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LAMPIRAN

Lampiran 1. Isolasi *Actinomycetes*

1. Pengambilan sampel tanah rizosfer tumbuhan *Peperomia pellucida*

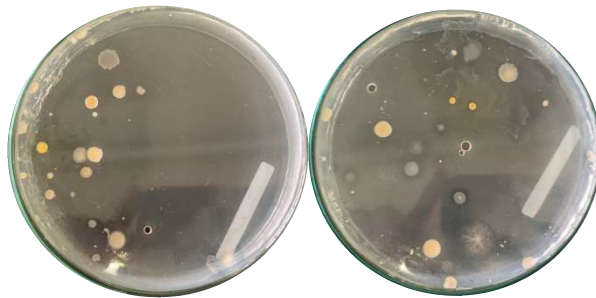


2. Pengenceran sampel tanah rizosfer tumbuhan *Peperomia pellucida* dalam lima seri



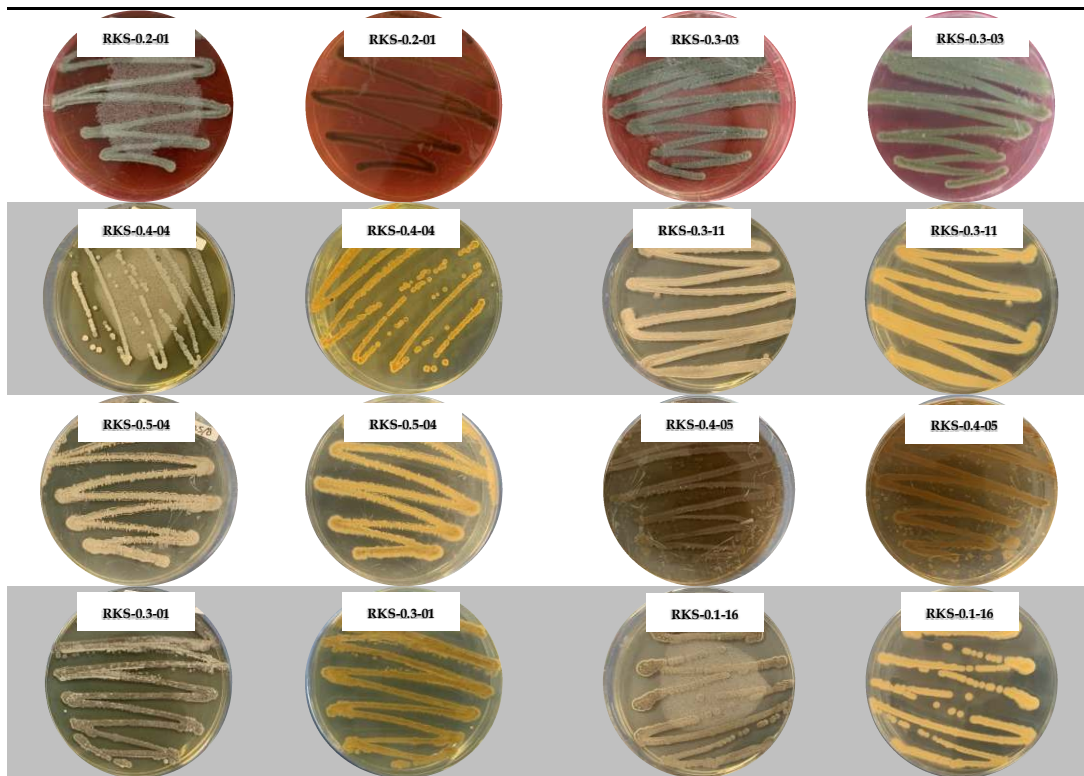
3. Isolasi *Actinomycetes* pada media ISP4 (*Inorganic salt starch agar*) yang berumur 7 hari





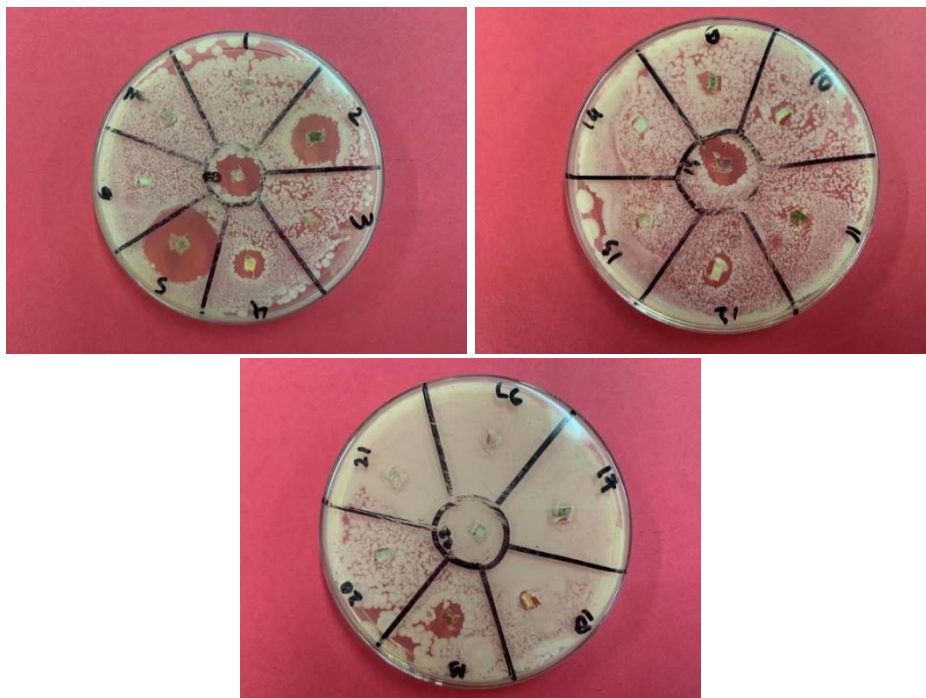
4. Pemurnian *Actinomycetes* pada media ISP2 (*Yeast malt agar*) yang berumur 7 hari.





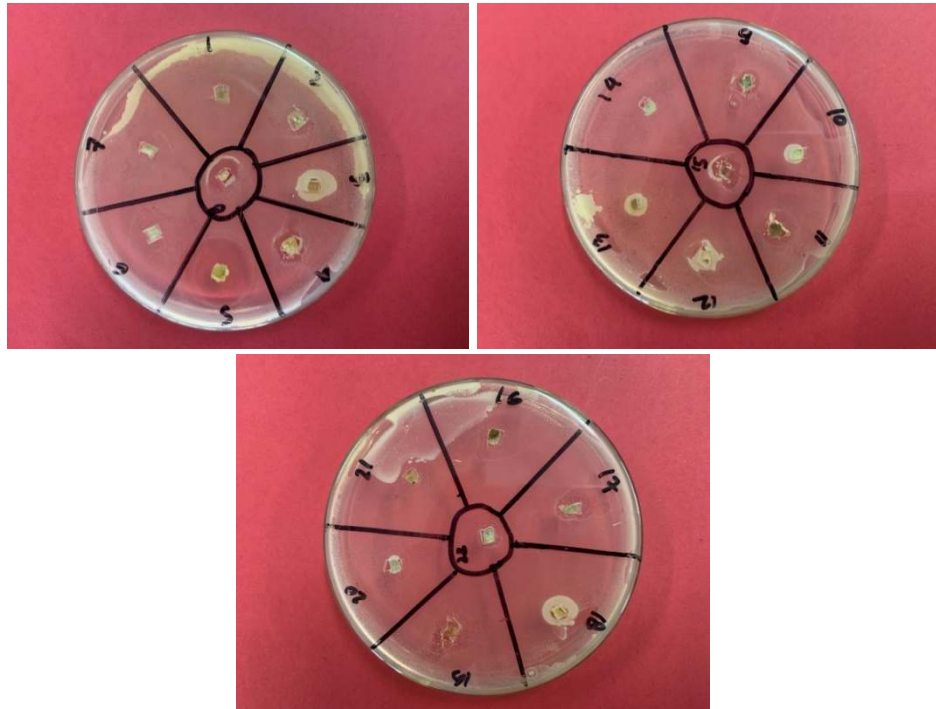
5. Pengujian antagonis isolat *Actinomycetes*

a. Bakteri *E. coli*



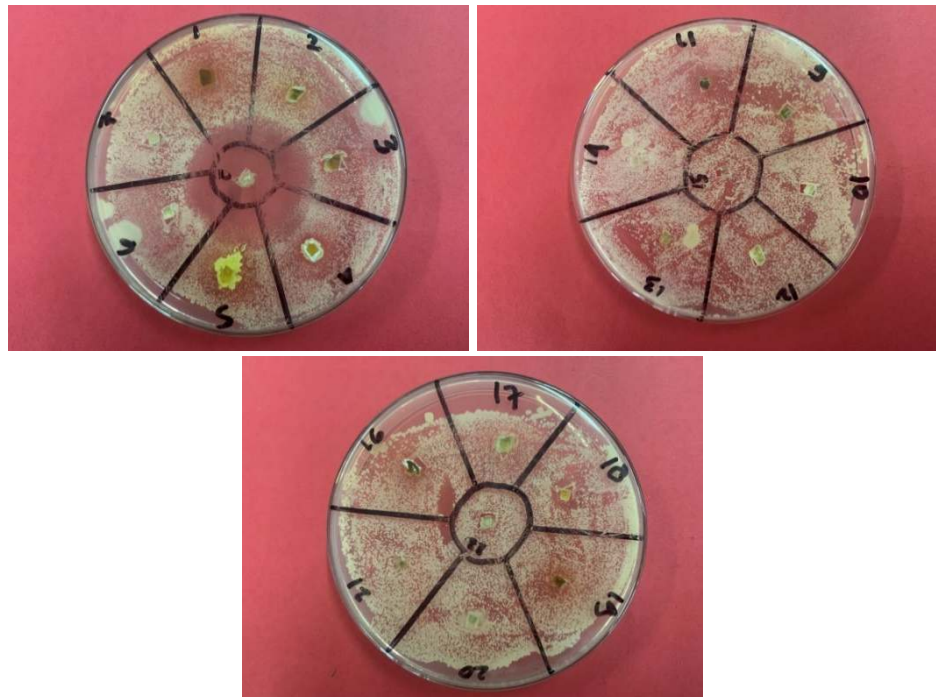
Ket. Kode isolat dapat dilihat pada tabel 2

b. Bakteri *S. aureus*



Ket. Kode isolat dapat dilihat pada tabel 2

c. Fungi *C. albicans*



Ket. Kode isolat dapat dilihat pada tabel 2

6. Fermentasi



a. Isolat RKS-0.3-13

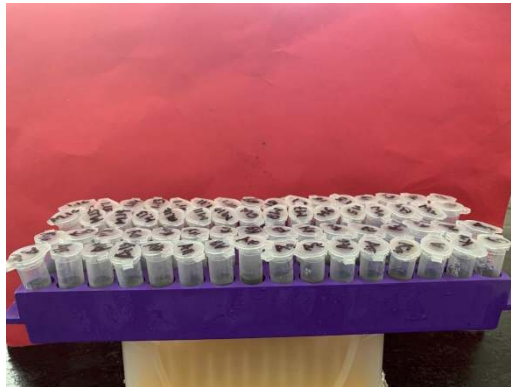


b. Isolat RKS-0.4-04





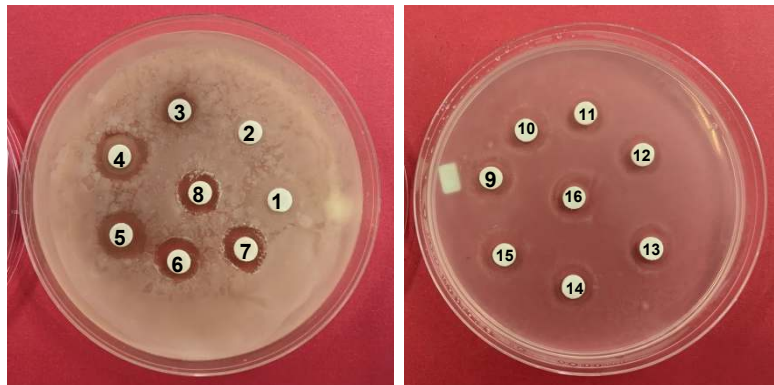
c. Sampling Fermentasi



7. Pengujian kurva hubungan antara zona hambat dengan waktu fermentasi

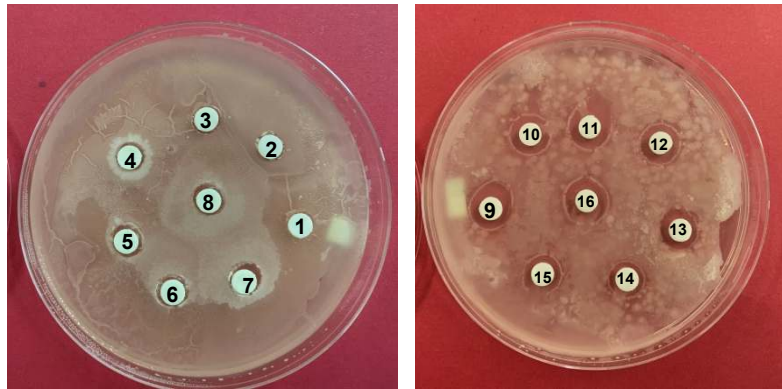
a. Isolat RKS-0.3-13

1). *E. coli*



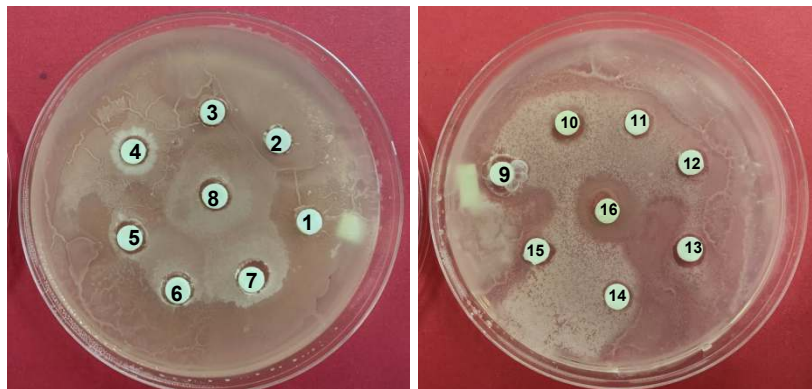
Ket. Nomor yang ada pada gambar adalah waktu/hari fermentasi

2). *S. aureus*



Ket. Nomor yang ada pada gambar adalah waktu/hari fermentasi

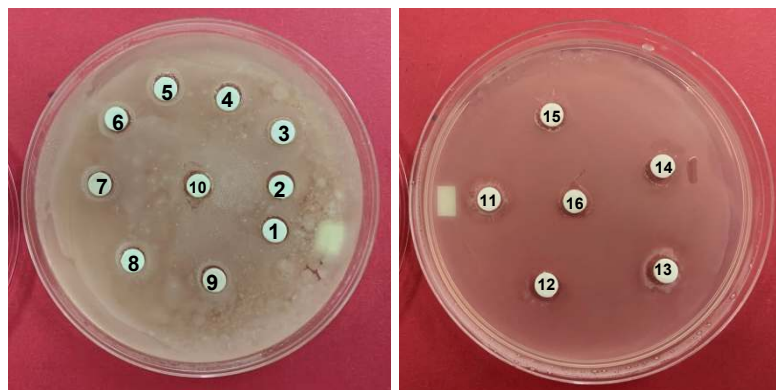
3). *C. albicans*



Ket. Nomor yang ada pada gambar adalah waktu/hari fermentasi

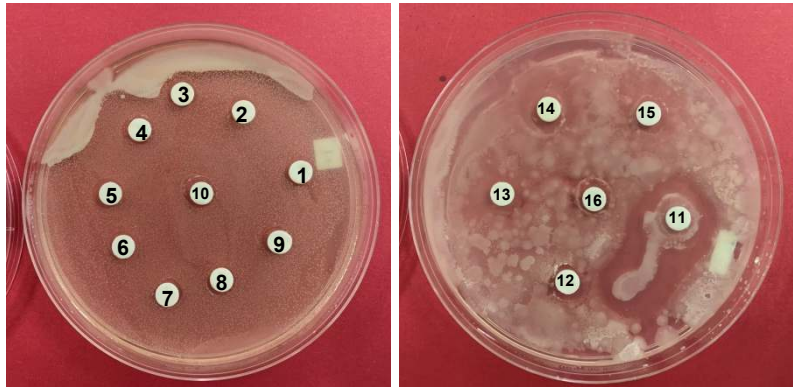
b. Isolat RKS-0.4-04

1). *E. coli*



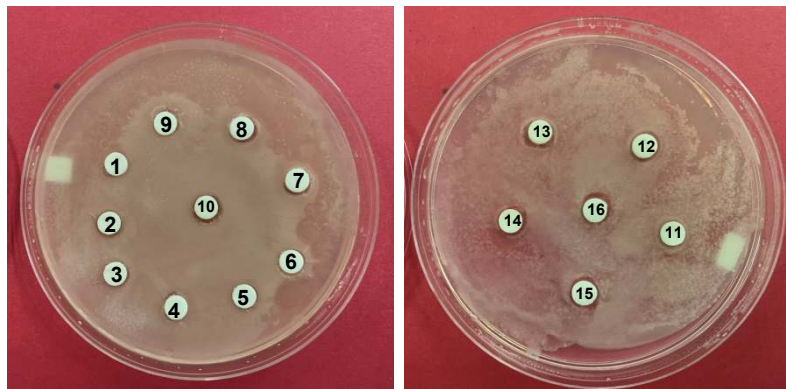
Ket. Nomor yang ada pada gambar adalah waktu/hari fermentasi

2). *S. aureus*



Ket. Nomor yang ada pada gambar adalah waktu/hari fermentasi

3). *C. albicans*



Ket. Nomor yang ada pada gambar adalah waktu/hari fermentasi

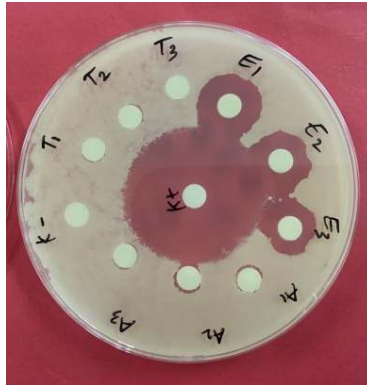
8. Ekstraksi Senyawa Metabolit Sekunder



9. Pengujian Aktivitas Antimikroba

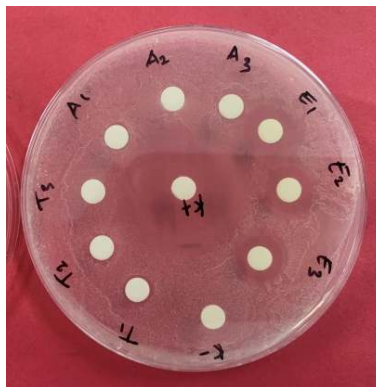
a. Isolat RKS-0.3-13

1). *E. coli*



Ket. A₁, A₂, A₃ (ekstrak air *Actinomyces*), E₁, E₂, E₃ (Ekstrak etil asetat *Actinomyces*) K⁺ (Kontrol positif), K⁻, T₁, T₂, T₃ (Kontrol Negatif)

2). *S. aureus*



Ket. A₁, A₂, A₃ (ekstrak air *Actinomyces*), E₁, E₂, E₃ (Ekstrak etil asetat *Actinomyces*) K⁺ (Kontrol positif), K⁻, T₁, T₂, T₃ (Kontrol Negatif)

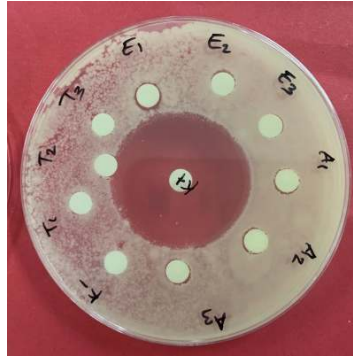
3). *C. albicans*



Ket. A₁, A₂, A₃ (ekstrak air *Actinomyces*), E₁, E₂, E₃ (Ekstrak etil asetat *Actinomyces*) K⁺ (Kontrol positif), K⁻, T₁, T₂, T₃ (Kontrol Negatif)

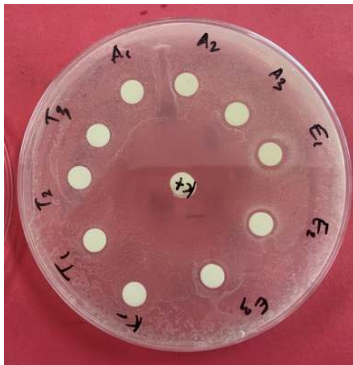
b. Isolat RKS-0.4-04

1). *E. coli*



Ket. A₁, A₂, A₃ (ekstrak air *Actinomyces*), E₁, E₂, E₃ (Ekstrak etil asetat *Actinomyces*) K⁺ (Kontrol positif), K⁻, T₁, T₂, T₃ (Kontrol Negatif)

2). *S. aureus*



Ket. A₁, A₂, A₃ (ekstrak air *Actinomyces*), E₁, E₂, E₃ (Ekstrak etil asetat *Actinomyces*) K⁺ (Kontrol positif), K⁻, T₁, T₂, T₃ (Kontrol Negatif)

3). *C. albicans*



Ket. A₁, A₂, A₃ (ekstrak air *Actinomyces*), E₁, E₂, E₃ (Ekstrak etil asetat *Actinomyces*) K⁺ (Kontrol positif), K⁻, T₁, T₂, T₃ (Kontrol Negatif)

10. Kromatografi Lapis Tipis

a. Maserasi

1). Daun Suruhan dari Ekosistem Karst



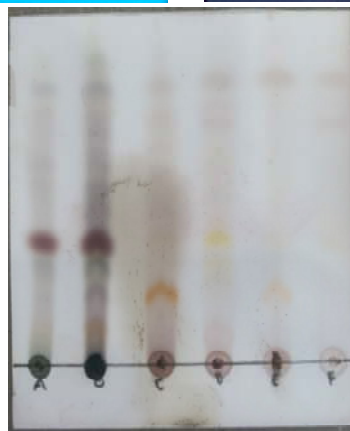
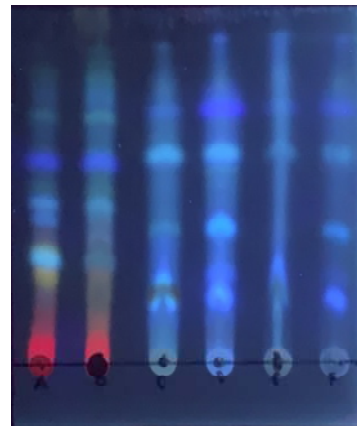
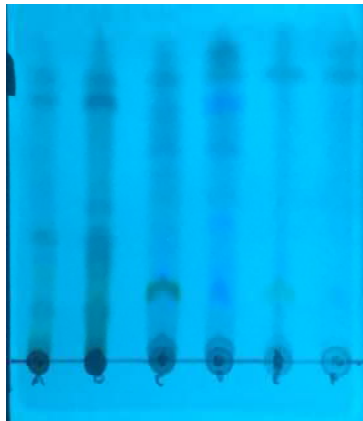
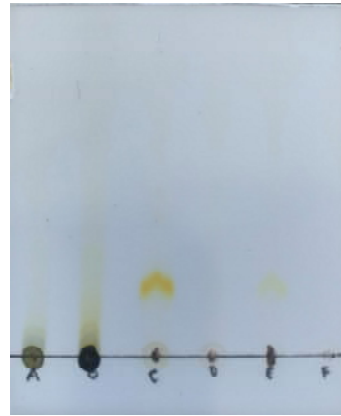
2). Daun Suruhan dari Ekosistem Non Karst



b. Ekstrak



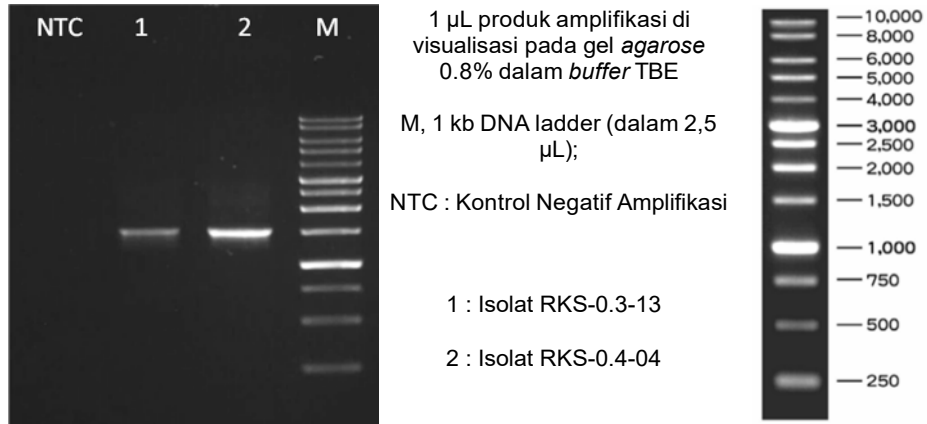
c. Pengujian KLT



Ket. Keterangan gambar dapat dilihat pada gambar 5

11. Identifikasi Molekuler

a. Elektroforesis Produk PCR



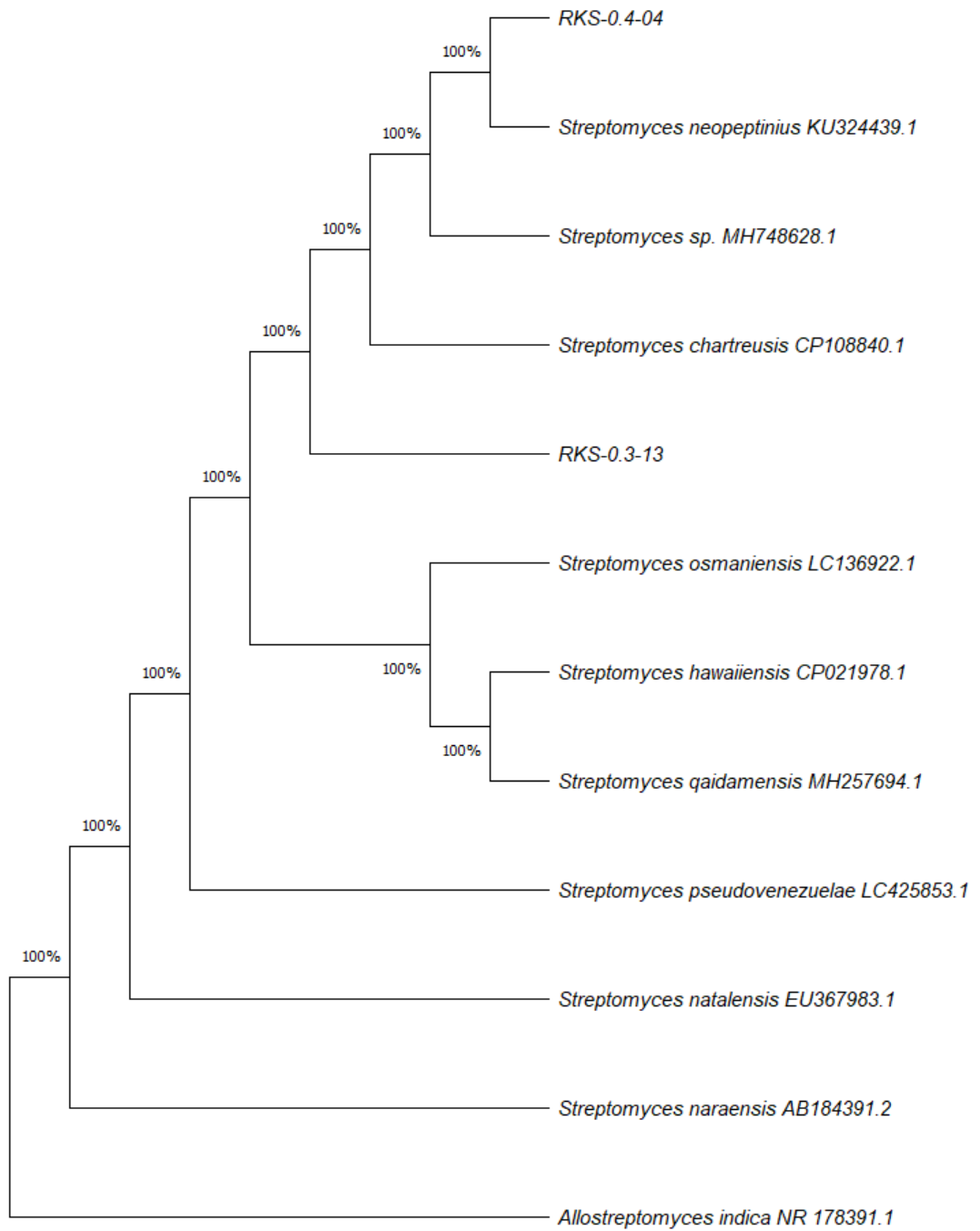
b. Hasil Sequencing – Produk Amplifikasi

NO	Kode Sampel	Sekuens
1	RKS-0.3-13	<i>Sequence Assembly 1390bp</i>
		1 TGCAGTCGAA CGATGAACCA CTTCGGTGGG GATTAGTGGC GAACGGGTGA GTAACACGTG
		61 GGCAATCTGC CCTTCACTCT GGGACAAGCC CTGGAACGGG GGTCTAATAC CGGATAACAC
		121 TTCCACTCTC CTGGGTGGAG GTTAAAAGCT CCGGCGGTGA AGGATGAGCC CGCGCCCTAT
		181 CAGCTTGTGG GTGAGGTAAT GGCTCACCAG GCGACGACG GGTAGCCGGC CTGAGAGGGC
		241 GACCGGCCAC ACTGGGACTG AGACACGGCC CAGACTCCTA CGGGAGGCAG CAGTGGGGAA
		301 TATTGCACAA TGGGCGAAAG CCTGATGCAG CGACGCCGCG TGAGGGATGA CGGCCTTCGG
		361 GTTGTAAACC TCTTTCAGCA GGAAGAAGC GAAAGTGACG GTACCTGCAG AAGAAGCGCC
		421 GGCTAACTAC GTGCCAGCAG CCGCGTAAT ACGTAGGGCG CAAGCGTTGT CCGGAATTAT
		481 TGGGCGTAAA GAGCTCGTAG GCGGCTTGTG ACGTGCGGTG TGAAAGCCCG GGCCTTAACC
		541 CCGGCTCTGC ATTCGATACG GGCTAGCTAG AGTGTGGTAG GGGAGATCGG AATTCTCGGT
		601 GTAGCGGTGA AATGCGCAGA TATCAGGAGG AACACCGGTG CCGAAGGCGG ATCTCTGGGC
		661 CATTACTGAC GCTGAGGAGC GAAAGCGTGG GGAGCGAACA GGATTAGATA CCCTGGTAGT
		721 CCACGCCGTA AACCGTGGGA ACTAGGTGTT GCGCACATTC CAGTCTGTCG GTGCCGCAGC
		781 TAACGCATTA AGTTCGCCGC CTGGGGAGTA CCGCCGCAAG GCTAAAATC AAAGGAATTG
		841 ACGGGGGCCC GCACAAGCAG CCGAGCATGT GGCTTAATTC GACGCAACGC GAAGAACCCT
		901 ACCAAGGCTT GACATACACC GGAACGCTCT GGAGACAGGC CCCCCTTGT GTTCGGTGTG
		961 CAGGTGGTGC ATGGCTGTGC TCAGCTCGTG TCGTGAGATG TTGGGTTAAG TCCCGCCAACG
		1021 AGCGCAACCC TTGTTCTGTG TTGCCAGCAT GCCCTTCGGG GTGATGGGGA CTCACAGGAG
1081 ACCGCCGGGG TCAACTCGGA GGAAGGTGGG GACGACGTCA AGTCATCATG CCCCTTATGT		
1141 CTTGGGCTGC ACACGTGCTA CAATGGCAGG TACAATGAGC TCGGATACCG TGAGTGGGAG		
1201 CGAATCTCAA AAGCCTGTCT CAGTTCGGTA TTGGGCTCTG CACTCGACC CCATGAAGTC		
1261 GGAGTTGCTA GTAATCGCAG ATCAGCATTG CTGCGGTGAA TACGTTCCCG GGCCTTGTAC		
1321 ACACCGCCCG TCACGTCACG AAAGTCGGTA ACACCCGAAG CCGGTGGCCC AACCCCTTGT		
1381 GGGAGGGAGC		
2	RKS-0.4-04	<i>Sequence Assembly 1377bp</i>
		1 GCAGTCGAAC GATGAACCAC TTCGGTGGGG ATTAGTGGCC AACGGGTGAG TAACACGTGG
		61 GCAATCTGCC CTTCACTCTG GGACAAGCCC TGGAACGGG GTCTAATACC GGATAACACT
		121 TTCCACTCTC TGGGTGGAGG TTTAAAAGCTC CCGGCGGTGA GGATGAGCCC GCGCCCTATC
		181 AGCTTGTGGG TGAGGTAATG GCTCACCAGG GCGACGACGG GTAGCCGGCC TGAGAGGGCG
		241 ACCGGCCACA CTGGGACTGA GACACGGCCC AGACTCCTAC GGGAGGCAGC AGTGGGGAAT
		301 ATTGACAAAT GGGCGAAAGC CTGATGCAGC GACGCCGCGT GAGGGATGAC GGCCTTCGGG
		361 TTGTAAACCT CCTTCAGCAG GGAAGAAGCG AAAGTGACGG TACCTGCAGA AGAAGCGCCG
		421 GCTAACTACG TGCCAGCAGC CCGGTAATA CGTAGGGCGC AAGCGTTGTG CCGAATTATT
		481 GGGCGTAAAG AGCTCGTAGG CCGCTTGTCA CGTGGGTGTG GAAAGCCCGG GGCTTAACCC
		541 CCGGTCTGCA TTCGATACGG GCTAGCTAGA GTGTGGTAGG GGAGATCGGA ATTCTGGTG
		601 TAGCGGTGAA ATGCGCAGAT ATCAGGAGGA ACACCGTGG CGAAGCGGGA TCTCTGGGCC
		661 ATTACTGACG CTGAGGAGCG AAAGCGTGGG GAGCGAACAG GATTAGATAC CCTGGTAGTC
		721 CACGCCGTAA ACGGTGGGAA CTAGGTGTTG GCGACATTCC ACGTCTGCGG TGCCGACGCT
		781 AAGCATTAA GTTCCCCGCC TGGGGAGTAC GGCCGCAAGG CTAATACTCA AAGGAATTGA
		841 CCGGGGCCCG CACAAGCAGC GGAGCATGTG GCTTAATTCC ACGCAACCGG AAGAACCCTTA
		901 CCAAGGCTTG ACATACACCG GAAACGCTCTG GAGACAGGGC CCCCCTTGTG GTCCGGTGTAC
		961 AGGTGGTGA TGGTGTCTGT CAGCTCGTGT CGTGAGATGT TGGTTAAGT CCCCACACGA
		1021 GCGCAACCC TGTCTGTGTG TGCCAGCATG CCCCTTCGGG TGATGGGGAC TCACAGGAGA
1081 CCGCCGGGGT CAACTCGGAG GAAGGTGGGG ACGACGTCFA GTCATCATGC CCCCTTATGTC		
1141 TTGGGCTGCA CACGTGTCTA AATGGCAGGT ACAATGAGCT GCGATACCGT GAGTGGGAGC		
1201 GAATCTCAA AAGCCTGTCT CAGTTCGGAT TTGGGCTCTG AACTCGACC CCATGAAGTC		
1261 GAGTTGCTAG TAATCGCAGA TCAGCATTGC TGCGGTGAA TACGTTCCCG GGCCTTGTAC		
1321 CACCGCCCGT CACGTCACGA AAGTCGGTAA CACCCGAAGC CCGTGGCCCA ACCCCTT		

c. Hasil Top 10 Hit BLAST terhadap NCBI, Excluding Uncultured Sample Sequences

No	Kode Sampel	Tautan Hasil						
		Description	Max Score	Total Score	Query Cover	E value	Per. Ident	Accession
1.	RKS-0.3-13	Streptomyces sp. strain CQC01 16S ribosomal RNA gene, partial sequence	2562	2562	100%	0.0	99.93%	MH748628.1
		Streptomyces neoceptini strain F18 16S ribosomal RNA gene, partial sequence	2562	2562	100%	0.0	99.93%	KU324439.1
		Streptomyces neoceptini strain 2-1-4-1-3 16S ribosomal RNA gene, partial sequence	2562	2562	100%	0.0	99.93%	OP108649.1
		Streptomyces sp. strain LK-Fr-3 16S ribosomal RNA gene, partial sequence	2562	2562	100%	0.0	99.93%	OQ456618.1
		Streptomyces sp. strain BCCO 10_2295 16S ribosomal RNA gene, partial sequence	2562	2562	100%	0.0	99.93%	ON493628.1
		Streptomyces neoceptini strain 12-8 16S ribosomal RNA gene, partial sequence	2562	2562	100%	0.0	99.93%	KJ571076.1
		Streptomyces sp. strain R391 16S ribosomal RNA gene, partial sequence	2556	2556	100%	0.0	99.86%	MH817409.1
		Streptomyces chartreusis strain NBC_00851 chromosome, complete genome	2556	15330	100%	0.0	99.86%	CP108840.1
		Streptomyces chartreusis strain NBC_00865 chromosome, complete genome	2556	15197	100%	0.0	99.86%	CP108820.1
		Streptomyces neoceptini strain T41 16S ribosomal RNA gene, partial sequence	2556	2556	100%	0.0	99.86%	KU317914.1
https://www.ncbi.nlm.nih.gov/nuccore/MH748628.1,KU324439.1,OP108649.1,OQ456618.1,ON493628.1,KJ571076.1,MH817409.1,CP108840.1,CP108820.1,KU317914.1								
2.	RKS-0.4-04	Streptomyces sp. strain CQC01 16S ribosomal RNA gene, partial sequence	2538	2538	100%	0.0	99.93%	MH748628.1
		Streptomyces neoceptini strain F18 16S ribosomal RNA gene, partial sequence	2538	2538	100%	0.0	99.93%	KU324439.1
		Streptomyces neoceptini strain 2-1-4-1-3 16S ribosomal RNA gene, partial sequence	2538	2538	100%	0.0	99.93%	OP108649.1
		Streptomyces sp. strain LK-Fr-3 16S ribosomal RNA gene, partial sequence	2538	2538	100%	0.0	99.93%	OQ456618.1
		Streptomyces sp. strain BCCO 10_2295 16S ribosomal RNA gene, partial sequence	2538	2538	100%	0.0	99.93%	ON493628.1
		Streptomyces neoceptini strain 12-8 16S ribosomal RNA gene, partial sequence	2538	2538	100%	0.0	99.93%	KJ571076.1
		Streptomyces neoceptini strain B451 16S ribosomal RNA gene, partial sequence	2534	2534	99%	0.0	99.93%	MK301165.1
		Streptomyces sp. strain R391 16S ribosomal RNA gene, partial sequence	2534	2534	99%	0.0	99.93%	MH817409.1
		Streptomyces chartreusis strain NBC_00851 chromosome, complete genome	2534	15197	99%	0.0	99.93%	CP108840.1
		Streptomyces chartreusis strain NBC_00865 chromosome, complete genome	2534	15047	99%	0.0	99.93%	CP108820.1
https://www.ncbi.nlm.nih.gov/nuccore/MH748628.1,KU324439.1,OP108649.1,OQ456618.1,ON493628.1,KJ571076.1,MK301165.1,MH817409.1,CP108840.1,CP108820.1								

d. Pohon Filogenik



Lampiran 2. Harmoni Warna

RAL 1000	RAL 1011	RAL 1019	RAL 1033	RAL 2009	RAL 3004	RAL 3015
RAL 1001	RAL 1012	RAL 1020	RAL 1034	RAL 2010	RAL 3005	RAL 3016
RAL 1002	RAL 1013	RAL 1021	RAL 2000	RAL 2011	RAL 3007	RAL 3017
RAL 1003	RAL 1014	RAL 1023	RAL 2001	RAL 2012	RAL 3009	RAL 3018
RAL 1004	RAL 1015	RAL 1024	RAL 2002	RAL 3000	RAL 3011	RAL 3020
RAL 1005	RAL 1016	RAL 1027	RAL 2003	RAL 3001	RAL 3012	RAL 3022
RAL 1006	RAL 1017	RAL 1028	RAL 2004	RAL 3002	RAL 3013	RAL 3027
RAL 1007	RAL 1018	RAL 1032	RAL 2008	RAL 3003	RAL 3014	RAL 3031

RAL 4001	RAL 4009	RAL 5008	RAL 5017	RAL 6000	RAL 6008	RAL 6016
RAL 4002	RAL 5000	RAL 5009	RAL 5018	RAL 6001	RAL 6009	RAL 6017
RAL 4003	RAL 5001	RAL 5010	RAL 5019	RAL 6002	RAL 6010	RAL 6018
RAL 4004	RAL 5002	RAL 5011	RAL 5020	RAL 6003	RAL 6011	RAL 6019
RAL 4005	RAL 5003	RAL 5012	RAL 5021	RAL 6004	RAL 6012	RAL 6020
RAL 4006	RAL 5004	RAL 5013	RAL 5022	RAL 6005	RAL 6013	RAL 6021
RAL 4007	RAL 5005	RAL 5014	RAL 5023	RAL 6006	RAL 6014	RAL 6022
RAL 4008	RAL 5007	RAL 5015	RAL 5024	RAL 6007	RAL 6015	RAL 6024

RAL 6025	RAL 7000	RAL 7009	RAL 7022	RAL 7034	RAL 7043	RAL 8008
RAL 6026	RAL 7001	RAL 7010	RAL 7023	RAL 7035	RAL 7044	RAL 8011
RAL 6027	RAL 7002	RAL 7011	RAL 7024	RAL 7036	RAL 8000	RAL 8012
RAL 6028	RAL 7003	RAL 7012	RAL 7026	RAL 7037	RAL 8001	RAL 8014
RAL 6029	RAL 7004	RAL 7013	RAL 7030	RAL 7038	RAL 8002	RAL 8015
RAL 6032	RAL 7005	RAL 7015	RAL 7031	RAL 7039	RAL 8003	RAL 8016
RAL 6033	RAL 7006	RAL 7016	RAL 7032	RAL 7040	RAL 8004	RAL 8017
RAL 6034	RAL 7008	RAL 7021	RAL 7033	RAL 7042	RAL 8007	RAL 8019

RAL 8022	RAL 8028	RAL 9003	RAL 9011
RAL 8023	RAL 9000	RAL 9004	RAL 9016
RAL 8024	RAL 9001	RAL 9005	RAL 9017
RAL 8025	RAL 9002	RAL 9010	RAL 9018

Die Farbangaben sind unverbindlich (Webdarstellung) und dienen lediglich der Orientierung.

Lampiran 3. Surat Izin Penelitian



**PEMERINTAH KABUPATEN MAROS
DINAS PENANAMAN MODAL, PELAYANAN TERPADU SATU PINTU DAN
KETENAGAKERJAAN**

Jl. Asoka No. 1 Telp. (0411)373884 Kabupaten Maros
email : admin@dpmpptsp.maroskab.go.id Website : www.dpmpptsp.maroskab.go.id

IZIN PENELITIAN

Nomor: 259/V/IP/DPMPPTSP/2023

DASAR HUKUM :

1. Undang-Undang Republik Indonesia Nomor 18 tahun 2002 tentang Sistem Nasional Penelitian, Pengembangan, dan Penerapan Ilmu Pengetahuan Teknologi;
2. Peraturan Menteri Dalam Negeri Nomor 7 Tahun 2014 tentang Perubahan Peraturan Menteri Dalam Negeri Nomor 64 Tahun 2011 tentang Pedoman Penerbitan Rekomendasi Penelitian;
3. Rekomendasi Tim Teknis Izin Penelitian Dinas Penanaman Modal dan Pelayanan Terpadu Satu Pintu Kabupaten Maros Nomor : 281/V/REK-IP/DPMPPTSP/2023

Dengan ini memberikan Izin Penelitian Kepada :

N a m a : AKMAL SAPUTRA ARDIONO
Nomor Pokok : NO12221007
Tempat/Tgl.Lahir : LANGI / 11 Juni 1999
Jenis Kelamin : Laki-Laki
Pekerjaan : MAHASISWA
Alamat : JL. TAMANGAPA RAYA 5 NO. 23 MAKASSAR
Tempat Meneliti : KARST BANTIMURUNG

Maksud dan Tujuan mengadakan penelitian dalam rangka Penulisan Tesis dengan Judul :

**"ISOLASI DAN IDENTIFIKASI MOLEKULER ACTINOMYCETES TANAH RIZOSFER
Peperomia pellucida L. DARI EKOSISTEM KARST SEBAGAI HASIL SENYAWA
ANTIBIOTIK"**

Lamanya Penelitian : 01 Juni 2023 s/d 01 Januari 2023

Dengan ketentuan sebagai berikut :

1. Mentaati semua peraturan perundang-undangan yang berlaku, serta menghormati Adat Istiadat setempat.
2. Penelitian tidak menyimpang dari maksud izin yang diberikan.
3. Menyerahkan 1 (satu) exemplar Foto Copy hasil penelitian kepada Dinas Penanaman Modal dan Pelayanan Terpadu Satu Pintu Kabupaten Maros.
4. Surat Izin Penelitian ini dinyatakan tidak berlaku, bilamana pemegang izin ternyata tidak mentaati ketentuan-ketentuan tersebut diatas.

Demikian Izin Penelitian ini diberikan untuk dipergunakan sebagaimana mestinya.



Maros, 29 Mei 2023

KEPALA DINAS,



NURYADI, S. Sos., M. A. P

Pangkat : Pembina Tk. I

Nip : 19741005 199803 1 010

Tembusan Kepada Yth.:

1. Dekan Fakultas Farmasi UNHAS di Makassar
2. Arsip

*Dokumen Ini Telah Ditandatangani Secara Elektronik Menggunakan Sertifikat Elektronik
iOTENTIK Badan Riset dan Inovasi Nasional (BRIN)*