3% CHX gel for 2 min (Forhans)		Dentinal excavator or a gracey curette

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
Tomic 2011	Third molar	DP-MSCs							
Tom-Kun 2011		hDPSCs			Serum-free culture medium	Immediately			
Trubiani 2010	Premolar	DP-MSCs					Pretreatment for a week with professional dental hygiene. Rinsed four times in PBS containing penicillin and streptomycin.		Sterile dentinal excavator
Trubiani 2012	Premolar						Pretreatment for a week with professional dental hygiene. Rinsed four times in PBS containing penicillin and streptomycin.	Cylindrical diamond bur (Intensiv, Grancia, Switzerland) mounted on a high-speed handpiece (Bora L; Bien-Air, Bienne, Switzerland) with water-spray cooling	Sterile dentinal excavator
Trubiani 2007			24-30 y				Rinsed four times in PBS containing penicillin and streptomycin		
Uchiyama 2009	Third molar	DPSCs	37 and 42 y	Female				Hammer	
Um 2011	Third molar	DPSCs	20-24 y	Male					
Vandomme 2014	Third molar	DPSCs			Realized (not described)		Cleaned (not described)		

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Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
	Premolar								
Varga 2011	Impacted third molar	hDPSCs	Mean of 27 y	Male Female	Sterile physiologic saline with gentamicin (Lék, Slovenia)	Immadiately	Rinsed with PBS (Oxoid, GB)	Cut around CEJ by using Luer's forceps	Excavator
Vasandan 2014	Impacted third molar	DPSCs	17-28 y				Washed with DPBS (Gibco, Grand Island, NY, USA) containing AA		
Ventura 2007	Molar	DPhMSCs					Immerssion in physiological solution containing antibiotics	Bone forceps	
Vishwanath 2013		DPSCs	Less than 25 y		FBS	Immediately	Cleaned (not described)	Sterilized dental bur	Small size broach and a blunt non cutting forceps
Wada 2009	Premolar	DPSCs							
Wang 2010	Third molar	DPSCs	15-25 y						
Wang 2014	Impacted third molar	hDPSCs	19-28 y						
Wang 2012	First premolar	DPSCs	18-20 y						
Wang 2013 (a)		DPSCs	14-25 y						
Wang 2013 (b)	Premolar	DPSCs	12-13 y						

(Contd...)

(Contd...)

Author and year of publication	*Tooth*	*Cell type*	*Age at the extraction*	*Sex*	*Storage method for transporting*	*Storage time*	*Hygiene*	*Methods section of tooth*	*Methods for removing the pulp from the interior of the chamber*
Weszl 2012	Impacted third molar	DPSCs	18-26 y					Cut around CEJ by using sterile dental fissure bur	
Woods 2009	Third molar	DPSCs	15-30 y		PBS (Sigma Chemical, St. Louis, MO)	24 h	Washed with sterile saline, exposure to 1% PVP-I for 2 min, 0,1% sodium thiosulfate in PBS for 1 min and another wash in sterile PBS. Immersion in Listerine antiseptic (Johnson and Johnson Healthcare Products, Langhorne, PA) for 1 min, followed by several final washes in sterile PBS.		Curette
Yan 2010		DPSCs							

(Contd...)

(Contd...)

Author and year of publication	Tooth	Cell type	Age at the extraction	Sex	Storage method for transporting	Storage time	Hygiene	Methods section of tooth	Methods for removing the pulp from the interior of the chamber
Yu 2009	Premolar	DPSCs					Cleaned with dental soaler to remove attached soft tissue	Splitting the teeth at the CEJ sterile dental drill	
Zhai 2013	First premolar	hDPSCs							
Zhang 2011	Third molar	DPSCs	16 y	Female				Vise	
Zhang 2010	Third molar	DMCs	16 y	Male					
Zhang 2006	Impacted third molar	DPSCs	18-24 y		α-MEM (Gibco BRL, Life Technologies B.V. Breda, The Netherlands) 0.5 mg/mL of gentamicin (Gibco BRL), 3mg/mL amphotericin B (Gibco BRL)		Cleaned (not described)	Cut around the CEJ by using a high-speed dental drill	
Zhang 2008 (a)	Impacted third molar	DPSCs	22 y	Male					
Zhang 2008 (b)	Impacted third molar	DPSCs	22 y	Male					
Zhao 2011	Third molar	hDPSCs	18-35 y	Male Female					
Zhao 2006	Lower premolar	hDPSCs							
Zhou 2014	Impactec third molar	DPCs	18-30 y						

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