Periode	: Januari – Maret
Tahun	: 2024
Skema Penelitian	: Penelitian Dasar
Tema RIP Penelitian	: Kualitas Kesehatan, Penyakit Tropis, Gizi dan Kesehatan

LAPORAN AKHIR
PENELITIAN DASAR

Increasing angiogenesis factors in hypoxic diabetic wounds using siRNA nanotherapeutics

TIM PENGUSUL:

Ketua Tim : Adam Astrada 1101059201

Fakultas Ilmu-Ilmu Kesehatan Program Studi Ilmu Keperawatan
Universitas Esa Unggul
Tahun 2024

Lembar Pengesahan Laporan Akhir
Program Penelitian
Universitas Esa Unggul

1. Judul Kegiatan Penelitian : INCREASING ANGIOGENESIS FACTORS IN HYPOXIC DIABETIC WOUNDS USING SIRNA NANOTHERAPEUTICS
2. Nama Mitra Sasaran :
3. Ketua Tim
   a. Nama Lengkap : ADAM ASTRADA, S.Kep., Ns. MHS,CNS,DHSc,FACCWS
   b. NIDN : 1101059201
   c. Jabatan Fungsional : Lektor (200)
   d. Fakultas/ Program Studi : Fakultas Ilmu-Ilmu Kesehatan/ Fikes/Program Studi Ilmu Keperawatan
   e. Bidang Keahlian :
   f. Nomor Telepon/ HP : 08975235030
   g. Email : adam.astrada@esaunggul.ac.id
4. Jumlah Anggota Dosen : -
5. Jumlah Anggota Mahasiswa : -
6. Lokasi Kegiatan Mitra
   Alamat
   Kabupaten/ Kota
   Provinsi
7. Periode/ Waktu Kegiatan : 22 Januari 2024 s/d 3 Februari 2024
8. Luaran yang Dihasilkan : Jurnal Internasional Bereputasi dan Berfaktor Dampak (Q1)
9. Usulan/ Realisasi Anggaran
   a. Dana Mandiri :
   b. Sumber Dana Lain (1) :

Jakarta, 25 Maret 2024
Ketua Peneliti,

(ADAM ASTRADA, S.Kep., Ns. MHS,CNS,DHSc,FACCWS)
NIDN/K. 1101059201

Menyetujui,
Dekan Fakultas Ilmu-Ilmu Kesehatan

(Prof. Dr. APRILITA RINA YANTI EFF, M.Biomed, Apt)
NIP/NIK. 215020572

Mengetahui,
Ketua Lembaga Penelitian dan Pengabdian Masyarakat Universitas Esa Unggul

(LARAS SITOAYU, S.Gz, M.K.M)
NIK. 215080596

IDENTITAS DAN URAIAN UMUM

1. Judul Penelitian : Increasing angiogenesis factors in hypoxic diabetic wounds using siRNA nanotherapeutics

2. Tim Peneliti

No	Nama	Jabatan	Bidang Keahlian	Instansi Asal	Alokasi Waktu (jam/minggu)
1	Adam Astrada	Ketua	Keperawatan Medikal Bedah	Universitas Esa Unggul	20 jam/ minggu

3. Objek Penelitian (jenis material yang akan diteliti dan segi penelitian):

   Objek pada penelitian ini adalah penelaahan sistematis disertai meta-analysis untuk mencari bukti terkait penggunaan terapi topikal dalam menangani masalah biofilm pada luka.

4. Masa Pelaksanaan

   Mulai : Januari tahun: 2024

   Berakhir : Januari tahun: 2024

5. Usulan Biaya: Tahun ke-1 : Rp -

6. Lokasi Penelitian (lab/studio/lapangan): Kantor.

7. Instansi lain yang terlibat (jika ada, dan uraikan apa kontribusinya)

8. Temuan yang ditargetkan (produk atau masukan untuk kebijakan)

   Hasil penelitian ini diharapkan dapat menemukan suatu bukti untuk penerapan *evidence-based practice* dalam penanganan masalah pada luka.

9. Kontribusi mendasar pada suatu bidang ilmu

   Penelitian ini didasari atas meningkatnya masalah sirkulasi pada luka kaki diabetic.

10. Jurnal ilmiah yang menjadi sasaran: Molecular Therapy Nucleic Acids

    Tahun : 2024

    Luaran HKI : -

DAFTAR ISI

DAFTAR TIM PELAKSANA DAN TUGAS

1. Ketua Pelaksana

Nama	: Adam Astrada, Ns., MHS, CNS, DHSc., FACCWS
NIDN	: 1101059201
Jabatan Fungsional	: Lektor (200)
Fakultas/ Prodi	: Ilmu-Ilmu Kesehatan/Ilmu Keperawatan
Tugas	: Bertanggung jawab terhadap pembuatan proposal, pelaksanaan riset, mengolah data dan menyusun laporan akhir, membuat pertanggung-jawaban keuangan, menyusun publikasi

BAB I
PENDAHULUAN

1. Latar Belakang

I read with interest the article by Shaabani et al. on siRNA-mediated prolyl hydroxylase domain protein 2 (PHD2) silencing to promote angiogenesis in diabetic wounds.(Shaabani et al., 2021) The authors demonstrate a layered gold nanoparticle delivery system for enhanced cytosolic release of PHD2 siRNA. Co-treatment with the endosomolytic agent desloratadine led to amplified gene knockdown in vitro. This is a significant advance given inefficient intracellular delivery remains a fundamental limitation of siRNA therapeutics.(Wang et al., 2010)

.

2. Permasalahan

However, some critical knowledge gaps remain. Firstly, it is unclear whether PHD2 silencing can induce sustained pro-angiogenic effects specifically in the chronically inflamed wound microenvironment, or whether compensatory negative feedback mechanisms may limit efficacy over time. Dynamic tracking of VEGF/FGF expression in cytokine-stimulated fibroblasts could provide insight.

3. Tujuan Penelitian

Shaabani et al. have developed an interesting PHD2 siRNA gold nanosystem as a pro-angiogenic therapeutic. Evaluation of sustained activity in inflammatory conditions, comparisons to miRNA-based approaches, benchmarking to other nanocarriers, and in-depth in vivo biocompatibility studies would strengthen conclusions regarding real-world efficacy and safety.

4. Manfaat Penelitian

5. Hasil yang Diharapkan

No	Jenis Luaran				Indikator Capaian		
	Kategori	**Sub Kategori**	**Wajib**	**Tambahan**	**TS[1)]**	**TS+1**	**TS+2**
1	Artikel ilmiah dimuat di jurnal[2)]	Internasional		√			
		Nasional terakreditasi			√		
		Nasional tidak terakreditasi					
2	Artikel ilmiah dimuat di prosiding[3)]	Internasional					
		Nasional					
3	*Invited speaker* dalam temu ilmiah[4)]	Internasional					
		Nasional					
4	Hak Kekayaan Intelektual (HKI)[6)]	Paten					
		Paten sederhana					
		Hak cipta					
		Merek dagang					
		Rahasia dagang					
		Desain produk industry					
		Indikasi geografis					
		Perlindungan varietas tanaman					
		Perlindungan topogarfi sirkuit terpadu					
5	Tehnologi tepat guna[7)]						
6	Model/Purwarupa/Desain/ Karya						

	seni/ Rekayasa sosial[8)]					
7	Buku ajar (ISBN)					
8	Tingkat kesiapan teknologi (TKT)[10)]			1-2	3	

BAB II

RENSTRA DAN PETA JALAN PENELITIAN PERGURUAN TINGGI

Dalam penelitian ini, mengacu kepada RIP Universitas Esa Unggul yaitu Kualitas Kesehatan, Penyakit Tropis, Gizi & Obat-Obatan (*Health, Disease, Nutrition & Medicine).*

BAB III
TINJAUAN PUSTAKA DAN LANDASAN TEORI

1. Tinjauan Pustaka

Dear Editor,

I read with interest the article by Shaabani et al. on siRNA-mediated prolyl hydroxylase domain protein 2 (PHD2) silencing to promote angiogenesis in diabetic wounds.(Shaabani et al., 2021) The authors demonstrate a layered gold nanoparticle delivery system for enhanced cytosolic release of PHD2 siRNA. Co-treatment with the endosomolytic agent desloratadine led to amplified gene knockdown in vitro. This is a significant advance given inefficient intracellular delivery remains a fundamental limitation of siRNA therapeutics.(Wang et al., 2010)

2. Tinjauan Teori

Compared to previous works on pro-angiogenic siRNA therapies for diabetic wounds, this study demonstrates several strengths:

- Selection of the PHD-2 target: Many other studies have similarly focused on PHD-2 silencing to stabilize HIF-1α and induce VEGF/FGF expression.(Wang et al., 2010; Wetterau et al., 2011) However, Shaabani et al. provide a more extensive mechanistic rationale for why PHD-2 inhibition can enhance angiogenesis in the ischemic and inflammatory context of diabetic wounds.
- Tunable LbL nanocarrier design: The assembly of siRNA polyplexes with gold nanoparticles and subsequent outer layer coating provides fine control over surface charge, stability, and endosomal escape properties. This represents an advance over simpler nanoparticle formulations used previously.
- Combination with desloratadine: Co-administration with an endosomolytic agent like desloratadine to amplify siRNA delivery is a novel approach not explored in other wound angiogenesis studies. This demonstrates the synergistic effects possible by integrating drug and gene delivery.

- Assessment in vitro: Many prior works examine siRNA or shRNA against PHD-2 was in vivo.(Paik et al., 2016) By first optimizing silencing in NIH-3T3 cells, Shaabani et al. provide better insights into the intracellular trafficking and gene knockdown efficiencies of their nanosystem.

BAB IV
METODE PENELITIAN

1. Metode Penelitian

However, some critical knowledge gaps remain. Firstly, it is unclear whether PHD2 silencing can induce sustained pro-angiogenic effects specifically in the chronically inflamed wound microenvironment, or whether compensatory negative feedback mechanisms may limit efficacy over time. Dynamic tracking of VEGF/FGF expression in cytokine-stimulated fibroblasts could provide insight.

2. Teknik Analisis Data

Secondly, the specificity of the gene knockdown approach could be improved. Silencing individual anti-angiogenic microRNAs like miR-26a or miR-29a allows finer tuning of the neovascularization response compared to broad PHD2 inhibition (Wu et al., 2019; Yang et al., 2021).(Wu et al., 2019; Yang et al., 2021) However, delivery of miRNA mimics or inhibitors poses additional pharmaceutical challenges. Direct comparative assessment between miRNA- and siRNA-based pro-angiogenic strategies is limited.

Furthermore, the study lacked comparisons to other leading siRNA nanocarriers. Lipid, polymer, and peptide-based systems have shown promise for wound applications, yet were not benchmarked.(Beiki et al., 2017) The additive effects of desloratadine-mediated endosomal escape were also rather modest. Testing in other cell types would better elucidate on-target effects and advantages over existing delivery platforms.

Finally, combination delivery with chemical enhancers like desloratadine requires an in-depth investigation of off-target toxicities and wound healing outcomes. Desloratadine may influence histamine receptor signaling on resident immune cells, possibly exacerbating inflammation. And while in vitro assays offer preliminary screening, ultimately prolonged in vivo biocompatibility studies in diabetic wound models are essential to validate clinical utility.

In summary, Shaabani et al. have developed an interesting PHD2 siRNA gold nanosystem as a pro-angiogenic therapeutic. Evaluation of sustained activity in inflammatory conditions, comparisons to miRNA-based approaches, benchmarking to other nanocarriers, and in-depth in vivo biocompatibility studies would strengthen conclusions regarding real-world efficacy and safety. I look forward to seeing this work address these gaps while evolving into a combination treatment approach optimized for the complex diabetic wound microenvironment.

BAB V

HASIL PENELITIAN

However, some critical knowledge gaps remain. Firstly, it is unclear whether PHD2 silencing can induce sustained pro-angiogenic effects specifically in the chronically inflamed wound microenvironment, or whether compensatory negative feedback mechanisms may limit efficacy over time. Dynamic tracking of VEGF/FGF expression in cytokine-stimulated fibroblasts could provide insight.

Secondly, the specificity of the gene knockdown approach could be improved. Silencing individual anti-angiogenic microRNAs like miR-26a or miR-29a allows finer tuning of the neovascularization response compared to broad PHD2 inhibition (Wu et al., 2019; Yang et al., 2021).(Wu et al., 2019; Yang et al., 2021) However, delivery of miRNA mimics or inhibitors poses additional pharmaceutical challenges. Direct comparative assessment between miRNA- and siRNA-based pro-angiogenic strategies is limited.

Furthermore, the study lacked comparisons to other leading siRNA nanocarriers. Lipid, polymer, and peptide-based systems have shown promise for wound applications, yet were not benchmarked.(Beiki et al., 2017) The additive effects of desloratadine-mediated endosomal escape were also rather modest. Testing in other cell types would better elucidate on-target effects and advantages over existing delivery platforms.

Finally, combination delivery with chemical enhancers like desloratadine requires an in-depth investigation of off-target toxicities and wound healing outcomes. Desloratadine may influence histamine receptor signaling on resident immune cells, possibly exacerbating inflammation. And while in vitro assays offer preliminary screening, ultimately prolonged in vivo biocompatibility studies in diabetic wound models are essential to validate clinical utility.

In summary, Shaabani et al. have developed an interesting PHD2 siRNA gold nanosystem as a pro-angiogenic therapeutic. Evaluation of sustained activity in inflammatory conditions, comparisons to miRNA-based approaches, benchmarking to other nanocarriers, and in-depth in vivo biocompatibility studies would strengthen

conclusions regarding real-world efficacy and safety. I look forward to seeing this work address these gaps while evolving into a combination treatment approach optimized for the complex diabetic wound microenvironment.

BAB VI

PEMBAHASAN

However, some critical knowledge gaps remain. Firstly, it is unclear whether PHD2 silencing can induce sustained pro-angiogenic effects specifically in the chronically inflamed wound microenvironment, or whether compensatory negative feedback mechanisms may limit efficacy over time. Dynamic tracking of VEGF/FGF expression in cytokine-stimulated fibroblasts could provide insight.

Secondly, the specificity of the gene knockdown approach could be improved. Silencing individual anti-angiogenic microRNAs like miR-26a or miR-29a allows finer tuning of the neovascularization response compared to broad PHD2 inhibition (Wu et al., 2019; Yang et al., 2021).(Wu et al., 2019; Yang et al., 2021) However, delivery of miRNA mimics or inhibitors poses additional pharmaceutical challenges. Direct comparative assessment between miRNA- and siRNA-based pro-angiogenic strategies is limited.

Furthermore, the study lacked comparisons to other leading siRNA nanocarriers. Lipid, polymer, and peptide-based systems have shown promise for wound applications, yet were not benchmarked.(Beiki et al., 2017) The additive effects of desloratadine-mediated endosomal escape were also rather modest. Testing in other cell types would better elucidate on-target effects and advantages over existing delivery platforms.

Finally, combination delivery with chemical enhancers like desloratadine requires an in-depth investigation of off-target toxicities and wound healing outcomes. Desloratadine may influence histamine receptor signaling on resident immune cells, possibly exacerbating inflammation. And while in vitro assays offer preliminary screening, ultimately prolonged in vivo biocompatibility studies in diabetic wound models are essential to validate clinical utility.

In summary, Shaabani et al. have developed an interesting PHD2 siRNA gold nanosystem as a pro-angiogenic therapeutic. Evaluation of sustained activity in inflammatory conditions, comparisons to miRNA-based approaches, benchmarking to other nanocarriers, and in-depth in vivo biocompatibility studies would strengthen

conclusions regarding real-world efficacy and safety. I look forward to seeing this work address these gaps while evolving into a combination treatment approach optimized for the complex diabetic wound microenvironment.

BAB VII
KESIMPULAN DAN SARAN

A. KESIMPULAN

In summary, Shaabani et al. have developed an interesting PHD2 siRNA gold nanosystem as a pro-angiogenic therapeutic. Evaluation of sustained activity in inflammatory conditions, comparisons to miRNA-based approaches, benchmarking to other nanocarriers, and in-depth in vivo biocompatibility studies would strengthen conclusions regarding real-world efficacy and safety. I look forward to seeing this work address these gaps while evolving into a combination treatment approach optimized for the complex diabetic wound microenvironment.

BAB VIII

BIAYA DAN JADWAL PENELITIAN

A. Anggaran Biaya

A. Jadwal Penelitian

No	Jenis Kegiatan	Bulan ke-											
		1	2	3	4	5	6	7	8	9	10	11	12
1													
2													
3													
4													
6													
7													
8	Pembuatan laporan												
9	Seminar												
10	Publikasi												

DAFTAR PUSTAKA

Beiki, B., Zeynali, B., & Seyedjafari, E. (2017). Fabrication of a three dimensional spongy scaffold using human Wharton's jelly derived extra cellular matrix for wound healing. *Materials Science and Engineering: C*, *78*, 627–638. https://doi.org/10.1016/j.msec.2017.04.074

Paik, K. J., Maan, Z. N., Zielins, E. R., Duscher, D., Whittam, A. J., Morrison, S. D., Brett, E. A., Ransom, R. C., Hu, M. S., Wu, J. C., Gurtner, G. C., Longaker, M. T., & Wan, D. C. (2016). Short Hairpin RNA Silencing of PHD-2 Improves Neovascularization and Functional Outcomes in Diabetic Wounds and Ischemic Limbs. *PLOS ONE*, *11*(3), e0150927. https://doi.org/10.1371/journal.pone.0150927

Shaabani, E., Sharifiaghdam, M., Lammens, J., De Keersmaecker, H., Vervaet, C., De Beer, T., Motevaseli, E., Ghahremani, M. H., Mansouri, P., De Smedt, S., Raemdonck, K., Faridi-Majidi, R., Braeckmans, K., & Fraire, J. C. (2021). Increasing Angiogenesis Factors in Hypoxic Diabetic Wound Conditions by siRNA Delivery: Additive Effect of LbL-Gold Nanocarriers and Desloratadine-Induced Lysosomal Escape. *International Journal of Molecular Sciences*, *22*(17), 9216. https://doi.org/10.3390/ijms22179216

Wang, J., Lu, Z., Wientjes, M. G., & Au, J. L. S. (2010). Delivery of siRNA Therapeutics: Barriers and Carriers. *The AAPS Journal*, *12*(4), 492–503. https://doi.org/10.1208/s12248-010-9210-4

Wetterau, M., George, F., Weinstein, A., Nguyen, P. D., Tutela, J. P., Knobel, D., Cohen BA, O., Warren, S. M., & Saadeh, P. B. (2011). Topical prolyl hydroxylase domain-2 silencing improves diabetic murine wound closure. *Wound Repair and Regeneration*, *19*(4), 481–486. https://doi.org/https://doi.org/10.1111/j.1524-475X.2011.00697.x

Wu, H., Li, F., Shao, W., Gao, J., & Ling, D. (2019). Promoting Angiogenesis in Oxidative Diabetic Wound Microenvironment Using a Nanozyme-Reinforced Self-Protecting Hydrogel. *ACS Central Science*, *5*(3), 477–485. https://doi.org/10.1021/acscentsci.8b00850

Yang, Y.-L., Chang, Y.-H., Li, C.-J., Huang, Y.-H., Tsai, M.-C., Chu, P.-Y., & Lin, H.-

Y. (2021). New Insights into the Role of miR-29a in Hepatocellular Carcinoma: Implications in Mechanisms and Theragnostics. *Journal of Personalized Medicine*, *11*(3), 219. https://doi.org/10.3390/jpm11030219

Lampiran 1. Surat Pernyataan Ketua Pelaksana

Surat Pernyataan Ketua Pelaksana
Penelitian

Yang bertadatangan di bawah ini:

Nama	: Adam Astrada, Ns., MHS, CNS, DHSc., FACCWS
NIDN/NIK	: 1101059201
Fakultas/ Prodi	: Ilmu-Ilmu Kesehatan
Jabatan fungsional	: Lektor (200)

Dengan ini saya menyatakan bahwa proposal program penelitian yang diajukan dengan judul:

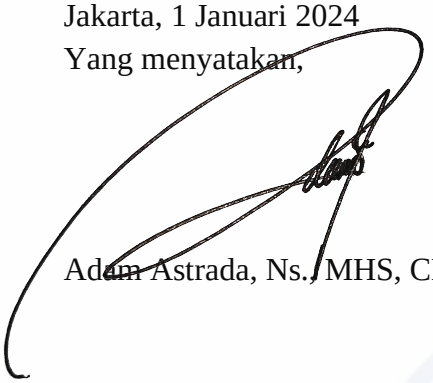
"Increasing angiogenesis factors in hypoxic diabetic wounds using siRNA nanotherapeutics"

Yang saya usulkan dalam skema penelitian mandiri tahun 2024 bersifat original dan belum pernah dibiayai oleh lembaga/ sumber dana lain.

Bilamana diketahui dikemudian hari adanya indikasi ketidakjujuran/ itikad kurang baik sebagaimana dimaksud di atas, maka kegiatan ini dibatalkan dan saya bersedia mengembalikan dana yang telah diterima kepada pihak Universitas Esa Unggul melalui LPPM.

Demikian pernyataan ini dibuat dengan sesumgguhnya dan dengan sebenar-benarnya.

Jakarta, 1 Januari 2024
Yang menyatakan,



Adam Astrada, Ns., MHS, CNS, DHSc., FACCWS

Lampiran 2. Biodata Pengusul dan Anggota

Biodata Pengusul