

BAB V

KESIMPULAN DAN SARAN

A. Kesimpulan

Berdasarkan penelitian yang telah dilakukan maka dapat diambil kesimpulan bahwa :

Pertama, bakteri *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853, *Salmonella typhi* ATCC 13311, *Serratia marcescens* ATCC 8100, dan *Staphylococcus aureus* ATCC 25923 menghasilkan enzim amilase.

Kedua, dari kelima amilase bakteri yang memiliki aktivitas enzim amilase paling besar adalah *Escherichia coli* ATCC 25922 dengan diameter zona bening sebesar 19,79 mm.

Ketiga, karakteristik *Escherichia coli* ATCC 25922 memiliki aktivitas paling besar pada suhu 30°C dan pH 8. *Pseudomonas aeruginosa* ATCC 27853 memiliki aktivitas paling besar pada suhu 50°C dan pH 8. *Salmonella typhi* ATCC 13311 memiliki aktivitas paling besar pada suhu 50°C dan pH 8. *Serratia marcescens* ATCC 8100 memiliki aktivitas paling besar pada suhu 50°C dan pH 8. *Staphylococcus aureus* ATCC 25923 memiliki aktivitas paling besar pada suhu 50°C dan pH 8.

B. Saran

Perlu dilakukan penelitian lanjutan untuk sampai pada tahap pemurnian enzim amilase dan pengukuran kadar protein yang terkandung, karena yang dilakukan dalam penelitian ini masih berupa ekstrak kasar amilase saja. Dan juga pada penelitian aktivitas enzim harus lebih teliti dan memahami faktor apa saja yang mempengaruhi aktivitas enzim selain pH, suhu, dan substrat.

DAFTAR PUSTAKA

- Allocati N, Massuli M, Alexeyev MF, Ilio CD. 2013. Escherichia coli in Europe : An Overview. *Int. J. Environ. Res Public Health* 10:6235-6254.
- Amri E, Widhyastuti N, Artika M. 2010. Aktivitas Amilase Bakteri yang Diisolasi dari Sumber Air Panas Ciseeng Bogor. *Jurnal Sainstek* 2:23-33.
- Arfah RA, Ahmad A, Djide MN, Anis M, Zakir M. 2015. Production Optimization and Characterization of Amylase Enzyme Isolated from Termofil Bacteria *Bacillus sp* RSAII-1b from Lejja Hot Spring South Sulawesi. *American Journal of Biomedical dan Life Sciences* 3:115-119.
- Bintang M. 2010. *Biokimia Teknik Penelitian*. Jakarta : Penerbit Erlangga.
- Bole S, Maji A, Dey A, Acharya A, Dubey S, Rakeslal. 2013. Isolation, Purification And Characterization Of Amylase From Airborne-Bacteria. *World Journal Of Pharmacy And Pharmaceutical Sciences* 2 (6).
- Cui SW, Liu Q, Xie SX. 2005. *Starch Modification and Application*. Taylor & Francis Group, LLC.
- Das S, Singh S, Sharma V, Soni ML. 2011. Biotechnological Applications Of Industrially Important Amylase Enzyme. *International Journal Of Pharma Bio Sciences* 2:486-496.
- Dehkordi MM, Javan FA. 2012. Application of Alpha-amylase in Biotechnology. *Journal of Biology and Today's World* 1:39-50.
- Dewi AK. 2013. Isolasi, Identifikasi dan Uji Sensitivitas Staphylococcus aureus terhadap Amoxicillin dari Sampel Susus Kambing Peranakan Ettawa (PE) Penderita Mastitis Di Wilayah Girimulyo, Kulonprogo, Yogyakarta. *Jurnal Sain Veteriner* 31:138-150.
- Elmarzugi NA, El Enshasy HA, Hamid MA, Hasham R, Aziz E, Elsayed EA, Othman NZ, Salama M. A Amylase Economic And Application Value. 2014. *World Journal of Pharmaceutical Research* 3:4890-4906.
- Fitriani A, Supriyanti FMT, Heryanto TE. 2013. Penentuan Aktivitas Amilase Kasar Termofil *Bacillus Subtilis* Isolat Kawah Gunung Darajat Garut, Jawa Barat. *Bionatura Jurnal Ilmu-ilmu Hayati dan Fisik* 15:107-113.
- Flyg C, Kenne K, Boman HG. 1983. Insect athogenic properties of *Serratia marcescens* ; phage- resistant mutants with a decreased resistance to *Cecropia* immunity and a decreased virulence to *Drosophila*. *Journal of General Microbiology* 120:173-181.

- Geciova J, Bury D, Jelen P. 2002. Methods For Disruption Of Microbial Cells For Potential Use In The Dairy Industry: A Review. *International Dairy Journal* 12:541-553.
- Giri AV, Anandkumar N, Muthukumar G, Pennathur G. 2004. A novel medium for the enhanced cell growth and production of prodigiosin from *Serratia marcescens* isolated from soil. *BMC Microbiology* 4:1-10.
- Harti AS. 2015. *Mikrobiologi Kesehatan*. Jakarta : Andi.
- Hayashida S, Teramoto Y, Inoue T. 1988. Production and Characteristic of Raw Potato-Starch-Digesting α -Amylase from *Bacillus subtilis* 65. *Applied and Environmental Microbiology* 54:1516-1522.
- [ISFI] Innovative Scientific Futuristic Informative. 2014. *Informasi Spesialite Obat Indonesia Vol. 49*. Jakarta : PT. ISFI.
- Jansen EH. 2015. Role of Biomarkers In Diagnosis and Prognostic Evaluation of Acute Pancreatitis. *Journal of Biomarkers*
- Jawetz E, Melnick JL, Adelberg EA. 2013. *Mikrobiologi Kedokteran, Edisi 25*. Jakarta: Buku Kedokteran EGC.
- Kandra L. 2003. α -Amylases of Medical and Industrial importance. *Journal of Molecular Structure* 487-489.
- Kaneko T, Ohno T, Ohisa N. 2005. Purification and Characterization of a Thermostable Raw Starch Digesting Amylase from a *Sterptomyces* sp. Isolated in a Milling Factory. *Biotechnol* 69:1037-1081.
- Khanafari A, Assadi MM, Fakhr FA. 2006. Review of Prodigiosin, Pigmentation in *Serratia marcescens*. *Journal of Biological Sciences* 6:1-13.
- Li, M. Liao, X. Zhang, D. Du, G. Chen, J. Yeast Extract Promotes Cell Growth and Induces Production of Polyvinyl Alcohol-Degrading Enzymes. *Enzyme Research*.
- Liu, Q. 2005. *Understanding Starch and Their Role in Foods*. Taylor & Francis Group, LLC.
- Maity S, Mallik S, Gupta S. 2015. Optimization of Solid State Fermentation Conditions and Characterization of Thermostable Alpha Amylase from *Bacillus subtilis*(ATCC 6633). *Journal of Bioprocessing & Biotechniques* 5 (4).
- Naiola E. 2006. Karakterisasi Enzim Kasar Glukoamilase Dari *Saccharomycopsis Sp.* *Berita Biologi* 8:187-192.

- Nangin D, Sutrisno A. 2015. Enzim Amilase Pemecah Pati Mentah Dari Mikroba : Kajian Pustaka. *Jurnal Pangan dan Agroindustri* 3:1032-1039.
- Ningsih DR, Rastuti U, Kamaludin R. 2012. Karakterisasi Enzim Amilase Dari Bakteri *Bacillus amyloliquefaciens*. *Pengembangan Sumber Daya Pedesaan dan Kearifan Lokal Berkelanjutan II. Prosiding Seminar Nasional: Purwokerto, 27-28 November 2012.*
- [NIOS] National Institute of Open Schooling. 1989. *Module Microbiology : Escherichia coli and Klebsiella*. India : NIOS.
- Peclzar MJ, Chan ECS. 1988. *Dasar-Dasar Mikrobiologi*. Jakarta : Universitas Indonesia.
- Phutucheary SD, Ngeow YF. 1981. *Serratia Marcescens : Biochemical Characteristics, Antimikrobal Sensitivity And Clinical Sinnificance*. *Malaysian J Pathol* 4:35-41.
- Poedjiadi A. 1994. *Dasar-dasar Biokimia*. Jakarta : Universitas Indonesia.
- Pollack RA, Findlay L, Mondschein W, Modesto RR. 2016. *Praktik Laboratorium Mikrobiologi Edisi 4*. Jakarta : Penerbit Buku Kedokteran.
- Radji M. 2010. *Buku Ajar Mikrobiologi*. Jakarta : Buku Kedokteran EGC.
- Reddy N.S, Nimmagadda A, Rao KR. 2003. An Overview of Themicrobial α amylase Family. *African Journal of Biotechnology* 2:645-648.
- Rokade MT, Archana. 2017. Isolation, Molecular Characterization And Phylogenic Analysis Of *Serratia marcescens* Isolated From Soil. *Ijariie* 3:3141-3147.
- Rosilawati E, Christinasari YC, Koestanti E. 2011. Isolasi Identifikasi serta Uji Kepekaan *Bacillus subtilis* dari Sedimen Tambak Udang Terhadap Bahan Antimikrobal. *Veterinara Medika* 4 (2).
- Sadikin M. 2002. *Biokimia Enzim*. Jakarta : Widya Medika.
- Setyorini, V.P. 2017. *Menristekdikti Resmikan Unit Produksi Enzim Pertama Indonesia*. ANTARANEWS.COM. Diakses 10 Oktober 2018.
- Sivaramakrishnan S., Gangadharan D., Nampoothiri, C.R. Sossol, and A. Pandey. 2006. α -amylase from Microbial Sources – An Overview on Recent Developments. *Food. Technol. Biotechnol Journal* 44:173-184.
- Suriawiria U. 1985. *Pengantar Mikrobiologi Umum*. Bandung: Angkasa.
- Susanti R, Fibriana F. 2017. *Teknologi Enzim*. Yogyakarta : Penerbit Andi.

- Sutrisno A. 2017. *Teknologi Enzim*. Jakarta : UB Press.
- Syowiecki J. 2007. *Industrial Enzymes: Structure, Function and Applications: The Use Of Starch Processing Enzymes In The Food Industry*. Netherlands: Springer.
- Todar K. 2008. *Salmonella*. *Todar's Online textbook of Bacteriology*. University of Wisconsin-Madison Departemen Bacteriology. Wisconsin.
- Uchendu IK, Agu CE, Orji OC, Offor MC, Nwosu TF. 2017. Review On Diagnosis Of Acute Pancreatitis. *Journal of Medical & Allied Sciences* 7:76-81.
- Varalakshmi KN, Pallavi NU, Arpita B, Bhuvaneshwari S, Mrudula P, Priyanka G. 2012. Production and partial purification of α -amylase from *Pseudomonas* sp. 2 under solid-state fermentation. *Turkish Journal of Biochemistry* 1:21-28.
- Yoo YJ, Hong J, Hatch RT. 1986. Comparison of α -Amylase Activities from Different Assay Methods. *Biotechnology and Bioengineering* XXX:147-151.

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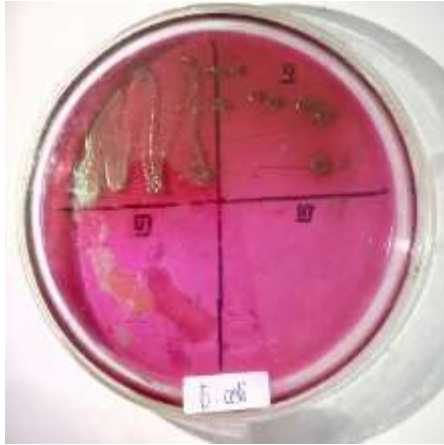
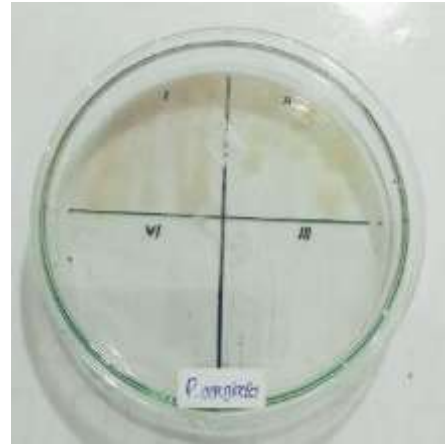
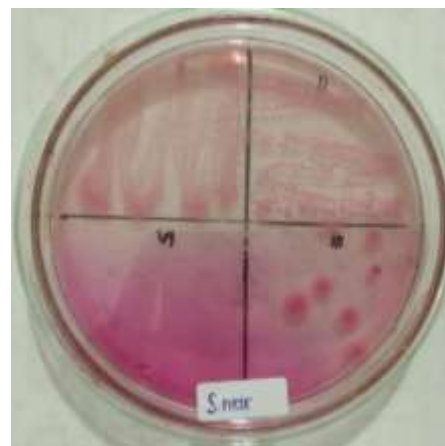
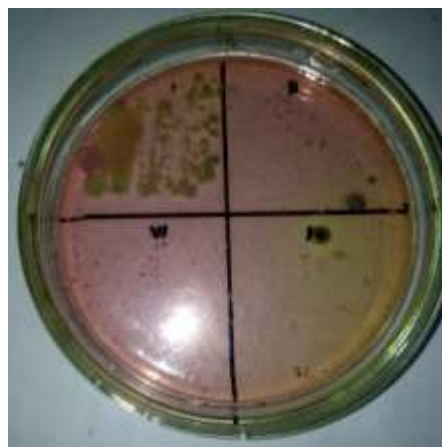
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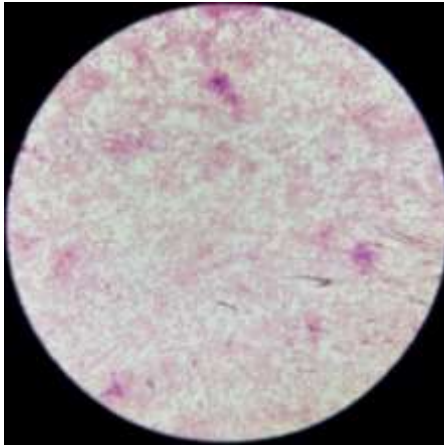
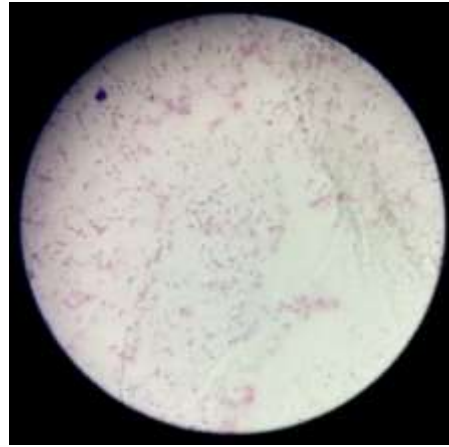
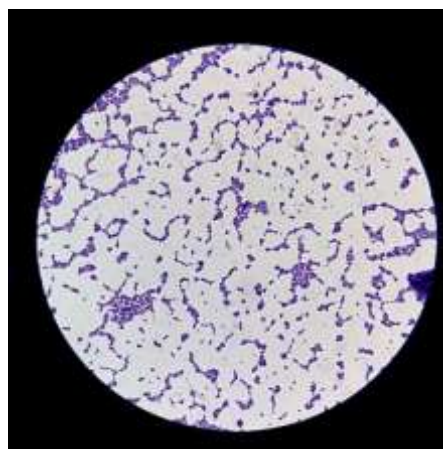
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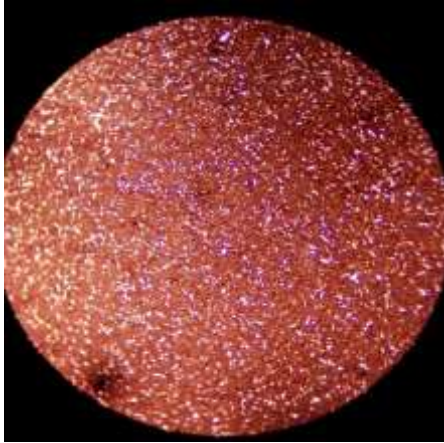
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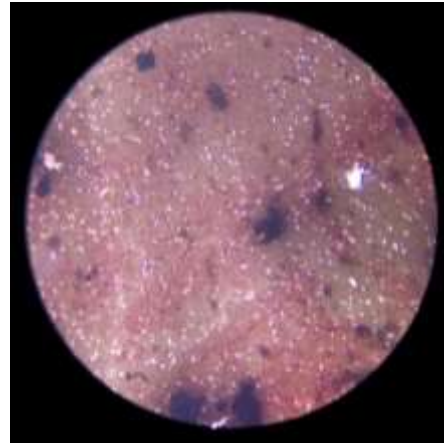
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Lampiran 1. Hasil Identifikasi Bakteri berdasarkan KoloniKoloni *E. coli*Koloni *P. aeruginosa*Koloni *S. typhi*Koloni *S. marcescens*Koloni *S. aureus*

Lampiran 2. Hasil Pewarnaan Gram Bakteri*E. coli**P. aeruginosa**S. typhi**S. marcescens**S. aureus*

















Lampiran 3. Hasil Pewarnaan Kapsul

P. aeruginosa



S. typhi

Lampiran 4. Hasil Uji Biokimia

Bakteri	SIM	KIA	LIA	SCA
<i>E. coli</i>				
<i>P. aeruginosa</i>				
<i>S. typhi</i>				
<i>S. marcescens</i>				

Lampiran 5. Hasil Uji Katalase dan Uji Koagulase *S. aureus*



Uji Katalase

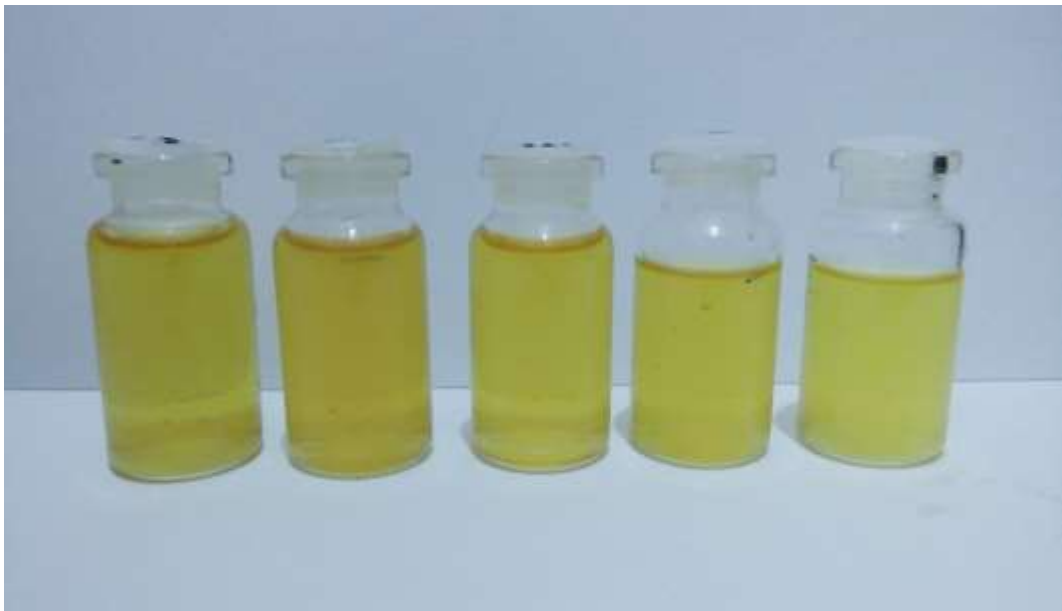


Uji Koagulase

Lampiran 6. Produksi Enzim Amilase

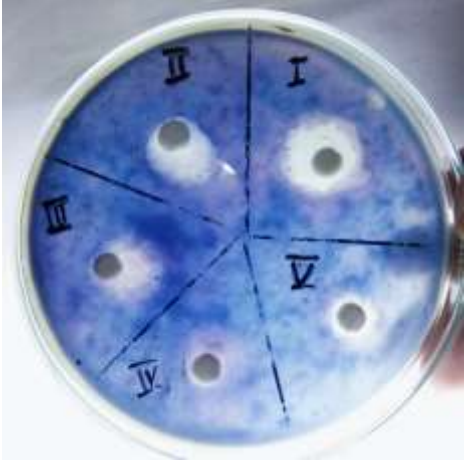
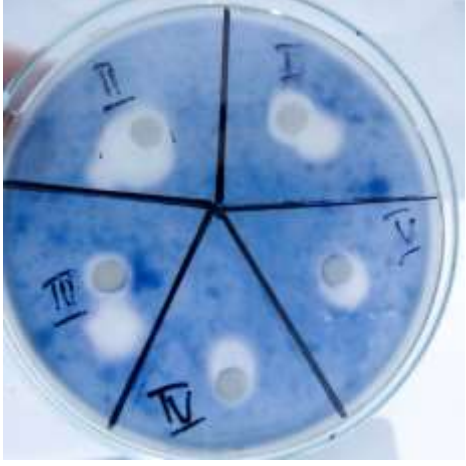



Fermentasi bakteri pada media produksi enzim



Ekstrak kasar enzim amilase setelah disentrifugasi

Lampiran 7. Hasil Zona Bening Amilase Bakteri

Replikasi	Zona Bening
1	
2	
3	

Lampiran 8. Larutan Buffer



Pengukuran pH



Larutan buffer sitrat (pH 4-6) dan buffer fosfat (pH 7-8)

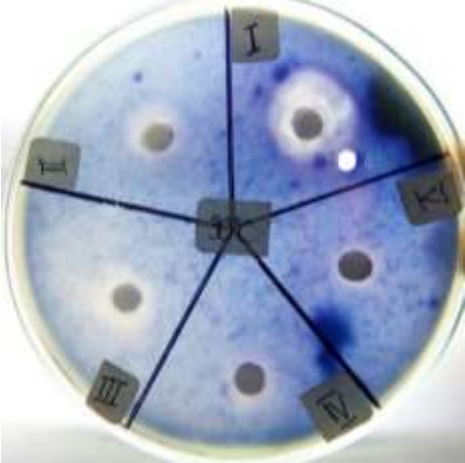
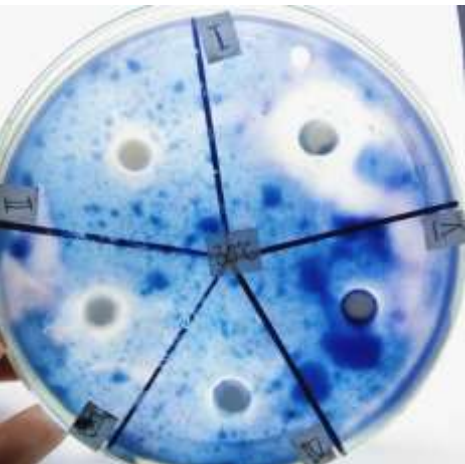
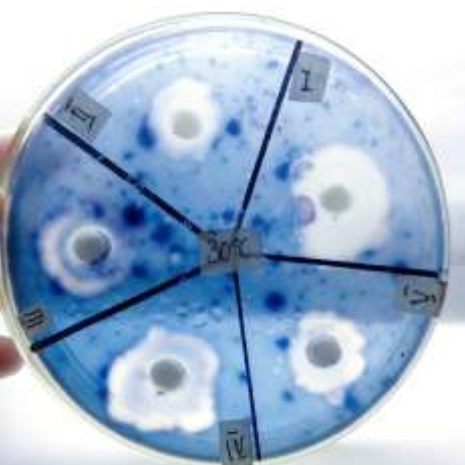
Lampiran 9. Ekstrak Kasar Amilase Variasi Suhu

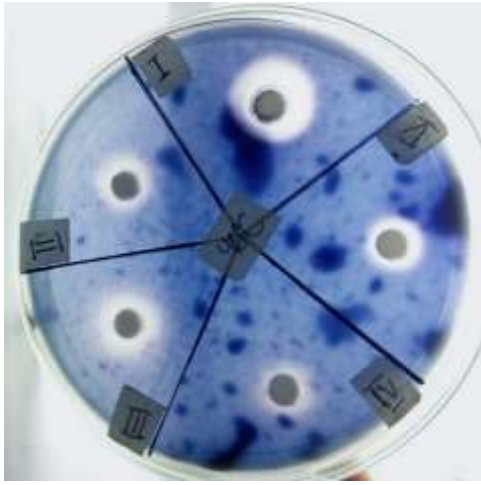
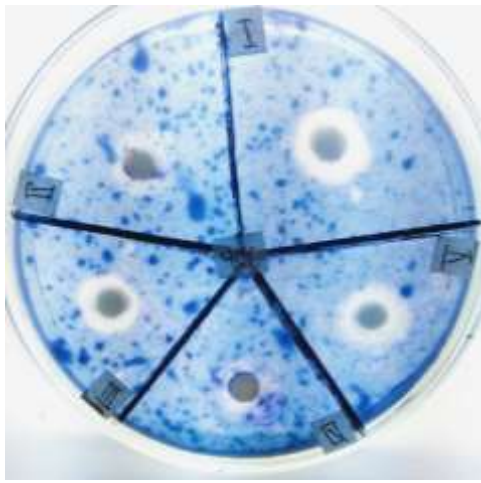
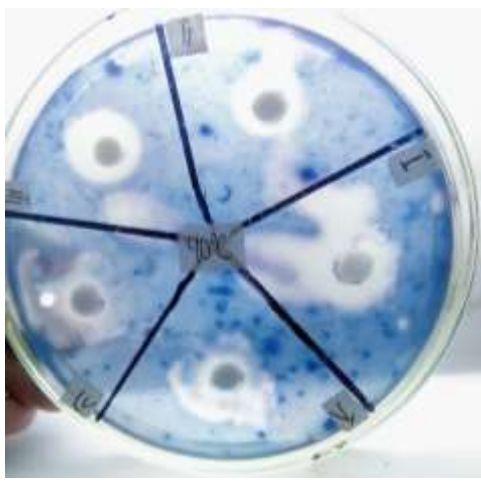


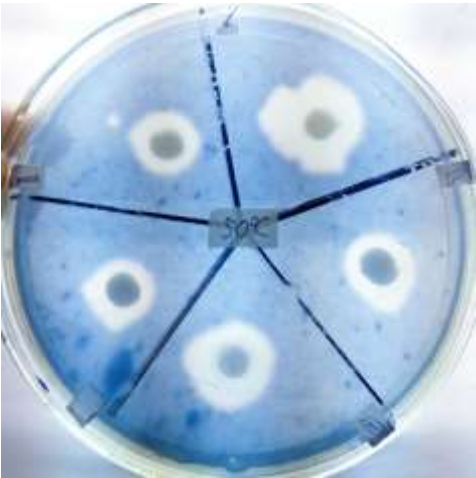
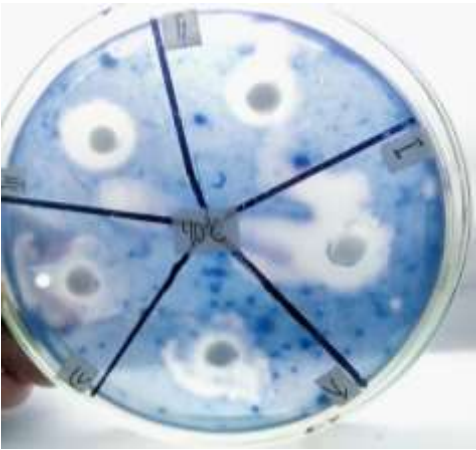
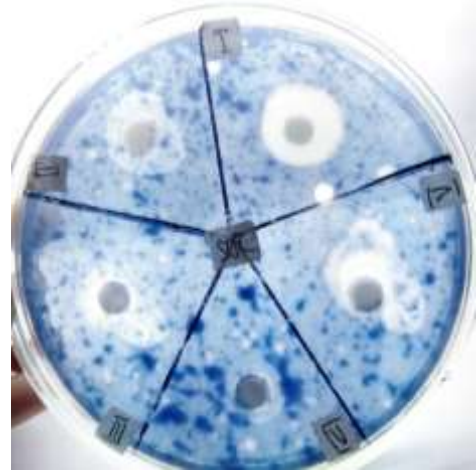
Lampiran 10. Ekstrak Kasar Amilase Variasi pH

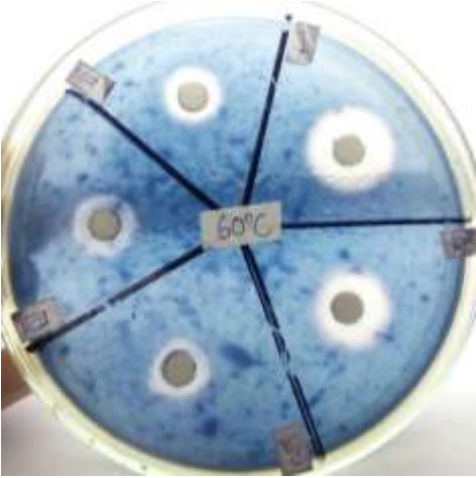
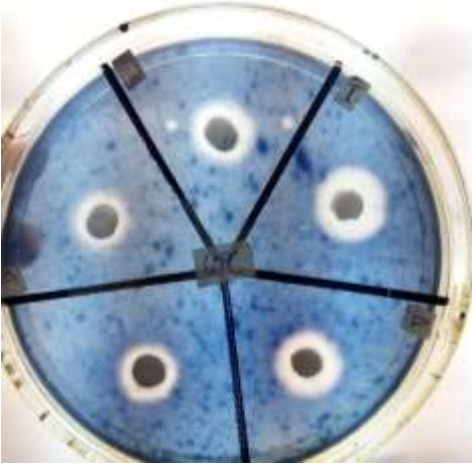
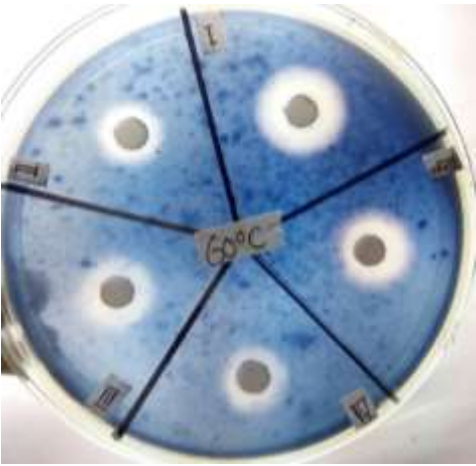


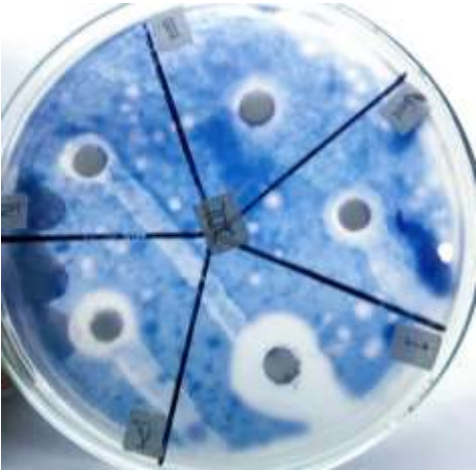
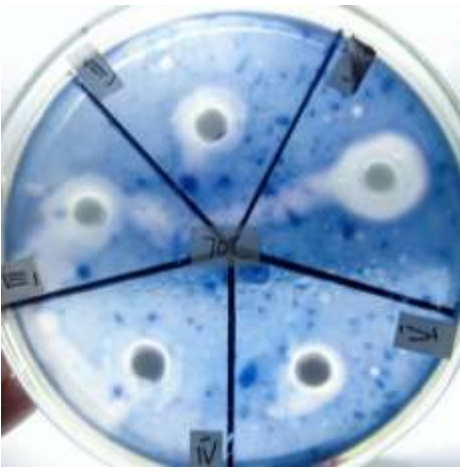
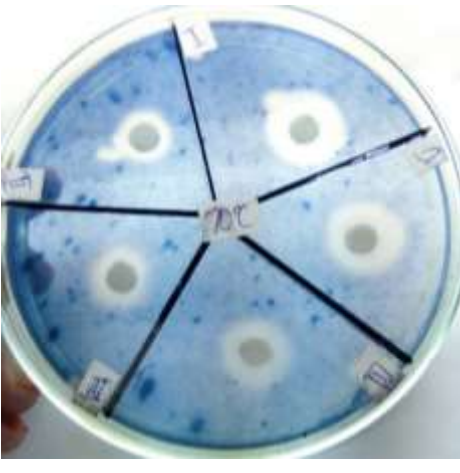
Lampiran 11. Zona Bening Aktivitas Amilase terhadap Suhu

Replikasi	Zona Bening
1	 A petri dish containing five replicates of amylase activity, labeled I through V. The medium is blue, and each replicate shows a distinct, clear, circular zone of activity. The zones are roughly circular and vary slightly in size. The background medium is a uniform blue color.
2	 A petri dish containing five replicates of amylase activity, labeled I through V. The medium is blue, and each replicate shows a clear, circular zone of activity. The zones are smaller and more irregular in shape compared to the 30°C condition. The background medium is a uniform blue color.
3	 A petri dish containing five replicates of amylase activity, labeled I through V. The medium is blue, and each replicate shows a very small, clear, circular zone of activity. The zones are significantly smaller and less distinct than those at 30°C and 40°C. The background medium is a uniform blue color.

Replikasi	Zona Bening
1	
2	
3	

Replikasi	Zona Bening
1	
2	
3	

Replikasi	Zona Bening
1	 A petri dish containing blue agar with six wells. A small label in the center reads "60°C". The wells are arranged in a circular pattern around the center. The agar shows some faint, irregular white spots, possibly indicating the beginning of a zone of inhibition.
2	 A petri dish containing blue agar with six wells. A small label in the center reads "60°C". The wells are arranged in a circular pattern around the center. The agar shows some faint, irregular white spots, possibly indicating the beginning of a zone of inhibition.
3	 A petri dish containing blue agar with six wells. A small label in the center reads "60°C". The wells are arranged in a circular pattern around the center. The agar shows some faint, irregular white spots, possibly indicating the beginning of a zone of inhibition.

Replikasi	Zona Bening
1	
2	
3	

Lampiran 12. Hasil Pengukuran Diameter Zona Bening Amilase terhadap Suhu

Karakterisasi suhu 30°C

Bakteri	Diameter Zona Bening (mm)			Rata-rata (mm)	SD
	Replikasi 1	Replikasi 2	Replikasi 3		
<i>E. coli</i>	19,65	21,98	24,60	22,08	2.48
<i>P. aeruginosa</i>	12,38	20,08	18,38	16,95	4.06
<i>S. typhi</i>	17,63	12,98	19,93	16,85	3.54
<i>S. marcescens</i>	13,48	13,18	23,23	16,63	5.72
<i>S. aureus</i>	12,25	12,13	17,33	13,90	2.97

Karakterisasi suhu 40°C

Bakteri	Diameter Zona Bening (mm)			Rata-rata (mm)	SD
	Replikasi 1	Replikasi 2	Replikasi 3		
<i>E. coli</i>	19,03	17,55	25,38	20,65	4,16
<i>P. aeruginosa</i>	15,58	14,78	15,78	15,38	0,53
<i>S. typhi</i>	17,45	19,15	18,58	18,39	0,87
<i>S. marcescens</i>	12,35	14,90	20,88	16,04	4,38
<i>S. aureus</i>	11,20	17,15	19,45	15,93	4,23

Karakterisasi suhu 50°C

Bakteri	Diameter Zona Bening (mm)			Rata-rata (mm)	SD
	Replikasi 1	Replikasi 2	Replikasi 3		
<i>E. coli</i>	18,55	18,68	20,18	19,14	0,91
<i>P. aeruginosa</i>	17,85	18,00	15,90	17,25	1,17
<i>S. typhi</i>	17,65	22,56	15,63	18,61	3,56
<i>S. marcescens</i>	13,85	26,88	19,10	19,94	6,56
<i>S. aureus</i>	20,38	17,95	16,45	18,26	1,98

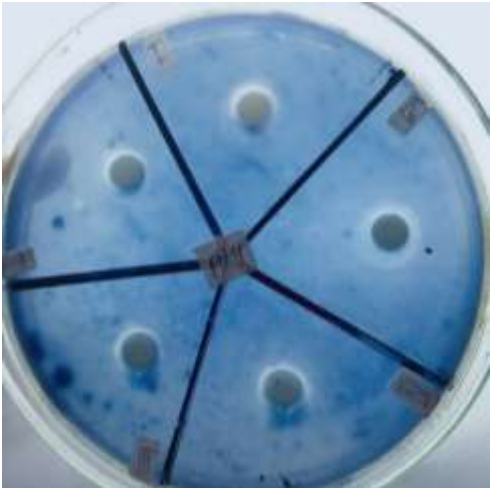
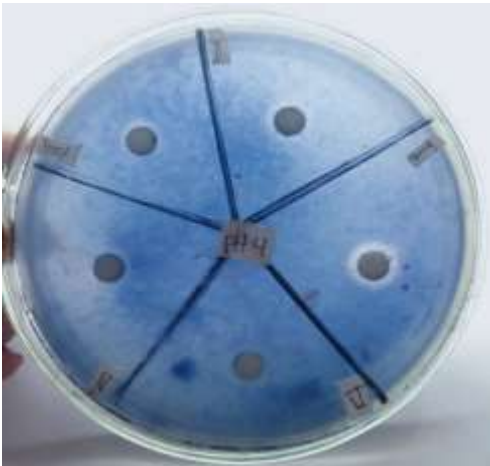
Karakterisasi suhu 60°C

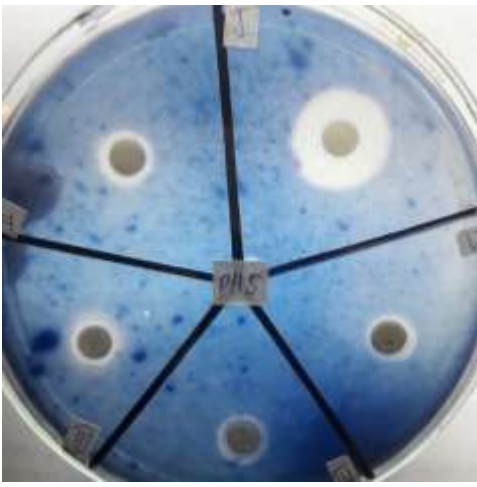
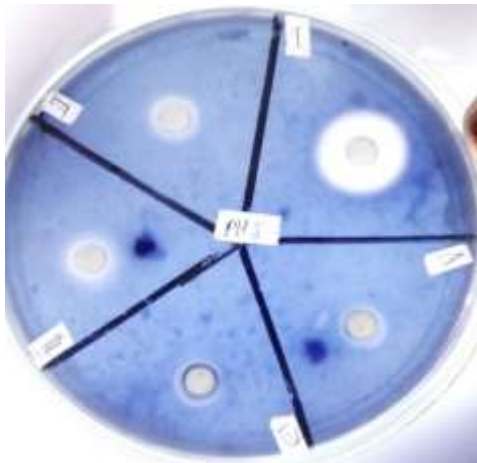

Bakteri	Diameter Zona Bening (mm)			Rata-rata (mm)	SD
	Replikasi 2	Replikasi 2	Replikasi 3		
<i>E. coli</i>	19,23	16,35	18,30	17,96	1,47
<i>P. aeruginosa</i>	14,25	14,20	13,08	13,84	0,66
<i>S. typhi</i>	14,40	13,88	13,78	14,02	0,33
<i>S. marcescens</i>	12,33	12,63	12,15	12,37	0,24
<i>S. aureus</i>	17,68	15,55	15,43	16,22	1,27

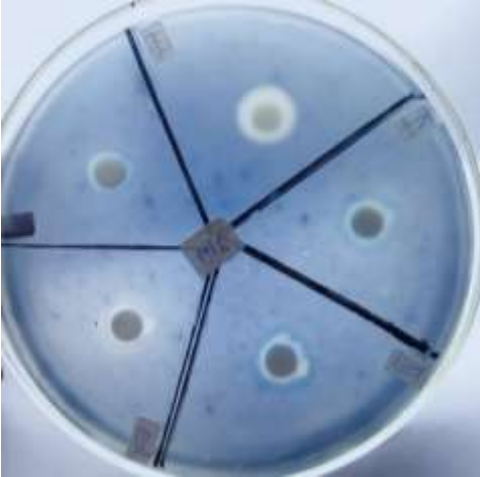
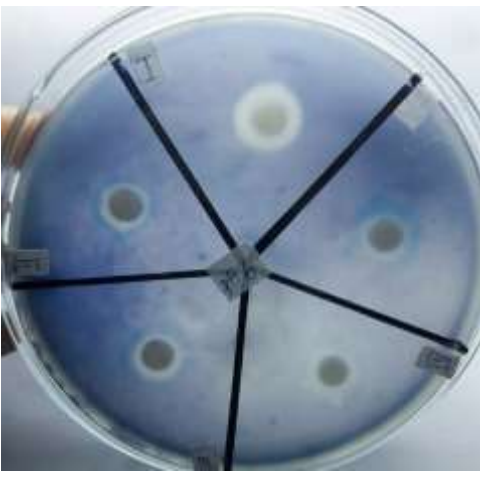
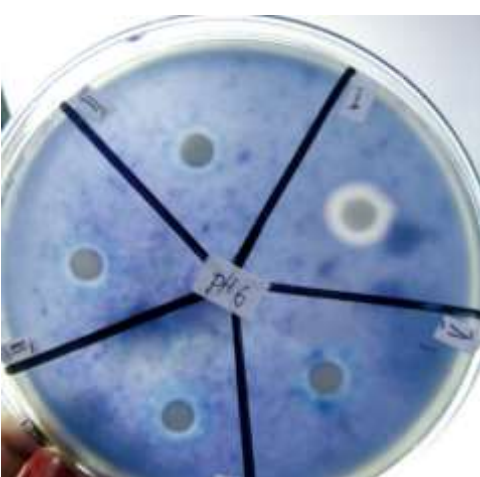
Karakterisasi suhu 70°C

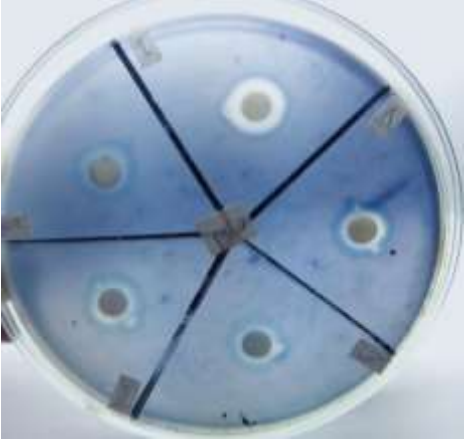
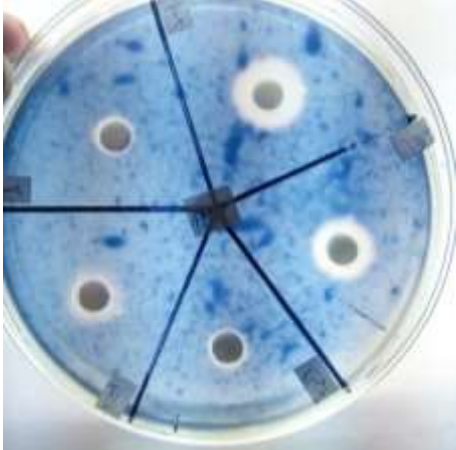
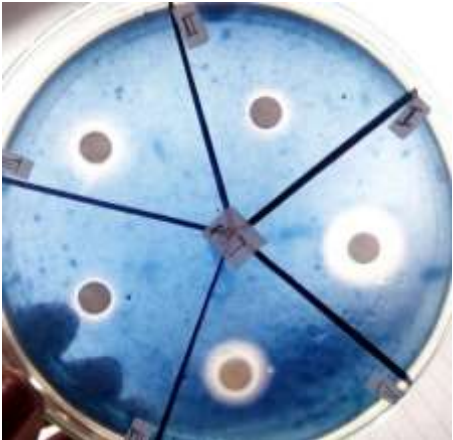
Bakteri	Diameter Zona Bening (mm)			Rata-rata (mm)	SD
	Replikasi 1	Replikasi 2	Replikasi 3		
<i>E. coli</i>	22,08	19,75	19,25	20,33	1,51
<i>P. aeruginosa</i>	12,55	14,43	14,33	13,77	1,06
<i>S. typhi</i>	11,53	19,05	16,15	15,58	3,79
<i>S. marcescens</i>	10,33	18,60	17,30	15,41	4,45
<i>S. aureus</i>	13,95	15,25	16,88	15,36	4,47

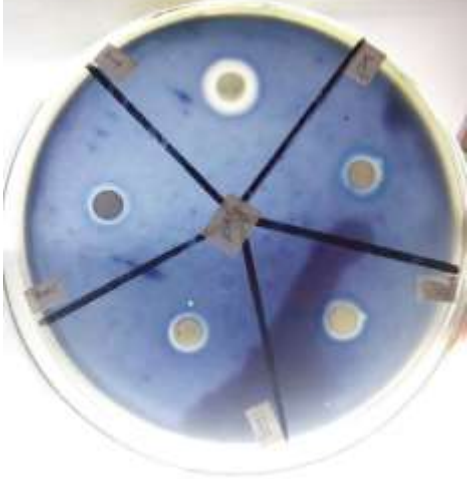
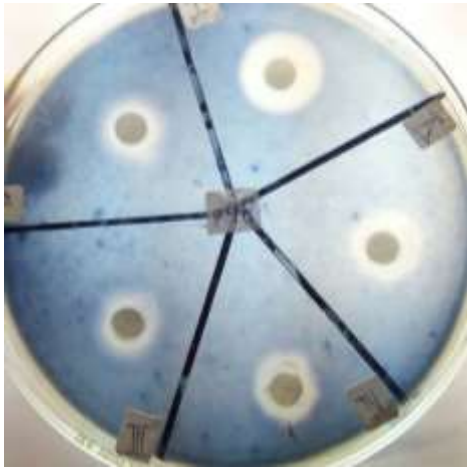
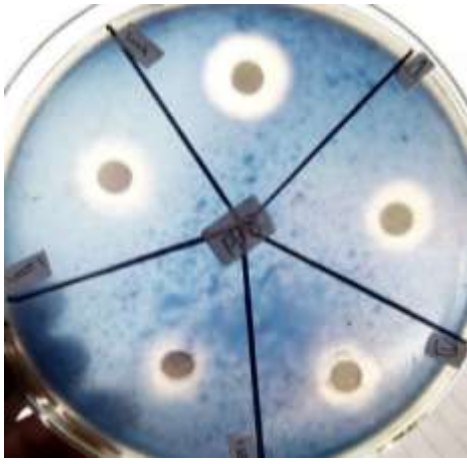
Lampiran 13. Zona Bening Aktivitas Amilase terhadap pH

Replikasi	Zona Bening
1	
2	

Replikasi	Zona Bening
1	
2	
3	

Replikasi	Zona Bening
1	 A petri dish with a blue agar surface, divided into five sectors by black lines. A central square label reads 'PH 6'. Each sector contains a circular well. The top well shows a distinct, clear, circular zone of inhibition, while the other four wells show no such zone.
2	 A petri dish with a blue agar surface, divided into five sectors by black lines. A central square label reads 'PH 6'. Each sector contains a circular well. The top well shows a distinct, clear, circular zone of inhibition, while the other four wells show no such zone.
3	 A petri dish with a blue agar surface, divided into five sectors by black lines. A central square label reads 'PH 6'. Each sector contains a circular well. The top well shows a distinct, clear, circular zone of inhibition, while the other four wells show no such zone.

Replikasi	Zona Bening
1	
2	
3	

Replikasi	Zona Bening
1	
2	
3	

Lampiran 14. Hasil Pengukuran Diameter Zona Bening Amilase terhadap pH

Karakterisasi pH 4

Bakteri	Diameter Zona Bening (mm)			Rata-rata (mm)	SD
	Replikasi 1	Replikasi 2	Replikasi 3		
<i>E. coli</i>	9,93	8,90	9,20	9,34	0,53
<i>P. aeruginosa</i>	0	0	0	0	0
<i>S. typhi</i>	0	0	0	0	0
<i>S. marcescens</i>	0	0	0	0	0
<i>S. aureus</i>	0	0	0	0	0

Karakterisasi pH 5

Bakteri	Diameter Zona Bening (mm)			Rata-rata (mm)	SD
	Replikasi 1	Replikasi 2	Replikasi 3		
<i>E. coli</i>	16,18	15,53	16,68	16,13	0,58
<i>P. aeruginosa</i>	8,70	9,13	9,38	9,07	0,34
<i>S. typhi</i>	8,80	8,80	10,38	9,33	0,91
<i>S. marcescens</i>	9,23	9,58	9,90	9,57	0,34
<i>S. aureus</i>	8,73	8,95	10,18	9,29	0,78

Karakterisasi pH 6

Bakteri	Diameter Zona Bening (mm)			Rata-rata (mm)	SD
	Replikasi 1	Replikasi 2	Replikasi 2		
<i>E. coli</i>	13,90	13,20	12,83	13,31	0,54
<i>P. aeruginosa</i>	9,83	10,13	9,75	9,90	0,20
<i>S. typhi</i>	8,75	10,95	9,60	9,77	1,11
<i>S. marcescens</i>	10,45	10,00	9,60	10,02	0,43
<i>S. aureus</i>	10,25	10,38	8,98	9,87	0,77

Karakterisasi pH 7

Bakteri	Diameter Zona Bening (mm)			Rata-rata (mm)	SD
	Replikasi 1	Replikasi 2	Replikasi 3		
<i>E. coli</i>	13,85	15,25	15,58	14,89	0,92
<i>P. aeruginosa</i>	12,48	9,38	8,23	10,03	2,20
<i>S. typhi</i>	10,50	11,00	10,18	10,56	0,41
<i>S. marcescens</i>	9,95	9,05	8,95	9,32	0,55
<i>S. aureus</i>	10,18	13,15	12,85	12,06	1,64

Karakterisasi pH 8

Bakteri	Diameter Zona Bening (mm)			Rata-rata (mm)	SD
	Replikasi 1	Replikasi 2	Replikasi 3		
<i>E. coli</i>	15,90	16,55	16,90	16,45	0,51
<i>P. aeruginosa</i>	9,20	13,03	14,05	12,09	2,56
<i>S. typhi</i>	8,78	12,13	12,30	11,07	1,99
<i>S. marcescens</i>	9,80	12,68	12,23	11,57	1,55
<i>S. aureus</i>	9,95	13,95	13,20	12,37	2,13

Lampiran 15. Hasil Analisis Data Karakterisasi Suhu dengan SPSS

Uji kolmogorov Smirnov

Tujuan : mengetahui kenormalan data sebagai syarat uji ANOVA

Kriteria uji : Sig. < 0,05 berarti H0 ditolak Sig. > 0,05 H0 diterima

Hasil:

One-Sample Kolmogorov-Smirnov Test		Aktivitas Amilase Suhu
N		75
Normal Parameters ^{a,b}	Mean	16.7857
	Std. Deviation	3.49584
	Absolute	.070
Most Extreme Differences	Positive	.070
	Negative	-.051
Kolmogorov-Smirnov Z		.605
Asymp. Sig. (2-tailed)		.858

a. Test distribution is Normal.

b. Calculated from data.

Kesimpulan : sig. 0,858 > 0,05 (H0 diterima) maka data diameter zona bening (aktivitas amilase) terdistribusi normal.

Uji Levene

Tujuan : untuk mengetahui homogenitas

Kriteria uji : Sig. < 0,05 berarti H0 ditolak. Sig. > 0,05 H0 diterima

Hasil :

Levene's Test of Equality of Error Variances^a

Dependent Variable: Aktivitas Amilase Suhu

F	df1	df2	Sig.
2.968	24	50	.001

Tests the null hypothesis that the error variance of the dependent variable is equal across groups.

a. Design: Intercept + Bakteri + Suhu + Bakteri *

Suhu

Kesimpulan : Sig. 0,001 < 0,05 (H0 ditolak) maka masing-masing kelompok data aktivitas amilase berasal dari populasi dengan varians yang berbeda (tidak homogen).

Uji Two Way Anova

Tests of Between-Subjects Effects

Dependent Variable: Aktivitas Amilase Suhu

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	444.534 ^a	24	18.522	2.014	.019
Intercept	21132.063	1	21132.063	2297.888	.000
Suhu	126.588	4	31.647	3.441	.015
Bakteri	210.583	4	52.646	5.725	.001
Suhu * Bakteri	107.362	16	6.710	.730	.751
Error	459.815	50	9.196		
Total	22036.412	75			
Corrected Total	904.348	74			

a. R Squared = .492 (Adjusted R Squared = .247)

Kesimpulan :

1. Faktor pertama bakteri (amilase) : karena signifikansi nilainya 0,015 < 0,05 (H0 ditolak), artinya dari ke lima bakteri tersebut memiliki aktivitas amilase yang tidak sama.
2. Faktor kedua suhu (amilase) : karena signifikansi nilainya 0,001 < 0,05 (H0 ditolak), artinya pengaruh suhu terhadap aktivitas amilase dari ke lima bakteri tersebut tidak sama.
3. Faktor interaksi/ hubungan (aktivitas amilase dengan pemberian variasi suhu) : karena signifikansi nilainya adalah 0,751 > 0,05 (H0 diterima), artinya amilase yang dihasilkan oleh ke lima bakteri tersebut memiliki interaksi dengan pemberian pengaruh suhu.

Aktivitas Amilase Suhu

Tukey HSD^{a,b}

Suhu	N	Subset	
		1	2
60oC	15	14.8827	
70oC	15	15.8440	15.8440
40oC	15	17.2807	17.2807
30oC	15	17.2807	17.2807
50oC	15		18.6407
Sig.		.210	.101

Means for groups in homogeneous subsets are displayed.

Based on observed means.

The error term is Mean Square(Error) = 9.196.

a. Uses Harmonic Mean Sample Size = 15.000.

b. Alpha = .05.

Kesimpulan : Menunjukkan bahwa pada suhu 30, 40, dan 70°C tidak memiliki perbedaan yang signifikan karena berada dalam satu subset, sedangkan pada suhu 50 dan 60°C memiliki perbedaan yang signifikan karena tidak berada dalam satu subset yang sama.

Lampiran 16. Hasil Analisis Data Karakterisasi pH dengan SPSS

Uji kolmogorov Smirnov

Tujuan : mengetahui kenormalan data sebagai syarat uji ANOVA

Kriteria uji : Sig. < 0,05 berarti H0 ditolak Sig. > 0,05 H0 diterima

Hasil:

		Aktivitas Amilase pH
N		75
Normal Parameters ^{a,b}	Mean	9.4401
	Std. Deviation	4.71396
	Absolute	.264
Most Extreme Differences	Positive	.137
	Negative	-.264
Kolmogorov-Smirnov Z		2.289
Asymp. Sig. (2-tailed)		.000

a. Test distribution is Normal.

b. Calculated from data.

Kesimpulan : Sig. 0,000 > 0,05 (H0 ditolak) maka data diameter zona bening (aktivitas amilase) tidak terdistribusi normal.

Uji Kruskal Wallis

Tujuan : mengetahui perbedaan data aktivitas amilase berdasarkan variasi pH

Kriteria uji : Sig. < 0,05 berarti H0 ditolak Sig. > 0,05 H0 diterima

Hasil :

		Aktivitas Amilase pH
Chi-Square		34.551
df		4
Asymp. Sig.		.000

a. Kruskal Wallis Test

b. Grouping Variable: pH

Kesimpulan : Menunjukkan nilai sig 0,000 < 0,05 (H0 ditolak) artinya pada hasil data aktivitas amilase pH terdapat perbedaan antara pH 4-8.

Lampiran 17. Alat Penelitian

Inkubator



Oven



Autoclav



Jangka Sorong



Waterbath

Lampiran 18. Cara Pembuatan Media

Sulfide Indol Motility (SIM)

Komposisi :	Pepton from casein	20	gram
	Pepton from meat	6	gram
	Ammonium Iron (II) citrate	0,2	gram
	Sodium thiosulfate	0,2	gram
	Agar-agar	0,2	gram

Cara pembuatan : Dilarutkan media SIM dalam aquadest sebanyak 1 L, dipanaskan sampai larut sempurna, kemudian disterilkan dengan autoklaf pada suhu 121°C selama 15 menit dan dituangkan dalam cawan petri.

Kligler Iron Agar (KIA)

Komposisi :	Pepton from casein	15	gram
	Pepton from meat	5	gram
	Ammonium Iron (II) citrate	0,5	gram
	Meat extract	3	gram
	Yeast extract	3	gram
	Sodium chloride	5	gram
	Laktosa	10	gram
	Glukosa	1	gram
	Sodium thiosulfate	0,5	gram
	Phenol red	0,024	gram
	Agar-agar	12	gram

Cara Pembuatan : Dilarutkan media KIA dalam aquadest sebanyak 1 L, dipanaskan sampai larut sempurna, kemudian disterilkan dengan autoklaf pada suhu 121°C selama 15 menit dan tuangkan dalam cawan petri.

Lysine Indol Agar (LIA)

Komposisi :	Pepton from meat	5	gram
	Yeast extract	3	gram

Glukosa	1	gram
Lysine monohydrochloride	10	gram
Sodium thiosulfate	0,04	gram
Ammonium Iron (II) citrate	0,5	gram
Bromo cresol purple	0,02	gram
Agar-agar	12,5	gram

Cara Pembuatan : Dilarutkan dalam aquadest sebanyak 1 L, dipanaskan sampai larut sempurna, kemudian disterilkan dengan autoklaf pada suhu 121°C selama 15 menit dan tuangkan dalam cawan petri.

Simmon Citrate Agar (SCA)

Komposisi :	Ammonium hydrogen fosfat	1	gram
	DI-Potassium hydrogen fosfat	1	gram
	Sodium chloride	5	gram
	Magnesium sulfate	0,2	gram
	Bromo thymol blue	0,08	gram
	Agar-agar	12,5	gram

Cara Pembuatan: Dilarutkan media SCA dalam aquadest sebanyak 1 L, dipanaskan sampai larut sempurna, kemudian disterilkan dengan autoklaf pada suhu 121°C selama 15 menit dan tuangkan dalam cawan petri.

Nutrient Agar (NA)

Komposisi :	Peptone	5	gram
	Sodium chloride	5	gram
	HM peptone B	1,5	gram
	Yeast extract	1,5	gram
	Agar	15	gram

Cara Pembuatan : Dilarutkan 28 gram media NA dalam 1 L aquadest. Dipanaskan sampai mendidih dan melarut sempurna. Disterilkan dengan autoklaf pada suhu 121°C selama 15 menit, lalu tambahkan 10 ml Kalium Tellurit 1 %. Kemudian dituang ke dalam cawan petri steril.

Endo Agar (EA)

Komposisi :	Peptone	10	gram
	Lactose	10	gram
	Dipotassium phosphate	3,5	gram
	Sodium sulphite	2,5	gram
	Basic fuchsin	0,5	gram
	Agar	15	gram

Cara Pembuatan : Dilarutkan 41,5 gram media EA dalam 1 L aquadest, lalu ditambahkan 1 ml natrium sulfit 10%. Dipanaskan sampai mendidih dan melarut sempurna. Disterilkan dengan autoklaf pada suhu 121°C selama 15 menit, kemudian dituang ke dalam cawan petri steril.

Pseudomonas Selective Agar (PSA)

Komposisi :	Pancreatic digest of gelatin	20	gram
	Magnesium chloride	1,4	gram
	Dipotassium sulphate	10	gram
	Cetrimide	0,3	gram
	Agar	13,6	gram

Cara Pembuatan : Dilarutkan 45,3 gram media PSA dalam 1 L aquadest, lalu tambahkan 10 ml gliserol. Dipanaskan sampai mendidih dan melarut sempurna. Disterilkan dengan autoklaf pada suhu 121°C selama 15 menit, kemudian dituang ke dalam cawan petri steril.

Salmonella-Shigella Agar (SSA)

Komposisi :	Proteose peptone	5	gram
	Lactose	10	gram
	Bile salts mixture	8,5	gram
	Sodium citrate	8,5	gram
	Sodium thiosulphate	8,5	gram
	Ferric citrate	1	gram

Brilliant green	0,0003 gram
Neutral red	0,025 gram
Agar	13,5 gram

Cara Pembuatan : Dilarutkan 60 gram media PSA dalam 1 L aquadest
Dipanaskan sampai mendidih dan melarut sempurna. Disterilkan dengan autoklaf
pada suhu 121°C selama 15 menit, kemudian dituang ke dalam cawan petri steril.

Vogel Johnson Agar (VJA)

Komposisi :	Tryptone	10	gram
	Yeast extract	5	gram
	Mannitol	10	gram
	Dibasic potassium phosphate	5	gram
	Lithium chloride	5	gram
	Glycine	10	gram
	Phenol red	0.025	gram
	Agar	16	gram

Cara Pembuatan : Dilarutkan 61 gram media PSA dalam 1 L aquadest
Dipanaskan sampai mendidih dan melarut sempurna. Disterilkan dengan autoklaf
pada suhu 121°C selama 15 menit, lalu tambahkan 10 ml Kalium Tellurit 1 %.
Kemudian dituang ke dalam cawan petri steril.

Lampiran 19. Cara Pembuatan Larutan Buffer

Buffer Sitrat pH 4

Dilarutkan 0,492 g Sodium Citrate Dihydrate dalam 40 ml aquadest steril, ditambahkan 0,639 g Citric Acid ke dalam larutan dan diaduk ad homogen. Ditambahkan larutan HCl atau NaOH sampai pH yang diinginkan sesuai, kemudian tambahkan aquadest steril ad 50 ml.

Buffer Sitrat pH 5

Dilarutkan 0,741 g Sodium Citrate Dihydrate dalam 40 ml aquadest steril, ditambahkan 0,476 g Citric Acid ke dalam larutan dan diaduk ad homogen. Ditambahkan larutan HCl atau NaOH sampai pH yang diinginkan sesuai, kemudian tambahkan aquadest steril ad 50 ml.

Buffer Sitrat pH 6

Dilarutkan 1,204 g Sodium Citrate Dihydrate dalam 40 ml aquadest steril, ditambahkan 0,174 g Citric Acid ke dalam larutan dan diaduk ad homogen. Ditambahkan larutan HCl atau NaOH sampai pH yang diinginkan sesuai, kemudian tambahkan aquadest steril ad 50 ml.

Buffer Fosfat pH 7

Dilarutkan 0,774 g Sodium phosphate dibasic heptahydrate dalam 40 ml aquadest steril, ditambahkan 0,291 g Sodium phosphate monobasic monohydrate ke dalam larutan dan diaduk ad homogen. Ditambahkan larutan HCl atau NaOH sampai pH yang diinginkan sesuai, kemudian tambahkan aquadest steril ad 50 ml.

Buffer Fosfat pH 8

Dilarutkan 0,814 g Sodium phosphate dibasic heptahydrate dalam 40 ml aquadest steril, ditambahkan 0,044 g Sodium phosphate monobasic monohydrate ke dalam larutan dan diaduk ad homogen. Ditambahkan larutan HCl atau NaOH sampai pH yang diinginkan sesuai, kemudian tambahkan aquadest steril ad 50 ml.

Lampiran 20. Hasil Clustal Omega

MSSIVNSDDDEIVIAIQERRIAESYSPWEEGAYPAKTESDRLRLAARI FADGHEVILGAEVVMRRVG	56
MSKEIDMRKSVYQIYPKSFNDTNGIGDINGIEKLDYIKLLGYDIWLT FVYESPMNDGDI SNYL	0
MKLLPFLMLLSPSALASWTLPGFPFADTAAGLFTSQALIPGQPLRILYQNHCHQPAEAIKLNQTL	37
MKLAACETLILPGFAVAASWTSFGFPFSEQGTGVFVSHAQLPGRTRPILNFDQQCQWQPADAIKLNQML	36
MKLAAPALILIPGIALASSWTSFGFPFLSTQETGRFTSHAALITGTRALTIHIDQQCQWQPSGAIKLNQML	36
MSSIVNSDDDEIVIAIQERRIAESYSPWEEGAYPAKTESDRLRLAARI FADGHEVILGAEVVMRRVG	107
MSKEIDMRKSVYQIYPKSFNDTNGIGDINGIEKLDYIKLLGYDIWLT FVYESPMNDGDI SNYL	0
MKLLPFLMLLSPSALASWTLPGFPFADTAAGLFTSQALIPGQPLRILYQNHCHQPAEAIKLNQTL	97
MKLAACETLILPGFAVAASWTSFGFPFSEQGTGVFVSHAQLPGRTRPILNFDQQCQWQPADAIKLNQML	96
MKLAAPALILIPGIALASSWTSFGFPFLSTQETGRFTSHAALITGTRALTIHIDQQCQWQPSGAIKLNQML	95
MSSIVNSDDDEIVIAIQERRIAESYSPWEEGAYPAKTESDRLRLAARI FADGHEVILGAEVVMRRVG	163
MSKEIDMRKSVYQIYPKSFNDTNGIGDINGIEKLDYIKLLGYDIWLT FVYESPMNDGDI SNYL	0
MKLLPFLMLLSPSALASWTLPGFPFADTAAGLFTSQALIPGQPLRILYQNHCHQPAEAIKLNQTL	149
MKLAACETLILPGFAVAASWTSFGFPFSEQGTGVFVSHAQLPGRTRPILNFDQQCQWQPADAIKLNQML	148
MKLAAPALILIPGIALASSWTSFGFPFLSTQETGRFTSHAALITGTRALTIHIDQQCQWQPSGAIKLNQML	147
MSSIVNSDDDEIVIAIQERRIAESYSPWEEGAYPAKTESDRLRLAARI FADGHEVILGAEVVMRRVG	208
MSKEIDMRKSVYQIYPKSFNDTNGIGDINGIEKLDYIKLLGYDIWLT FVYESPMNDGDI SNYL	0
MKLLPFLMLLSPSALASWTLPGFPFADTAAGLFTSQALIPGQPLRILYQNHCHQPAEAIKLNQTL	202
MKLAACETLILPGFAVAASWTSFGFPFSEQGTGVFVSHAQLPGRTRPILNFDQQCQWQPADAIKLNQML	201
MKLAAPALILIPGIALASSWTSFGFPFLSTQETGRFTSHAALITGTRALTIHIDQQCQWQPSGAIKLNQML	200
MSSIVNSDDDEIVIAIQERRIAESYSPWEEGAYPAKTESDRLRLAARI FADGHEVILGAEVVMRRVG	262
MSKEIDMRKSVYQIYPKSFNDTNGIGDINGIEKLDYIKLLGYDIWLT FVYESPMNDGDI SNYL	50
MKLLPFLMLLSPSALASWTLPGFPFADTAAGLFTSQALIPGQPLRILYQNHCHQPAEAIKLNQTL	257
MKLAACETLILPGFAVAASWTSFGFPFSEQGTGVFVSHAQLPGRTRPILNFDQQCQWQPADAIKLNQML	256
MKLAAPALILIPGIALASSWTSFGFPFLSTQETGRFTSHAALITGTRALTIHIDQQCQWQPSGAIKLNQML	255

ETAERRELLPEGNDFWSAQITPPCGRLYFRLEAWID- REAGRRRELRVHGAAR --- LP	163
-----	0
----- SLHPCGALPP --- YSNRQFRAGYQVYVDRSGETIQLS	149
----- SLQPCSNTP --- QRLLFRDGEVTLQIDRSGETILMIS	148
----- SLICEGAPP --- QRLLFRDGVYITIVDRSGETIILLS	147
LDLELR EGDELL QRC AERGG FIA AACA- PLAE -- RLQ ----- ACQSV EE R VALW LAAQ	163
-----	0
LSSV ESAA VAV SC FR W CG EV TV DV VT ST TE GE L RF S GE TA Q YS Q W K TI L Q P APA	149
IQNA AE FVA SL Y AE CG AW D GL EL TV DV SA IF E GA AV RV YS QQ IA IV NG Q IM LQ P AAI	148
IKTE PER TI Q A Y Q CF V W D S EL IV DR Q TE EG TV RV YS Q I DI Y Q W K TI LL PA - D	147
TG ELL LV GER EL Y R SK EY EV ER EL AR FA SN EL FF SE SG DT H GT DD VI	163
-----	0
SG GL LL ES AA TA----- K PA AF SW NA TV F VL DR EN GN PA ND HS YGR SD GLQ	149
SN GL LL ER AE TD----- TS AP FW NA TV F VL DR EN GF SD SN DQ SY GR H D GM A	148
SH GL LL ER AE TH----- AS AP FW NA TV F VL DR EN GF TD HS YGR H D GM A	147
----- ER LP Q FA AV GE D VL Y FP P - I HP IG TR K GR NS LA E AG D GS PY	208
-----	0
E IG TH GG LA GL Q L D Y L Q Q L G W N AL W ISS PE Q I H G W Y GG T ----- Q GF F HY A	202
E IG TH GG LR GL W L D Y L Q Q L G W N AL W IS AP FE Q I H G W Y GG T----- K GF F HY A	201
E IG TH GG LR GL T S Q L D Y L Q Q L G W N AL W ISS PE Q I H G W Y GG T ----- K GF F HY A	200
AI SG EE GG EA HP EL GB RE DR EL IV AV RE H GV E L AL D FA IQ CS - P H EW LE H -----	262
----- E IN ED FE MD FE KL IV A H Q DL K W L DI V IN HT ST HE W TE AR SK D	50
YH GY AL W RL D AM NG TE Q EL IV E Q A HR GR IL IF V W N H GV Y AT LA ----- DM Q I	257
YH GY Y Q W N L D AM NG E AD RL IV DS A H Q GR IL IF V W N H GV Y AT LA ----- DM Q E	256
YH GY Y Q W T L D AM NG S E AD RL IV D GA H Q GR IL IF V W N H GV Y AT LA----- DM Q E	255

500LWVWXYZ[]^_`~!@#%&'()*+,-./:;<=>?@AB
 501CDEFGHIJKLMNOPQRSTUVWXYZ[]^_`~!@#%&'()*+,-./:;<=>?@AB
 502CDEFGHIJKLMNOPQRSTUVWXYZ[]^_`~!@#%&'()*+,-./:;<=>?@AB

536QWERTYUIOPASDFGHJKLZXCVBNM[]^_`~!@#%&'()*+,-./:;<=>?@AB
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 539CDEFGHIJKLMNOPQRSTUVWXYZ[]^_`~!@#%&'()*+,-./:;<=>?@AB
 540CDEFGHIJKLMNOPQRSTUVWXYZ[]^_`~!@#%&'()*+,-./:;<=>?@AB

569QWERTYUIOPASDFGHJKLZXCVBNM[]^_`~!@#%&'()*+,-./:;<=>?@AB
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581QWERTYUIOPASDFGHJKLZXCVBNM[]^_`~!@#%&'()*+,-./:;<=>?@AB
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594QWERTYUIOPASDFGHJKLZXCVBNM[]^_`~!@#%&'()*+,-./:;<=>?@AB
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Lampiran 21. Phylogenetic Tree

Results for job clustalo-20190617-033301-0456-55119104-p1.m

Alignments Result Summary **Phylogenetic Tree** Submission Details

Phylogenetic Tree

This is a Neighbour-joining tree without distance corrections.

Download Phylogenetic Tree Data

Branch length: Cladogram Heat

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MSVIRNSDDPLVAGQPRVIESYSPVEEGAPKTESDRLAARIFADGHEVLGAEVMIRRVG 0.41
MSKEDNRKSWYQPKSFNDITGNGDNGHEKLDYKLLGVDMMLTPVYESPMNDNGYDINYL 0.392
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```