

**LEMBAR
HASIL PENILAIAN SEJAWAT SEBIDANG ATAU *PEER REVIEW*
KARYA ILMIAH : JURNAL ILMIAH**

Judul Jurnal Ilmiah (Artikel)	HUMAN BONE MARROW-DERIVED MESENCHYMAL CELL REACTIONS TO 316L STAINLESS STEEL: AN IN VITRO STUDY ON CELL VIABILITY AND INTERLEUKIN-6 EXPRESSION		
Jumlah Penulis	6 orang (Iwan Budiwan Anwar, Asep Santoso, Eko Saputra, <u>Rifky Ismail</u> , J. Jamari, Emile Van der Heide)		
Status Pengusul	Penulis Keempat		
Identitas Jurnal Ilmiah	<p>a. Nama Jurnal : Advanced Pharmaceutical Bulletin b. Nomor ISBN/ISSN : 22285881, 22517308 c. Vol, No., Bln Thn : Volume 7 (2), Juni 2017 d. Penerbit : Tabriz University of Medical Sciences e. DOI artikel (jika ada) : 10.15171/apb.2017.040 f. Alamat web jurnal : https://apb.tbzmed.ac.ir/ Alamat Artikel : https://apb.tbzmed.ac.ir/Article/APB_19332_20170425113627</p>		
Scopus Link	https://www.scopus.com/record/display.uri?eid=2-s2.0-85021405698&origin=resultslist&sort=plff&src=s&st1=ismail&st2=rifky&nlo=1&nlr=20&nls=count-f&sid=13d4c2cccd319c03b14caaf2e5d0d8109&sot=anl&sdt=aut&sl=34&s=AU-ID%28%22Ismail%2c+Rifky%22+35183057200%29&relpos=0&citeCnt=0&searchTerm=(SJR%200.632)%20(Q2)		
g. Terindex	(SJR 0,632) (Q2)		
SJR Link	http://www.scimagojr.com/journalsearch.php?q=21100232413&tip=sid		

Hasil Penilaian Peer Review :

Komponen yang dinilai	Nilai Reviewer		Nilai rata-rata
	Reviewer 1	Reviewer 2	
a. Kelengkapan unsur isi jurnal (10%)	4,00	4,00	4,00
b. Ruang lingkup dan kedalaman pembahasan (30%)	12,00	12,00	12,00
c. Kecukupan dan kemutahiran data/informasi dan metodologi (30%)	11,00	11,00	11,00
d. Kelengkapan unsur dan kualitas terbitan/jurnal (30%)	12,00	12,00	12,00
Total = (100%)	39,00	39,00	39,00
Nilai Pengusul = (40% x 39,00) / 5 = 3,12			

Reviewer 1

Prof. Dr. Ir. A.P. Bayuseno, MSc
NIP. 196205201989021001

Unit Kerja : Departemen Teknik Mesin FT UNDIP

Semarang, 11 Maret 2021

Reviewer 2

Dr. Agus Suprihanto, ST., MT
NIP. 197108181997021001

Unit Kerja : Departemen Teknik Mesin FT UNDIP

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KARYA ILMIAH : JURNAL ILMIAH**

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Status Pengusul	:	Penulis Keempat
Identitas Jurnal Ilmiah	a.	Nama Jurnal
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	c.	Vol. No., Bln Thn
	d.	Penerbit
	e.	DOI artikel (jika ada)
	f.	Alamat web jurnal
		Alamat Artikel
		Scopus Link
	g.	Terindex
		SJR Link

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- Jurnal Ilmiah Internasional Bereputasi
 Jurnal Ilmiah Internasional
 Jurnal Ilmiah Nasional Terakreditasi
 Jurnal Ilmiah Nasional Tidak Terakreditasi

Hasil Penilaian *Peer Review* :

Komponen Yang Dinilai	Nilai Maksimal Jurnal Ilmiah				Nilai Akhir Yang Diperoleh
	Internasional Bereputasi	Internasional	Nasional Terakreditasi	Nasional Tidak Terakreditasi	
a. Kelengkapan unsur isi jurnal (10%)	4,00				4,00
b. Ruang lingkup dan kedalaman pembahasan (30%)	12,00				12,00
c. Kecukupan dan kemutahiran data/informasi dan metodologi (30%)	12,00				11,00
d. Kelengkapan unsur dan kualitas terbitan/jurnal (30%)	12,00				12,00
Total = (100%)	40,00				39,00
Nilai Pengusul (kontribusi pengusul penulis) = (40% x 39,00) / 5 = 3,12					

Catatan Penilaian artikel oleh Reviewer :

1. Kesesuaian dan kelengkapan unsur isi jurnal:

Artikel ditulis dengan lengkap sesuai dengan panduan penulisan dari Jurnal *Advanced Pharmaceutical Bulletin*. Abstrak, Pendahuluan, Material dan Metode, Hasil Pembahasan, Kesimpulan, Daftar Pustaka dipresentasikan dengan baik

2. Ruang lingkup dan kedalaman pembahasan:

Viabilitas sel dan interleukin-6 dari kultur hBMC setelah pengobatan dengan implan 316L-SS dibahas cukup mendalam dan detil. Penemuan penelitian bahwa material 316L-SS tidak menimbulkan pengaruh racun dalam tubuh dibahas dengan lengkap.

3. Kecukupan dan kemutahiran data/informasi dan metodologi:

Artikel ini meskipun terdiri dari 4 halaman, membahas hal-hal terkini, yaitu isu biokompatibilitas. Hanya 2 dari 7 artikel ini yang tidak berasal dari artikel terkini. Meskipun demikian itu, metodologi penelitian dipresentasikan sangat baik sehingga menghasilkan kesimpulan yang relevan. Turnitin similarity index juga cukup rendah yaitu sebesar 14%.

4. Kelengkapan unsur dan kualitas terbitan:

Kelengkapan unsur penerbit cukup baik dan terindeks Scopus. SJR juga cukup tinggi sebesar 0,632.

Semarang, 8 Maret 2021

Reviewer 1

Prof. Dr. Ir. A.F. Bayuseno, Msc

NIP. 196205201989021001

Unit Kerja : Departemen Teknik Mesin FT UNDIP

**LEMBAR
HASIL PENILAIAN SEJAWAT SEBIDANG ATAU PEER REVIEW
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Jumlah Penulis	:	6 orang (Iwan Budawan Anwar, Asep Santoso, Eko Saputra, Rifky Ismail , J. Jamari, Emile Van der Heide)
Status Pengusul	:	Penulis Keempat
Identitas Jurnal Ilmiah	a.	Nama Jurnal
	b.	Nomor ISBN/ISSN
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	d.	Penerbit
	e.	DOI artikel (jika ada)
	f.	Alamat web jurnal
		Alamat Artikel
		Scopus Link
	g.	Terindex
		SJR Link
Kategori Publikasi Jurnal Ilmiah (beri √ pada kategori yang tepat)	:	<input checked="" type="checkbox"/> Jurnal Ilmiah Internasional Bereputasi <input type="checkbox"/> Jurnal Ilmiah Internasional <input type="checkbox"/> Jurnal Ilmiah Nasional Terakreditasi <input type="checkbox"/> Jurnal Ilmiah Nasional Tidak Terakreditasi

Hasil Penilaian *Peer Review* :

Komponen Yang Dinilai	Nilai Maksimal Jurnal Ilmiah				Nilai Akhir Yang Diperoleh
	Internasional Bereputasi	Internasional	Nasional Terakreditasi	Nasional Tidak Terakreditasi	
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b. Ruang lingkup dan kedalaman pembahasan (30%)	12,00				12,00
c. Kecukupan dan kemutahiran data/informasi dan metodologi (30%)	12,00				11,00
d. Kelengkapan unsur dan kualitas terbitan/jurnal (30%)	12,00				12,00
Total = (100%)	40,00				39,00
Nilai Pengusul (kontribusi pengusul penulis) = (40% x 39,00) / 5 = 3,12					

Catatan Penilaian artikel oleh Reviewer :

1. **Kesesuaian dan kelengkapan unsur isi jurnal:**

Artikel yang disajikan sudah sesuai dengan *Guide for Author* dari *Advanced Pharmaceutical Bulletin*. Isi dari bagian-bagian seperti Pendahuluan, Material dan Method, Hasil dan Pembahasan, Kesimpulan, Daftar Pustaka ditulis dengan sangat baik. Topik yang dibahas sesuai dengan bidang pengusul, yaitu Teknik Mesin.

2. **Ruang lingkup dan kedalaman pembahasan:**

Ruang linkup artikel ini adalah tentang apakah 316L-SS berbahaya bagi tubuh apa tidak. Ketika dijadikan implant. Hasil penelitian dibahas dengan baik dalam artikel ini. Selain itu, tata bahasa yang digunakan juga disajikan dengan baik.

3. **Kecukupan dan kemutahiran data/informasi dan metodologi:**

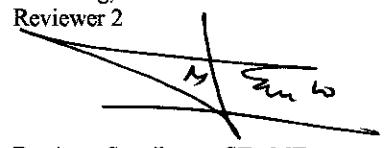
Kebaruan artikel sangat baik. Meskipun demikian, jumlah daftar pustaka sangat sedikit, hanya 7 untuk kategori jurnal internasional. Meskipun demikian, 80% pustaka merupakan terbitan terkini kurang dari 10 tahun. Metodologi yang digunakan sangat sistematis dengan tahapan-tahapan kegiatan yang jelas dan runut. Hasil-hasil simulasi juga disajikan secara menarik dan jelas (kontur, grafik) dengan analisis yang mendalam. Turnitin similarity index = 14%.

4. **Kelengkapan unsur dan kualitas terbitan:**

Jurnal ini telah terindex oleh database ternama Scopus. SJR sebesar 0,632.

Semarang, 11 Maret 2021

Reviewer 2


Dr. Agus Suprihanto, ST., MT

NIP. 197108181997021001

Unit Kerja : Departemen Teknik Mesin FT UNDIP



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Advanced Pharmaceutical Bulletin [Open Access](#)

Volume 7, Issue 2, 1 June 2017, Pages 335-338

Human bone marrow-derived mesenchymal cell reactions to 316L stainless steel: An in vitro study on cell viability and interleukin-6 expression (Article)

(Open Access)

Anwar, I.B.^{a,b}✉, Santoso, A.^b, Saputra, E.^{a,c}, Ismail, R.^c, Jamari, J.^c, van der Heide, E.^a

^aLaboratory for Surface Technology and Tribology, Faculty of Engineering Technology, University of Twente Drienerlolaan, 5, Postbox 217, Enschede, 7500 AE, Netherlands

^bOrthopaedic and Traumatology Department, Prof. Dr. R. Soeharso Orthopaedic Hospital, Jl. A. Yani Pabelan, Surakarta, 57162, Indonesia

^cLaboratory for Engineering Design and Tribology, Department of Mechanical Engineering, Diponegoro University, Jl. Prof. Soedharto, Tembalang, Semarang, 50275, Indonesia

Abstract

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Purpose: Human bone marrow-derived mesenchymal cell (hBMC) reactions to 316L stainless steel (316L-SS) have never been evaluated. The objective of this study was to assess cell viability and interleukin-6 expression of hBMC cultures upon treatment with a 316L-SS implant. Methods: A cytotoxicity analysis was conducted with a 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium (MTT) assay after a period of 24, 48 and 72 hours of incubation. Expression of interleukin-6 was measured using enzyme-linked immunosorbent assay (ELISA). Results: Cell viability measurement was performed via IC₅₀ formula. All treatment group showed a > 50 % cell viability with a range of 56,5 - 96,9 % at 24 hours, 51,8-77,3% at 48 hours and 70,1- 120 % at 72 hours. Interleukin-6 expression was downregulated subsequent to treatment with 316L-SS compared to the control group. Conclusion: We found that 316L-SS did not exhibit toxicity towards hBMC culture. © 2017 The Authors.

SciVal Topic Prominence

Topic: Tooth Avulsion | Autografting | Root Resorption

Prominence percentile: 79.538

Author keywords

316L stainless steel, Cell viability, Human bone marrow-derived mesenchymal cells, Interleukin-6

Indexed keywords

EMTREE drug terms: interleukin 6, stainless steel

EMTREE medical terms:

Article, bone marrow derived mesenchymal stem cell, cell viability, controlled study, cytotoxicity test, down regulation, enzyme linked immunosorbent assay, human, human cell, IC50, in vitro study, MTT assay, protein expression, reaction analysis, stainless steel implant, stem cell culture

Metrics [View all metrics](#)

3 Citations in Scopus

57th percentile

0.61 Field-Weighted

Citation Impact



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Usage, Captures, Mentions, Social Media and Citations beyond Scopus.

Cited by 3 documents

Computational Analysis of Different Designed Hip Joint Prostheses Using Finite Element Method

Taqriban, R.B., Ismail, R., Jamari, J. (2020) *7th International Conference on Information Technology, Computer, and Electrical Engineering, ICITACEE 2020 - Proceedings*

Study of Sensor Deflection in Hip Simulator: Numerical and Experimental Method

Wicaksono, F.S., Towijaya, T., Saputra, E. (2018) *E3S Web of Conferences*

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A review on surface treatment of stainless steel orthopedic implants
(2016) *International Journal of Pharmaceutical Sciences Review and Research*, 36 (1), art. no. 33, pp. 190-194. Cited 8 times.
<http://globalresearchonline.net/journalcontents/v36-1/33.pdf>
- 2 Dimić, I., Cvijović-Alagić, I., Obradović, N., Petrović, J., Putić, S., Rakin, M., Bugarski, B.
In vitro biocompatibility assessment of Co-Cr-Mo dental cast alloy ([Open Access](#))
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doi: 10.2298/JSC150505070M
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IL-6-dependent PGE2 secretion by mesenchymal stem cells inhibits local inflammation in experimental arthritis ([Open Access](#))
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<http://www.plosone.org/article/fetchObjectAttachment.action?uri=info%3Adoi%2F10.1371%2Fjournal.pone.0014247&representation=PDF>
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Immunobiology of mesenchymal stem cells ([Open Access](#))
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doi: 10.1038/cdd.2013.158
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Cytotoxicity difference of 316L stainless steel and titanium reconstruction plate
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Biological behaviour of human umbilical artery smooth muscle cell grown on nickel-free and nickel-containing stainless steel for stent implantation ([Open Access](#))
(2016) *Scientific Reports*, 6, art. no. 18762. Cited 6 times.
www.nature.com/srep/index.html
doi: 10.1038/srep18762
View at Publisher
- 7 Martinesi, M., Bruni, S., Stio, M., Treves, C., Bacci, T., Borgioli, F.
Biocompatibility evaluation of surface-treated AISI 316L austenitic stainless steel in human cell cultures ([Open Access](#))
(2007) *Journal of Biomedical Materials Research - Part A*, 80 (1), pp. 131-145. Cited 25 times.
doi: 10.1002/jbm.a.30846
View at Publisher

MicroRNA-301a inhibition enhances the immunomodulatory functions of adipose-derived mesenchymal stem cells by induction of macrophage M2 polarization

Hsu, L.-W. , Huang, K.-T. , Nakano, T.
(2020) *International Journal of Immunopathology and Pharmacology*

Topographical cues regulate the crosstalk between MSCs and macrophages

Vallés, G. , Bensiamar, F. , Crespo, L.
(2015) *Biomaterials*

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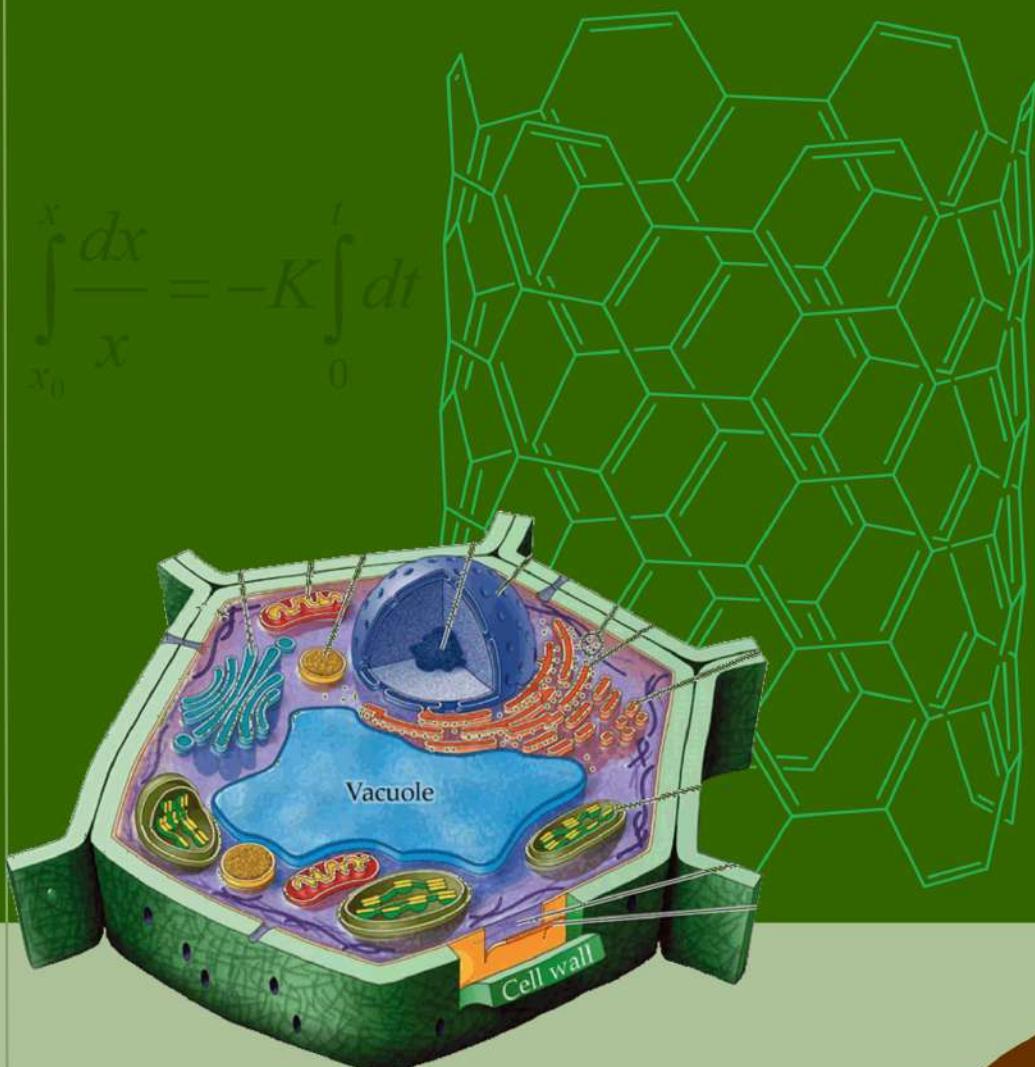
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Research Article

Spectroscopic and Spectrometric Methods Used for the Screening of Certain Herbal Food Supplements Suspected of Adulteration

(/Article/APB_15898_20161205152824)

Cristina Mateescu, Anca Mihaela Popescu*, Gabriel Lucian Radu, Tatiana Onisei, Adina Elena Raducanu

Adv Pharm Bull. 2017;7(2): 251-259. doi: 10.15171/apb.2017.030 (<https://doi.org/10.15171/apb.2017.030>)

PMCID: PMC5527239 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC5527239>) **PMID:** 28761827

(<http://www.ncbi.nlm.nih.gov/pubmed/28761827>) **Scopus ID:** 85021391660

(<https://www.scopus.com/inward/record.url?partnerID=HzOxMe3b&scp=85021391660&origin=inward>)

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Research Article

Formulation of Menthol-Loaded Nanostructured Lipid Carriers to Enhance Its Antimicrobial Activity for Food Preservation

(/Article/APB_304_20161212212610)

Parizad Piran, Hossein Samadi Kafil, Saeed Ghanbarzadeh, Rezvan Safdari, Hamed Hamishehkar*

Adv Pharm Bull. 2017;7(2): 261-268. doi: 10.15171/apb.2017.031 (<https://doi.org/10.15171/apb.2017.031>)

PMCID: PMC5527240 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC5527240>) **PMID:** 28761828

(<http://www.ncbi.nlm.nih.gov/pubmed/28761828>) **Scopus ID:** 85021405803

(<https://www.scopus.com/inward/record.url?partnerID=HzOxMe3b&scp=85021405803&origin=inward>)

 Article (/Article/APB_304_20161212212610)

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Scopus 10 (<https://www.scopus.com/inward/citedby.url?partnerID=HzOxMe3b&scp=85021405803&origin=inward>)

PubMed 0 (http://www.ncbi.nlm.nih.gov/pmc/?linkname=pubmed_pmc_refs&from_uid=28761828)

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Research Article

Effects of Pomegranate (*Punica Granatum L.*) Seed and Peel Methanolic Extracts on Oxidative Stress and Lipid Profile Changes Induced by Methotrexate in Rats

(/Article/APB_3809_20161002125700)

Farideh Doostan, Roxana Vafafar, Parvin Zakeri-Milani, Aliasghar Pouri, Rogayeh Amini Afshar, Mehran Mesgari Abbasi*

Adv Pharm Bull. 2017;7(2): 269-274. doi: 10.15171/apb.2017.032 (<https://doi.org/10.15171/apb.2017.032>)

PMCID: PMC5527241 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC5527241>) PMID: 28761829

(<http://www.ncbi.nlm.nih.gov/pubmed/28761829>) Scopus ID: 85021420852

(<https://www.scopus.com/inward/record.url?partnerID=HzOxMe3b&scp=85021420852&origin=inward>)

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Research Article

Studying the Effect of Sertraline in Reducing Aggressive Behavior in Patients with Major Depression

(/Article/APB_14580_20160825101452)

Alireza Farnam, Arezoo MehrAra, Hossein Dadashzadeh, Golamreza Chalabianlou, Salman Safikhanlou*

Adv Pharm Bull. 2017;7(2): 275-279. doi: 10.15171/apb.2017.033 (<https://doi.org/10.15171/apb.2017.033>)

PMCID: PMC5527242 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC5527242>) **PMID:** 28761830

(<http://www.ncbi.nlm.nih.gov/pubmed/28761830>) **Scopus ID:** 85021442544

(<https://www.scopus.com/inward/record.url?partnerID=HzOxMe3b&scp=85021442544&origin=inward>)

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Research Article

Preparation and Microstructural Characterization of Griseofulvin Microemulsions Using Different Experimental Methods: SAXS and DSC

(/Article/APB_293_20160829142248)

Eskandar Moghimipour*, Anayatollah Salimi*, Sahar Changizi

Adv Pharm Bull. 2017;7(2): 281-289. doi: 10.15171/apb.2017.034 (<https://doi.org/10.15171/apb.2017.034>)

PMCID: PMC5527243 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC5527243>) **PMID:** 28761831

(<http://www.ncbi.nlm.nih.gov/pubmed/28761831>) **Scopus ID:** 85021397319

(<https://www.scopus.com/inward/record.url?partnerID=HzOxMe3b&scp=85021397319&origin=inward>)

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Scopus* 5 (<https://www.scopus.com/inward/citedby.url?partnerID=HzOxMe3b&scp=85021397319&origin=inward>)

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 Crossref 3 (/citation_report/APB_293_20160829142248/crossref)

Research Article

Anticonvulsant and Antioxidant Effects of Pitavastatin Against Pentylenetetrazol-Induced Kindling in Mice

(/Article/APB_4168_20170124182937)

Nastaran Faghihi, Mohammad Taghi Mohammadi*

Adv Pharm Bull. 2017;7(2): 291-298. doi: 10.15171/apb.2017.035 (<https://doi.org/10.15171/apb.2017.035>)

PMCID: PMC5527244 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC5527244>) **PMID:** 28761832

(<http://www.ncbi.nlm.nih.gov/pubmed/28761832>) **Scopus ID:** 85021429734

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Research Article

Expression of Functional Anti-p24 scFv 183-H12-5C in HEK293T and Jurkat T Cells

(/Article/APB_19315_20170420112001)

Mohammad Tasyriq Che Omar*

Adv Pharm Bull. 2017;7(2): 299-312. doi: 10.15171/apb.2017.036 (<https://doi.org/10.15171/apb.2017.036>)

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Research Article

Development and Optimization of a New Chemoenzymatic Approach for the Synthesis of Peracetylated Lactosamine (Intermediate for the Synthesis of Pharmacologically Active Compounds) Monitored by RP-HPLC Method

(/Article/APB_1387_20161231183306)

Qais Ibrahim Abualassal*, Khaldun Mohammad Al Azzam*, Zead Helmi Abudayeh, Loay Khaled Hassounah

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Research Article

Evaluation of Vitamin D₃ and D₂ Stability in Fortified Flat Bread Samples During Dough Fermentation, Baking and Storage

(/Article/APB_19381_20170514002509)

Mehrnaz Tabibian, Mohammadali Torbati, Mohammad Reza Afshar Mogaddam, Maryam Mirlohi, Malihe Sadeghi,

Javad Mohtadinia*

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Short Communication

Study of Thermal Analysis Behavior of Fenbendazole and Rafoxanide

(/Article/APB_450_20170101191511)

Ali Kamal Attia*, Ahmed Sayed Saad, Manal Sami Alaraki, Eman Saad Elzanfaly

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Short Communication

Human Bone Marrow-Derived Mesenchymal Cell Reactions to 316L Stainless Steel: An in Vitro Study on Cell Viability and Interleukin-6 Expression

(/Article/APB_19332_20170425113627)

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Short Communication



Human Bone Marrow-Derived Mesenchymal Cell Reactions to 316L Stainless Steel: An in Vitro Study on Cell Viability and Interleukin-6 Expression

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Abstract

Purpose: Human bone marrow-derived mesenchymal cell (hBMC) reactions to 316L stainless steel (316L-SS) have never been evaluated. The objective of this study was to assess cell viability and interleukin-6 expression of hBMC cultures upon treatment with a 316L-SS implant.

Methods: A cytotoxicity analysis was conducted with a 3-(4,5-dimethylthiazol 2-yl)-2,5-diphenyltetrazolium (MTT) assay after a period of 24, 48 and 72 hours of incubation. Expression of interleukin-6 was measured using enzyme-linked immunosorbent assay (ELISA).

Results: Cell viability measurement was performed via IC₅₀ formula. All treatment group showed a > 50 % cell viability with a range of 56,5 - 96,9 % at 24 hours, 51,8-77,3% at 48 hours and 70,1- 120 % at 72 hours. Interleukin-6 expression was downregulated subsequent to treatment with 316L-SS compared to the control group.

Conclusion: We found that 316L-SS did not exhibit toxicity towards hBMC culture.

Introduction

Presently, 316L stainless steel (316L-SS) implants are still widely utilized in the field of medical practice, especially orthopedic surgery. It has excellent mechanical properties, corrosion resistance and is cost effective.¹ It is mandatory to evaluate biomedical safety before using such implants in humans.² As one of the dominant cellular components of medullary bone marrow, mesenchymal cells have important roles in several human tissue regeneration processes. They also possess the capability of producing several cytokines (include interleukin-6/IL-6) after contact with foreign material.^{3,4} Human bone marrow mesenchymal cells (hBMC) reactions to 316L-SS have never been evaluated. Therefore, herein, we conducted an *in vitro* cytotoxicity study of 316L-SS on hBMC along with evaluating the expression of IL-6.

Materials and Methods

Human bone marrow-derived mesenchymal cell isolation and culture

Bone marrow samples were obtained from a patient experiencing total hip arthroplasty surgery with written informed consent and ethical approval from the Ethics

Commission of Prof. Dr. R. Soeharso Orthopedic Hospital, Solo, Indonesia. During surgery, an approximately 10 ml of bone marrow suspension was harvested from the intramedullary canal of the femur. It was captured in a 20 ml tube (Falcon, BD Bioscience) containing the same volume of heparinized (10 U/mL) phosphate – buffered saline (PBS) to prevent clotting. The mixture of bone marrow and heparinized PBS was kept at 4°C prior to further processing in the laboratory. hBMC culturing was conducted in the Laboratory of Cell Culture at the Department of Physiology, Gadjah Mada University School of Medicine, Yogyakarta, Indonesia. Bone marrow was loaded into a centrifuge tube and centrifuged at 2500 rpm for 10 minutes at room temperature. The top layer of mononuclear cells was collected and washed with PBS twice and Dulbecco's Modified Eagle Media (DMEM) once. The isolated cells were suspended in DMEM, supplemented with 15% PBS, 1% antibiotic – mycotic (Sigma – Aldrich, Co, USA) and seeded into 25 cm² flasks. The cells were incubated at 37°C, 5% CO₂. After three days, non-adherent cells were removed by washing twice with PBS and the new medium was applied. The cell density and

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Research Article



Development and Optimization of a New Chemoenzymatic Approach for the Synthesis of Peracetylated Lactosamine (Intermediate for the Synthesis of Pharmacologically Active Compounds) Monitored by RP- HPLC Method

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- *Candida rugosa*

Abstract

Purpose: To describe a chemoenzymatic approach joining an enzymatic regioselective hydrolysis of peracetylated *N*-acetyl- α -D-glucosamine (**A**) with a mild controlled acyl relocation which resulted 2-acetamido-2 deoxy-1,3,6-tri-O-acetyl- α -D-glucopyranose (**1B**).

Methods: Immobilization of lipase on decaoctyl (DSEOD) and octyl agarose (OSCL) was carried out as reported by the work of Bastida *et al.* The newly developed RP-HPLC method for examining the enzymatic hydrolysis was carried out isocratically utilizing a HPLC system.

Results: The new approach resulted the target compound (**B**) in 95% yield after purification utilizing flash column chromatography. *Candida rugosa*-lipase immobilized ondecaoctyl-sepabeads was the best catalyst in terms of activity and region-selectivity in the hydrolysis of substrate (**A**), delivering the deacetylation at C6 position (98% general yield).

Also, a reversed-phase high-performance liquid-chromatographic (RP-HPLC) method for controlling the region-selective hydrolysis of peracetylated *N*-acetyl- α -D-glucosamine (**A**) with a mild monitored acyl movement which led to 2-acetamido-2-deoxy-1,3,6-tri-O-acetyl- α -D-glucopyranose (**1B**) has additionally been developed. The developed RP-HPLC method was utilized as fingerprints to follow the hydrolysis of substrate (**A**) and to determine its purity and additionally yield. Furthermore, the acquired compound (**B**) was further purified by flash chromatography. Compound (**B**) was further characterized utilizing ¹H NMR and mass spectrometry.

Conclusion: An efficient chemoenzymatic procedure to optimize the preparation of peracetylated lactosamine **B** containing acetyl ester as extraordinary protecting group is presented. Compound **B** is a significant intermediate for the synthesis of pharmacologically active compound (e.g. complex oligosaccharides for biochemical, biophysical, or biological examinations). Besides, reaction monitoring utilizing HPLC proposes more exact information than spectroscopic methods.

Introduction

Carbohydrates are viewed as a key part in various biological processes. Given their differing difficulty in all cells, it is not surprising that glycans have various assorted part in different physiological processes. The physiological processes are made out of sophisticated multi-cell living organism form. Honestly, a generous number of bioactive compounds are glycosylated and the sugar moiety is considered as a key for their bioactivity.¹ Of them, glycoproteins found in cell-cell recognition of various pathologies are of remarkable interest.² Actually, oligosaccharides found in glycoproteins are recognized by lectin receptors, the key part for carbohydrate-mediated recognition actions.³ For example, lacto oligosaccharides series are incorporated into a couple of structures with high biological concern (e.g., glycolipids and glycoproteins) (Figure1).⁴⁻⁶

The destiny of carbohydrate science will be great by the use of its products. The access to oligosaccharides by separation from natural sources is believed tedious and subsequently it gives simply little material that consistently needs the targeted level of purity. The synthesis of high purity oligosaccharides is considered a challenge. Additionally, it is critical for improvement of glycobiology branch.⁷ The absence of fruitful and successful methods for synthesis decreased the usage and the examination of oligosaccharides for therapeutic and diagnostic applications. The preparation of monodeprotected sugar is one of the basic building stones in accomplishing complex oligosaccharides.^{8,9} Due to the complexity of structure as well as variability of oligosaccharides, it has been highlighted more than in case of proteins or DNA. Moreover, they are not

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Review Article



Legionella Pneumophila and Dendrimers-Mediated Antisense Therapy

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- Vesicular trafficking

Abstract

Finding novel and effective antibiotics for treatment of *Legionella* disease is a challenging field. Treatment with antibiotics usually cures *Legionella* infection; however, if the resultant disease is not timely recognized and treated properly, it leads to poor prognosis and high case fatality rate. *Legionella pneumophila* DrrA protein (Defects in Rab1 recruitment protein A)/also known as SidM affects host cell vesicular trafficking through modification of the activity of cellular small guanosine triphosphatase (GTPase) Rab (Ras-related in brain) function which facilitates intracellular bacterial replication within a supporter vacuole. Also, *Legionella pneumophila* LepA and LepB (Legionella effector protein A and B) proteins suppress host-cell Rab1 protein's function resulting in the cell lysis and release of bacteria that subsequently infect neighbour cells. Legionella readily develops resistant to antibiotics and, therefore, new drugs with different modes of action and therapeutic strategic approaches are urgently required among antimicrobial drug therapies; gene therapy is a novel approach for *Legionnaires disease* treatment. On the contrary to the conventional treatment approaches that target bacterial proteins, new treatment interventions target DNA (Deoxyribonucleic acid), RNA (Ribonucleic acid) species, and different protein families or macromolecular complexes of these components. The above approaches can overcome the problems in therapy of *Legionella* infections caused by antibiotics resistance pathogens. Targeting *Legionella* genes involved in manipulating cellular vesicular trafficking using a dendrimer-mediated antisense therapy is a promising approach to inhibit bacterial replication within the target cells.

Introduction

Legionnaires' disease is a serious form of pneumonia and lung inflammation, which is caused by intracellular bacterium Legionella. Although early therapeutic intervention using antibiotics usually cures Legionnaires' disease, some patients experience complications that could lead to death.¹ Legionella rapidly develops resistance to commonly used antibacterial agents.² Therefore, there is an urgent demand for discovery of new antibacterial targets to overcome the resistance problem. Bacterial pathogens deliver effector proteins which interfere with host cell physiological functions and hijack their target cell machinery leading to specific clinical symptoms.^{3,4} To escape degradation by its host cells, a Legionella-containing vacuole (LCV) is formed and protects the bacterium from cell immune defense,

possibly through secretion of bacterial proteins into the host cytosol.⁵ Therefore, specific antibiotics with high levels of permeability are required to pass cell membrane barrier and reach the bacterium within the cells.⁶ This review describes different gene therapy approaches including antisense therapy mediated by dendrimers to target and eliminate or disarm pathogen, novel method for specific targeting of effective types of antibiotics to intracellular *L. pneumophila* (*Legionella pneumophila*). We also describe antisense therapy for *L. pneumophila* treatment targeting bacterial protein synthesis aiming to disturb host trafficking pathway through interference with phagosome and lysosome fusion in macrophages, therefore targeting bacteria in the cytoplasm by different methods such as RNA interference type would be an

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