Indexed by : DOAJ Google Crossref Dimension sinta GARUDA

J.Biomed.Transl.Res
ISSN: 2503-2178

JOURNAL OF BIOMEDICINE AND TRANSLATIONAL RESEARCH

Available online at JBTR website: https://jbtr.fk.undip.ac.id

Original Research Article

The Association Between Intake of Saturated, Monounsaturated, and Polyunsaturated Fatty Acids with Bifidobacterium Abundance Among Obese Adults Without Metabolic Syndrome

Yuli Nugraeni[1*], Ranny Rachmawati[2], Akyla Oktriza Az Zahra[3], Adelia Adzra Nugraha[3]

[1]*Department of Conservative and Endodotic, Faculty of Dentistry, Universitas Brawijaya, Indonesia*
[2]*Department of Periodontics, Faculty of Dentistry, Universitas Brawijaya, Indonesia*
[3]*Faculty of Dentistry, Universitas Brawijaya, Indonesia*

Article Info
History
Received: 21 Mar 2025
Accepted: 31 Aug 2025
Available: 31 Aug 2025

Abstract

Background: *Dental Pulp Stem Cell* (DPSC) is one of the ideal *mesenchymal stem cells* (MSCs) for the repair of damaged cells, as it has great proliferative potential and the ability to regenerate pulp-like complex dentins. However, DPSC has limitations as a stem cell sources due to its small number. Induction with human umbilical cord cells (hUC-MSC) isolates is currently widely developed, considering its advantages such as being easy to obtain, having proliferative potential and anti-inflammatory effects. However, its influence and effectiveness on Dental Pulp Stem Cells is not widely known.

Objective: To prove the effect of umbilical cord cell isolate on the expression of Stro-1 and CD34 in *Dental Pulp Stem Cell* (DPSC) culture.

Methods: This study is experimental laboratory with simple random sampling which is divided into 4 groups, namely 2 control groups and 2 groups of umbilical cord isolate administration which are observed for 12 hours and 24 hours. Observation of expression by immunohistochemical staining was then calculated using Qupath software in 5 fields of view.

Results: The treatment group that was given umbilical cord cell isolate showed the highest increase in Stro-1 and CD34 expression at 24-hour observation compared to the untreated group.

Conclusion: There was an increase in the expression of Stro-1 and CD34 in Dental Pulp Stem Cell cultures after being given umbilical cord cell isolate.

Keywords: *Dental Pulp Stem Cell; Umbilical cord cell isolate; mesenchymal stem cells; Stro-1; CD34.*

Permalink/ DOI: https://doi.org/10.14710/jbtr.v11i2.26269

INTRODUCTION

The prevalence of pulp and periapical diseases in Indonesia is categorized as high. Based on the National Basic Health Research (Riskesdas) data from the Ministry of Health, pulp and periapical diseases occupy the 11th position out of all outpatient diseases in Indonesian hospitals with a total of 30.06%. Even in 2018, according to the results of basic health research in 2018, the largest proportion of dental problems in Indonesia was damaged/cavities/diseased teeth by 45.3%, which shows that the prevalence of pulp disease in Indonesia is categorized as high.[1]

Pulp is a soft tissue located in the center of the tooth and surrounded by enamel and dentin which are the tissues that form and support the teeth.[1] Dental pulp in pulp chambers comes from mesenchymal tissue and has various functions.[2] Dental complaints can be caused by fractures, chemical factors, microorganism infections, and thermal factors that will cause an inflammatory response.[3] If these complaints are not treated immediately, it will cause bacteria to enter the open pulp.[1]

*Corresponding author:
E-mail: *yulinugraeni@ub.ac.id*
(Yuli Nugraeni)

Pulp treatment is carried out to maintain the vitality of teeth damaged by caries or dental trauma, keep the tooth structure intact and maintain optimal pulp function, namely formative, sensory, nutritive and protective. Pulp is involved in the maintenance, support and advanced formation of dentin.[4]

In dentistry, treatment in the field of stem cell-based tissue regeneration is developing rapidly.[5] Although human tissue has limitations in regeneration, much research on stem cells has been conducted and the latest developments in stem cell research promise new medical therapeutic prospects in the practice of dentistry in the future.[6] Regenerative endodontics promise the restoration of pulp-dentin complexes in teeth with necrotic pulp.[7]

Mesenchymal stem cells (MSCs) are adult stem cells that are often used in stem therapy. MSCs have the ability to renew cells independently and have the ability to differentiate into a wide variety of cell types. MSCs are suitable for the development of regenerative medicine because of their multipotent ability, immunomodulator, and capacity to migrate to affected tissues and directly initiate tissue repair. The source of MCSs in humans can be isolated, one of which is from dental tissue, namely *Dental Pulp Stem cells* (DPSCs).[8]

Dental Pulp Stem cells (DPSCs) were first isolated from the third molar permanent tooth by Gronthos et al in 2000.[9] DPSCs isolated from pulp tissue showed characteristics such as mesenchymal stem cells considered to be ideal candidates for regenerating damaged dental pulp tissue due to their multipotent nature and high proliferation rate Research conducted by Murray, et al and Iohara, et al concluded that DPSCs have the ability to differentiate into odontoblasts and play a role in dentin regeneration. DPSCs are known to express several stem cell markers such as stromal-1 (STRO-1) which is known to be the best MSC marker and express CD34 but only <2% in number.[10]

Although DPSC is ideal to be a source of stem cells for pulp tissue regeneration, collecting human dental pulp cells is difficult due to some limitations; they need to be obtained from healthy dental pulp tissue, and the number is very small.[11] Therefore, researchers consider inducing with another source of MSC, that is *human Umbilical Cord Mesenchymal Stem Cell* (hUC-MSC), because it is easier to isolate, have anti-inflammatory effects, and has fewer ethical issues, as it is a biological waste that is no longer needed.[12,13,14]. When compared to other sources of MSC such as *Bone Marrow Mesenchymal Stem Cell* (BM-MSC), hUC-MSCs have more primitive cells so that they can differentiate into different cells, are multipotent, non-hematopoetic, can repair themselves and differentiate into other cells such as osteoblasts, odontoblasts, fatty tissue and chondroblast.[15]

This study was carried out by observing cultures to identify the expression of biomarkers Stro-1 and CD34[16] in a controlled manner. In vitro studies using cells were carried out first.[17]

The research combining DPSC with hUC-MSC has never conducted before, so researchers are interested in conducting this study to see whether or not the MSCs can help regenerate dental pulp tissue, by looking at the expression of Stro-1 and CD34.

MATERIALS AND METHODS

Research Design

This research is a laboratory expreriment and the treatment of all samples was carried out simultaneously and after a long period of treatment, observations were made using the *Postest Only Control Group Design*.

Umbilical Cord Isolation

The cord solution for placental collection media was made by adding 90 mL of deionized water to 10 mL of HBSS, then adding 100µg/mL gentamicin, 10 mM HEPES, and 3,7 g/L NaHCO3. The pH was adjusted to 7.4. The solution was then sterilized with a 0.2 µm filter, and stored at 4°C (for 1 week). As much as 25 ml of collagenase 0.5 mg/ml was prepared by mixing trypLE with collagenase (Worthington).

Cord blood was collected into a tube containing the anticoagulant CDP-A (Terumo, Japan). After the arteries and veins were removed, the remaining tissue and umbilical cord are cut into 5 cm sections and placed in the media (cord solution), and transferred to the laboratory at a cold temperature. Next, the umbilical cord is cleaned using sterile water or PBS.

The endothelial isolation of the vein on the umbilical cord was performed by inserting a cannula into the vein and then washed with FBS. Collagenase solution were put into the vein and the umbilical cord was gently massaged so that the endothelium, fibroblasts, and inner layer are detached. The detached cells were taken and placed in a 15 ml tube. The tube was then centrifuged at 2000 rpm for 3 minutes, and the cells pellet were placed in cell banker. The cells were stored at a temperature of -80°C. When the umbilical cord isolate will be used, it can be incubated at 37°C for 2-3 minutes, centrifuged at 800 rpm for 8 minutes, then the pellet is suspended in culture medium.

Collection of Dental Pulp Cells

Dental pulp cells were obtained from newly extracted caries-free intact teeth in less than 6 hours. Then, the teeth were soaked in a physiological solution of 0.9% NaCl or DMEM in a 15 ml tube, and stored in a box filled with ice. After that, it is transfered to a petri dish and disinfected with 0.2% chlorhexidine digluconate. The soft tissue attached to the tooth was removed with a scalpel knife, then the teeth were burred using a round bur to open the pulp chamber. The pulp tissue was taken using an extirpation needle, and put into a tube containing culture medium, namely *Hepes-buffered Dulbecco's Modified Eagle Medium* (HDMEM). The tube was then centrifuged at 1000g/min for 10 minutes, the supernatant is removed and the remaining pellets were dissolved again in the complete culture medium and stored in an incubator at 37°C [18].

Dental pulp culture

The dental pulp cells were cultured on several petri dishes in DMEM medium containing 2.5 g/l HEPES and 10% *fetal bovine serum* (FBS). The medium is changed after 72 hours to remove non-attached cells. After reaching confluency, the cells were dissociated using trypsin, expanded, counted, and transferred to a fresh culture medium. They were then collected into a 15 ml tube and centrifuged at 1000g/min for 10 minutes at

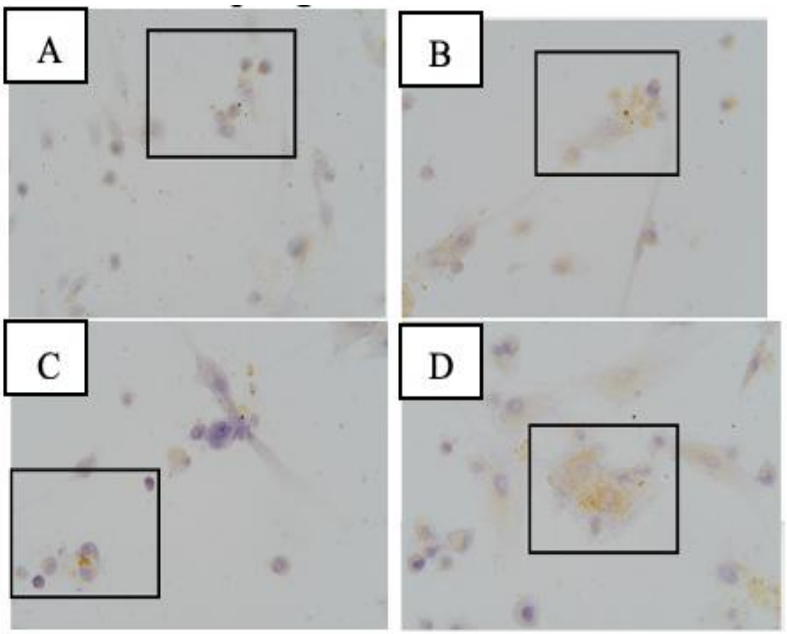

Figure 1. Stro-1 expression staining with 400x magnification (A) K(12 hour); (B) P(12 hour); (C) K(24 hour); (D) P(24 hour). Please provide Figures that have minimum 300 dpi in resolution

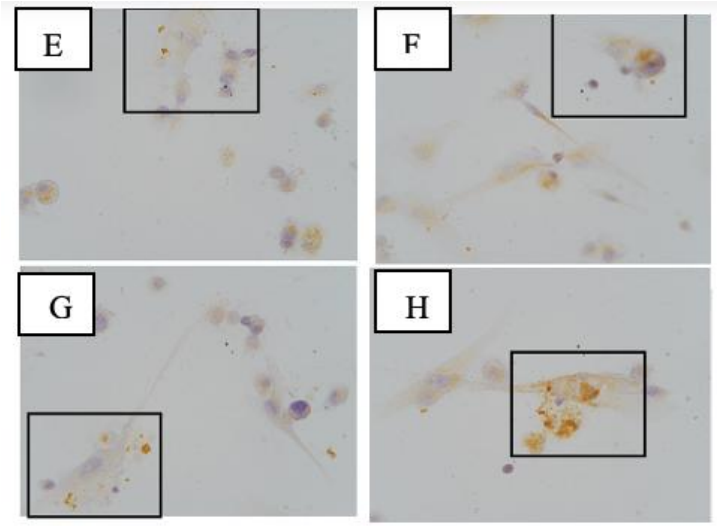

Figure 2. CD34 expression staining with 400x magnification (E) K(12 hour); (F) P(12 hour); (G) K(24 hour); (H) P(24 hour). Please provide Figures that have minimum 300 dpi in resolution

24℃. The supernatant is discarded, and the precipitated pellets are re-dissolved in a complete culture medium. The number of cells was then calculated using a hemocytometer under a Nikon Diaphot 300 microscope (4x/0.10 magnification). Furthermore, the pulp cells were recultured on a 24-well plate with the number of cells in each well being 2 x 10^5 cells. Cells wereallowed to multiply and would form colonies [19].

Stro-1 and CD34 Expression Tests

Immunoperoxidase against STRO-1 and CD34 cell culture preparations were washed with dH_20 twice for 5 minutes and incubated with PBS solution for 5 minutes. The preparation was placed in a glass box containing citrate buffer and then put into the autoclave for 15 minutes to optimize its antigenicity. It was then cooled at room temperature for 1 hour, and after drying for a while, the tissue is capped using a pap pen. The preparation was washed with dH_20 for 5 minutes and PBS for 5 minutes before incubating with 0.3% H_2O_2 for 15 minutes, rinsed with PBS pH 7.2 3x for 5 minutes each. Incubation with the blocking solution was carried out for 30 minutes, followed by overnight incubation at 4℃ with a Stro-1 monoclonal Mouse and CD34. The preparation was then rinsed with PBS pH 7.4 and incubate on a secondary, HRP-labeled IgG anti-mouse antibody for 1 h. Next, it was washed with PBS pH 7.4 and incubated on substrates for HRP and DAB for 5 minutes, washed again twice with PBS pH 7.4. After washing, it was counterstained with Mayer hematoxilen for 10 minutes, rinsed with dH_2O, dried and covered with coverglass. Positive expression of Stro-1 and CD34 is characterized by a brown color on the preparation. The image was calculated using Qupath software in 5 fields of view.

Table 1. Average expression of Stro-1 and CD 34

Group		Stro-1	Average	P-value	CD 34	Average	P-value
12 hour	K	0.365	0.296[a]	<0.001	0.235	0.231[a]	<0.001
		0.226			0.326		
		0.225			0.125		
	P	0.369	0.454[b]		0.239	0.485[b]	
		0.523			0.564		
		0.552			0.456		
		0.356			0.421		
24 hours	K	0.384	0.318[a,b]		0.498	0.385[a,b]	
		0.256			0.369		
		0.336			0.325		
		0.335			0.421		
	P	0.345	0.687[c]		0.425	0.790[c]	
		0.695			0.897		
		0.697			0.856		
		0.598			0.652		
		0.756			0.756		

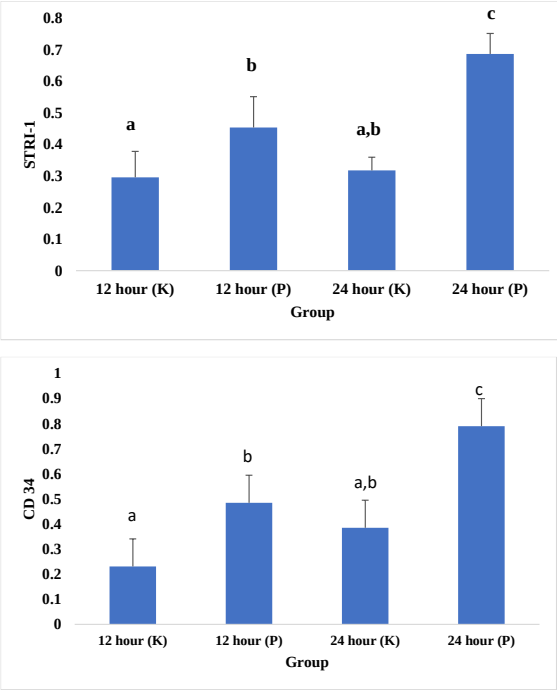

Figure 3. Graphic expression of Stro-1 and CD34.

Data Analysis

Data analysis was carried out using a program SPSS version 27 with a significance level or probability value of 0.05 (p=0.05) and a confidence level of 95% (α=0.05). Based on the Mann-Whitney test on Stro-1 expression data observed at 12 hours and 24 hours, p>0.05 showed that there was no significant difference in the average number of Stro-1 expression in *Dental Pulp Stem Cell* cultures given umbilical cord isolates. Based on the T-test of CD34 expression data observed at 12 hours, a p value of <0.05 indicated a significant difference in the average number of Stro-1 expressions in *Dental Pulp Stem Cell* cultures treated with umbilical cord isolates at 12 hours. Meanwhile, the T-test on CD34 expression data observed at 24 hours showed no significant difference in the average number of Stro-1 expressions in *Dental Pulp Stem Cell* cultures isolated from umbilical cord observed at 24 hours. The results of the Pearson correlation test that the expression of Stro-1 and CD34 in *Dental Pulp Stem Cell* cultures treated with umbilical cord cell isolate showed a correlation between the administration of umbilical cell isolate and the expression of Stro-1 and CD34 observed at 12 hours and 24 hours, meaning that the number of expressions produced after the administration of umbilical cord cell isolate will increase.

RESULTS

In this study, molar 3 tooth pulp cell cultures were randomly assigned to either the experimental (treatment) group, or the control group. Treatment group 1 was DPSC without administration of umbilical cord cell isolate, and treatment 2 was DPSC with administration of umbilical cord cell isolate. The groups were observed, and the number of Stro-1 and CD34 expressions were calculated after testing.

There are 4 groups in this study. The control group consisted of DPSC without administration of umbilical cord cell isolate, observed at 12 and 24 hours. Treatment group 1 consisted of DPSC with the administration of umbilical cord cell isolate, which was observed for 12 hours. Treatment group 2 consisted of DPSC with the administration of umbilical cord cell isolate, which was observed for 24 hours.

Preparation for the calculation of Stro-1 and CD34 was carried out using Qpath with immunohistochemical staining, then observations were made with Qupath software in 5 fields of view. The image of Stro-1 and CD34 expressions were presented in Figure 1 and 2, respectively.

The results of the calculation and average expression of Stro-1 and CD34 in the treatment group are presented in Table 1:

Based on figure 3 the results of the Stro-1 and CD34 expression graphs, it was shown that the 12-hour treatment group (K) (a) was significantly different from the 12-hour (P) (b) group ($p<0.05$), but not significantly different from the 24-hour group (K) (a,b) ($p>0.05$) while the 24-hour group (P) had significant differences with all treatment groups marked with notation (c).

DISCUSSION

The purpose of this study was to determine the effect of umbilical cord cell isolate on the expression of Stro-1 and CD34 in *Dental Pulp Stem Cells*. The *Dental Pulp Stem Cells* culture and umbilical cord isolate used in this study were processed in the Biomedical Laboratory, Faculty of Medicine, Universitas Brawijaya. The observations were done at 12 and 24 hours to determine cell proliferation. Observing at 12 and 24 hours provide enough intervals to see significant changes in cell proliferation without waiting too long.[20] Another study showed a decrease in proliferation after 48 hours.[21]

Stro-1 Expression in Groups Given Umbilical Cord Isolate Observed at 12 Hours and 24 Hours

The treatment group that was given umbilical cord isolate and observed at 12 hours showed no difference in the number of expressions compared to the untreated group. The treatment group that was given umbilical cord isolate observed at 24 hours showed a difference in the increase in the number of expressions compared to the group that was not treated. The administration of umbilical cord cell isolate in the Stro-1 group had the highest average expression in the treatment group, observed at 24 hours. In the first 12 hours, stable Stro-1 expression showed the ability of cells to maintain multipotent properties. After 24 hours, a significant change occurs, signaling a shift in the cell's potential differentiation. Although there was no increase in expression in the group observed at 12 hours, Stro-1 remained the best MSC marker.[22]

Naturally, MSCs express Stro-1. Stro-1 is a highly reliable marker for recognizing and isolating different types of MSCs, especially in dental tissue.[23] Expression of Stro-1 in DPSC shows that DPSC is a multipotent source of MSCs and can be differentiated into odontoblasts for pulp regeneration.[22] Research conducted by George *et al.*, in 2015 inoculated umbilical cord cell stem cells on the surface scaffold of human tooth roots, and the result was that the umbilical cord cells were attached to the root surface scaffold and the number of cell attachments increased significantly. These results showed that the umbilical cord cell isolate tends to differentiate into odontoblast-like cells.[24] Chen, et al in 2015 also conducted a study whose results showed that umbilical cord cell isolates can be induced to differentiate into odontoblast-like cells and dentin-like matrices. *Mesenchymal Stem cells* (MSCs) of the *umbilical cord* (hUC-MSC) have the ability to secrete various molecules paracrinally.[25] Increased Stro-1 expression can occur due to several factors, one of which is interaction with growth factors.

Growth factors are biologically active molecules that can be secreted and affect cell growth. Growth factors can act on specific cell surface receptors that then transmit their growth signaling molecules to other intracellular components and ultimately produce altered gene expression.[26] Odontoblasts-like cells originate from pulp cell progenitors that undergo differentiation. These progenitor cells induce molecular signals to induce the proliferation, migration, and differentiation of odontoblast-like cells. After the differentiated odontoblast occurs, there is a release of growth factors by the dentin matrix, which is a molecular signal for the process of reparative dentin formation by differentiated odontoblast-like cells. The growth factors that exist in both *dental pulp stem cells* and umbilical cord are TGF-β, which plays a role in proliferation, differentiation, and other cellular processes. Several growth factors, i.e. TGF–β, are directly involved in the differentiation of odontoblast cells.[27] TGF-β1 and TGF-β3 are growth factors that can regulate the formation of the dentin matrix by odontoblasts.[28]

In addition, mesenchymal umbilical cord growth factors such as BDNF (*Brain-Derived Neurotrophic Factor*), NGF (*Nerve Growth Factor*), and VEGF (*Vascular Endothelial Growth Factor*). These growth factors play an important role in the development and regeneration of mesenchymal cells, including *mesenchymal stem cells* (MSCs) that have the potential to replace damaged tissue. *Vascular Endothelial Growth Factor* (VEGF) itself has been used in therapy to support tissue regeneration, including in the use of DPSCs. VEGF can affect the growth and expression of biomarkers in DPSCs, indicating the potential of VEGF in facilitating cell differentiation and new tissue development.[29]

Expression of CD34 in Groups Given Umbilical Cord Isolate Observed at 12 Hours and 24 Hours

The treatment group that was given umbilical cord isolate observed at 12 hours and 24 hours showed a difference in the number of expressions compared to the group that was not treated. Administration of umbilical cord cell isolate in the CD 34 group had the highest average number of expressions in the treatment group observed at 24 hours. The results of these observations show that the number of cells proliferating increases. In previous studies, *Dental Pulp Stem Cells* (DPSC) naturally only slightly expressed CD34, which was <2%.[10]

Human Umbilical Cord Mesenchymal Stem Cell (hUC- MSC) is a rich source of Mesenchymal Stem Cell[30] also consists of hematopoietic cells that naturally express CD34.[31] Positive expression of CD34 indicates angiogenesis or the formation of new blood vessels. This is supported by research conducted by Zhang, et al, 2020 where hUC-MSC cultures form structures such as blood vessels and regenerate tissues such as dental pulp. Angiogenesis is the beginning of the process of tissue regeneration in the pulp.[32] The mechanism of pulp regeneration begins with the presence of pulp cells that can release angiogenic and dentinogenesis factors as a tissue repair process.[33] An increase in CD34 expression can occur due to several factors, one of which is the interaction with growth factors.

The interaction between stem cells in umbilical cord isolates and DPSCs can stimulate increased CD34 expression through the transfer of growth factors or signals because odontoblasts also produce cytokines as paracrine signals to recruit immune system cells by suppressing the secretion of inflammatory factors, reducing inflammation and oxidative stress, thereby suppressing cell apoptosis.[14] MSC paracrination is a condition in which MSC secretes certain molecules related to the regeneration process such as angiogenesis molecules, namely *vascular endhothelial growth factor* (VEGF), *fibroblast growth factor-2* (FGF-2), *angiopoietin-1* (Ang-1), and proliferation and differentiation molecules, namely *platelet-derived growth factor* (PDGF), insulin like growth factor (IGF).[25] The results of the correlation test showed a strong correlation, indicating that the administration of umbilical cord cell isolate could increase the expression of each marker, namely Stro-1 and CD34.

CONCLUSION

The administration of umbilical cord isolate showed that it affected the expression of markers in *Dental Pulp Stem Cell*. From the data obtained, the Stro-1 marker experienced an increase in expression levels at 24-hour observations. Meanwhile, the CD34 marker experienced an increase in the number of expressions at 12 hours and 24 hours of observation. The Stro-1 and CD34 markers experienced the highest number of expressions increases at 24-hour observations.

Further research is needed with longer observation times of 36 hours and 72 hours, as the 12-hour observation period has not revealed any significant differences. Additionally, more in-depth research is required to investigate the effect of increasing markers on *mesenchymal stem cells*. In addition, further research is also needed to see the interactions between cells that cause increased expression of markers and what growth factors play a role in the differentiation of odontoblasts. It is hoped that in future research, the isolation of umbilical cord cells can be carried out using cell culture.

ACKNOWLEDGMENTS

The researcher would like to express his deepest gratitude to all authors who contributed to the writing of this manuscript, as well as to all related parties in the Faculty of Dentistry who have provided moral and spiritual support. This research was awarded a research grant by the PNBP Faculty of Dentistry, Brawijaya University.

REFERENCES

1. Dewiyani, S. Distribusi Frekuensi Pulpitis Reversibel Dan Pulpitis Ireversibel Di Rsgm Fkg Moestopo (B) Tahun 2014-2016 (Berdasarkan Jenis Kelamin, Usia dan Lokasi Gigi). *J. Ilm. dan Teknol. Kedokt. Gigi* 15, 41 (2019). DOI:10.32509/jitekgi.v15i2.892
2. Arvetona, A. V. & Wirya Pratama, I. W. A. Prevalensi Pulpitis Reversible di Poliklinik Gigi Puskesmas. *JPP (Jurnal Kesehat. Poltekkes Palembang)* 19, 154–159 (2024). DOI:10.36086/jpp.v19i2.2416
3. Sugiaman, V. K. Peranan Sel Punca dalam Meregenerasi Jaringan Pulpa Gigi. 4 (1), 43-51 (2019). DOI:10.28932/sod.v4i1.1770
4. Kartinawanti, A. T. & Asy'ari, A. K. Penyakit Pulpa Dan Perawatan Saluran Akar Satu Kali Kunjungan: Literature Review. *J. Ilmu Kedokt. Gigi* 4, 64–72 (2021). ISSN 2580-0523
5. Abdullah, K. Identifikasi biomarker protein membran cd105 pada sel punca pulpa molar tiga dengan metode flow cytometry. *SKRIPSI-2018* (2022).
6. Hakim, R. F. Stem Sel dan Regenerasi Jaringan Dalam Bidang Kedokteran Gigi. *Stem Sel dan Regen. Jar. Dalam Bid. Kedokt. Gigi* (2020) doi:10.52574/SYIAHKUALAUNIVERSITYPRESS.321.
7. Gutmann, J. L. Grossman's Endodontic Practice – 13th Edition. *J. Conserv. Dent.* 19, 494 (2016). doi: 10.4103/0972-0707.190011
8. Narmada, I. B., Ernawati, D. S. & Prahasanti, C. Potensi Human Dental Pulp Stem Cells (HDPSCS) Untuk Regenerasi Defek Kelenjar Saliva Tikus Wistar. (2018). DOI: 10.3389/fcell.2023.1196023
9. Intanries, M. Isolasi dan kultur sel pulpa gigi dengan kombinasi teknik outgrowth dan enzim disagregasi (Laporan penelitian). *SKRIPSI-2015* (2016).
10. Anggrarista, K. A., Cecilia, P., Nagoro, A. A., Saskianti, T. & Surboyo, M. D. SHED, PRF, and Chitosan as Three-Dimensional of Tissue Engineering for Dental Pulp Regeneration. *Dent. Hypotheses* 12, 43–46 (2021). DOI:10.4103/denthyp.denthyp_121_20
11. Hadi, R. S. & Kusuma, I. Evaluasi Kemampuan Proliferasi, Diferensiasi dan Viabilitas Sel Punca Asal Pulpa Gigi Manusia Pasca Vitrifikasi. (2019) doi:10.33476/MKP.V11I1.951.
12. Li, T., Xia, M., Gao, Y., Chen, Y. & Xu, Y. Human umbilical cord mesenchymal stem cells: an overview of their potential in cell-based therapy. *Expert Opin. Biol. Ther.* 15, 1293–1306 (2015). DOI: 10.1517/14712598.2015.1051528
13. Nike Hendrijantini, -. Potensi Human Umbilical Cord Mesenchymal Stem Cells (hUCMSCs)Pada Terapi Regenerasi Tulang Mandibula Osteoporosis Dengan Indikator Peningkatan Ekspresi tgf-p1, runx 2, dan Jumlah Sel Osteoblas Tahun 2 Dari Rencana 2 Tahun. (2018).
14. Yao, J. *et al.* Extracellular vesicles derived from human umbilical cord mesenchymal stem cells alleviate rat hepatic ischemia-reperfusion injury by suppressing oxidative stress and neutrophil inflammatory response. *FASEB J.* 33, 1695–1710 (2019). DOI: 10.1096/fj.201800131RR
15. Zarrabi, M., Mousavi, S. H., Abroun, S. & Sadeghi, B. Potential Uses for Cord Blood Mesenchymal Stem Cells. *Cell J.* 15, 274 (2013). PMID: 24381851
16. Rosdiana, A. & Hadisaputri, E. Review Artikel: Studi Pustaka Tentang Prosedur Kultur Sel. *Farmaka* 14, 236–249 (2016).
17. Khumairoh, I. & Puspitasari, I. M. Kultur Sel. *Farmaka* Suplemen V, 98–110 (2016).

18. Dutra, A., Camassola, M. & Borba, P. De. Isolation and characterization of dental pulp stem cells from permanent third molars. 22, 153-157 (2016). (ISSN: 1545-1003).
19. Lampiasi, N. The Migration and the Fate of Dental Pulp Stem Cells. *Biology (Basel).* 12, 742 (2023). doi.org/10.3390/biology12050742
20. Sugawara, H. *et al.* A highly sensitive strategy for monitoring real-time proliferation of targeted cell types in vivo. *Nat. Commun. 2023 141* 14, 1–14 (2023).
21. Rizqi, J. & Amestiasih, T. PENGARUH KOMBINASI MADU DAN LIDAH BUAYA (ALOE VERA) TERHADAP PROLIFERRASI PADA SEL LINE FIBROBLAST NIH3T3: STUDI IN VITRO. (2021).
22. Nugraeni, Y., Riawan, W., Permatasari, N., Widjajanto, E. & Dradjat, R. S. Modulation of endogenous stem cells markers in the periodontal ligament of dental rat post traumatic avulsion following Aloe vera gel exposure. *Med. Plants* 9, 225–230 (2017). DOI:10.5958/0975-6892.2017.00036.3
23. Ning, H., Lin, G., Lue, T. F. & Lin, C. S. Mesenchymal stem cell marker Stro-1 is a 75 kd endothelial antigen. *Biochem. Biophys. Res. Commun.* 413, 353–357 (2011). DOI: 10.1016/j.bbrc.2011.08.104
24. George, J. P., Chakravarty, P., Chowdhary, K. Y., Purushothama, H. & Rao, J. A. Attachment and Differentiation of Human Umbilical Cord Stem Cells on to the Tooth Root Surface with and without the Use of Fibroblast Growth Factor-An In Vitro Study. *Int. J. stem cells* 8, 90–98 (2015). DOI: 10.15283/ijsc.2015.8.1.90
25. Putra, A. Basic Molecular Stem Cell. *Basic Mol. stem cell* 236 (2019).
26. Stone, W. L., Leavitt, L. & Varacallo, M. A. Physiology, Growth Factor. *StatPearls* (2023). PMID: 28723053
27. Lubis, D. S. H. R. L. Pengaruh Kultur Statis (2D) dan Kultur Dinamis (3D) terhadap Proliferasi Sel, Kadar Total Protein Conditioned Medium, dan Sitokin TGF-β pada Sel Punca Mesenkimal Asal Tali Pusat = Effects of Static Culture (2D) and Dynamic Culture (3D) on Cell Proliferati. (2021). URI: https://lib.ui.ac.id/detail?id=20512982&lokasi=lokal
28. Majaya, M. Ekspresi Dentin Matrix Protein-1 (DMP-1) Setelah Aplikasi Pasta Cangkang Telur Ayam Ras (*Gallus sp*) Pada Pulpa Gigi Kelinci New Zealand (*Oryctolagus cuniculus*) Yang Terin. *Tesis* 33, 1–12 (2022).
29. Mullins, E., Forgues, C. & Kingsley, K. Effects of Vascular Endothelial Growth Factor (VEGF) on Dental Pulp Stem Cells (DPSC). *Curr. Res. Med.* 9, 1–8 (2019). DOI: 10.3844/amjsp.2019.1.8
30. 30. Joanna, N. Pemberian *Human Umbilical Cord Mesenchymal Stem Cells* (hUCMSCs) Dalam Meningkatkan Jumlah Osteoblas Pada Model Peri-Implantitis (+CD). (2023).
31. Naeim, F., Naeim, F. P. N. & Song, S. X. . *Atlas of Hematopathology*. (Academic Press, 2018).
32. Ariesdyanata, C. *et al.* Perbedaan Angiogenesis pada Pulpa setelah Aplikasi Ekstrak Propolis dan Kalsium Hidroksida. *Conserv. Dent. J.* 9, 48–53 (2019). doi.org/10.20473/cdj.v9i1.2019.48-53
33. Elline, Ismiyatin, K. & Budhi, T. I. Potensi Regenerasi Kompleks Dentin Pulpa Menggunakan Scaffold Hidrogel - Universitas Airlangga Official Website. (2022).