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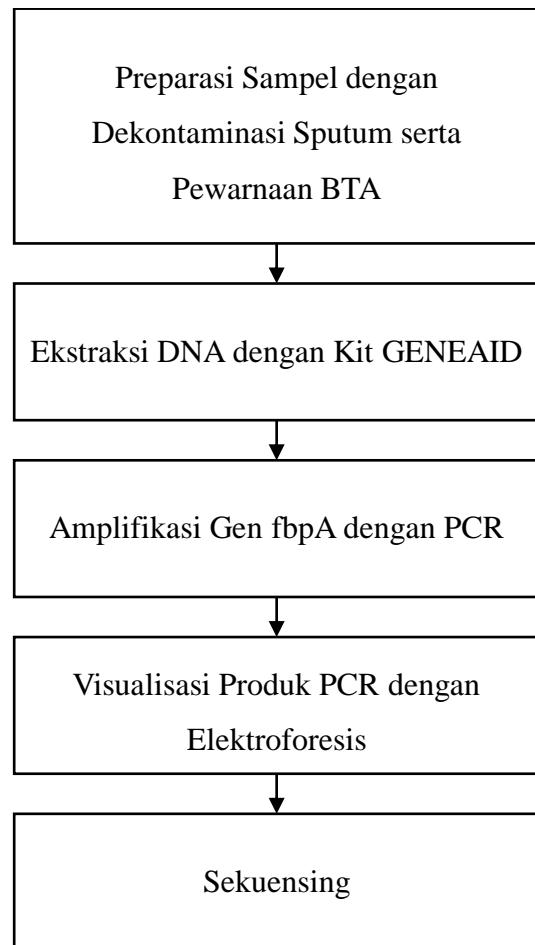
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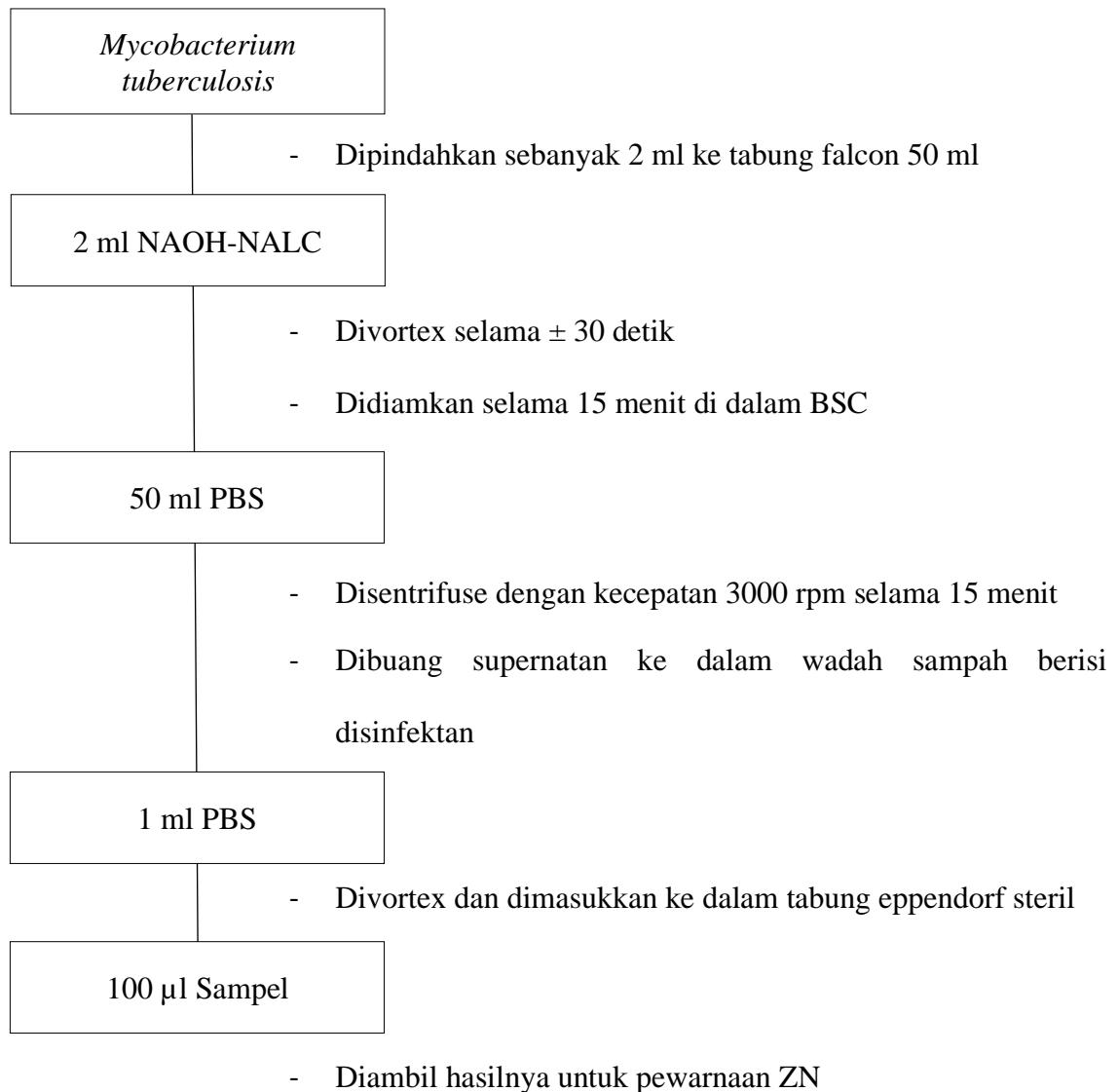
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## LAMPIRAN

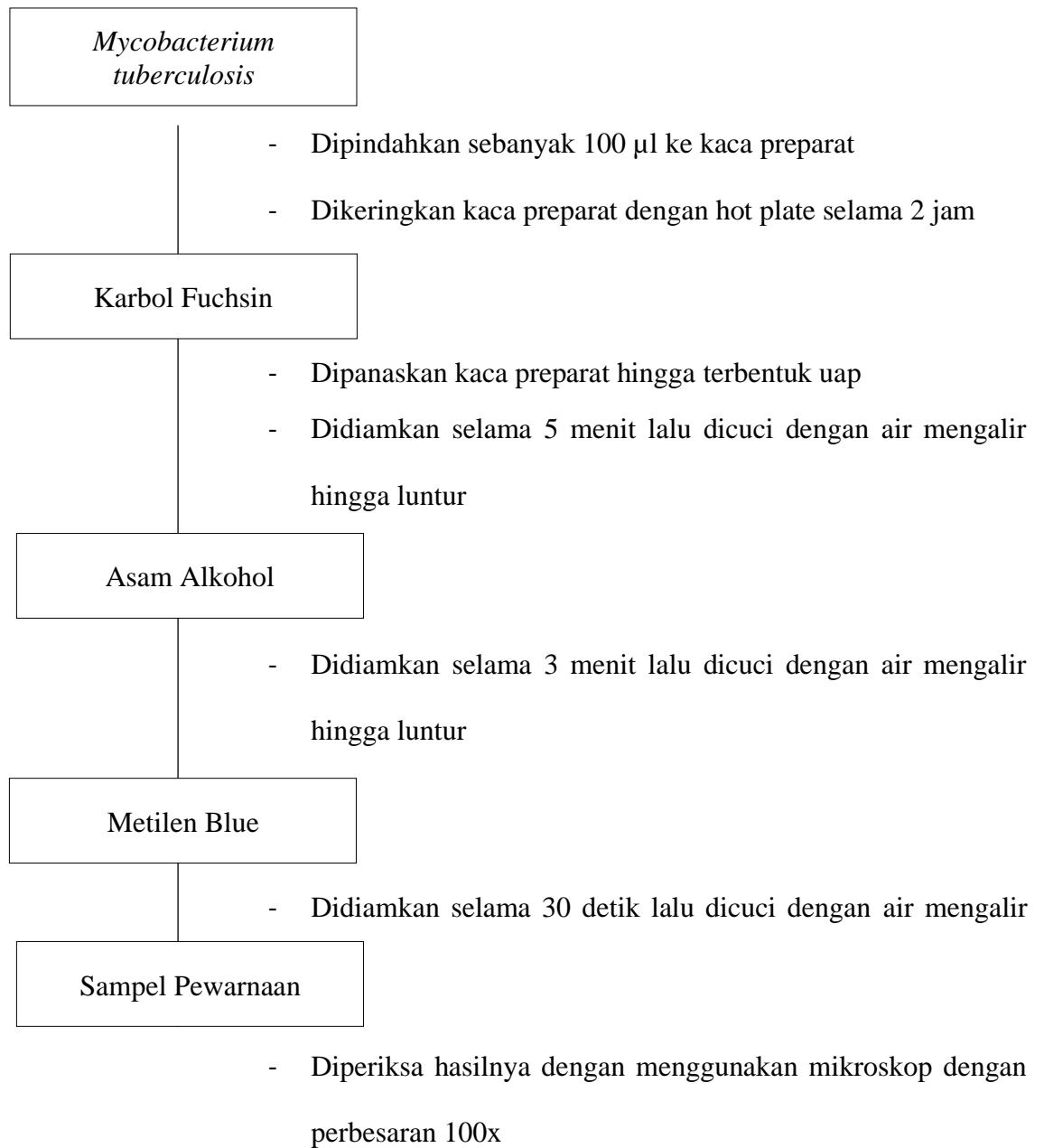
### Lampiran 1. Skema Kerja



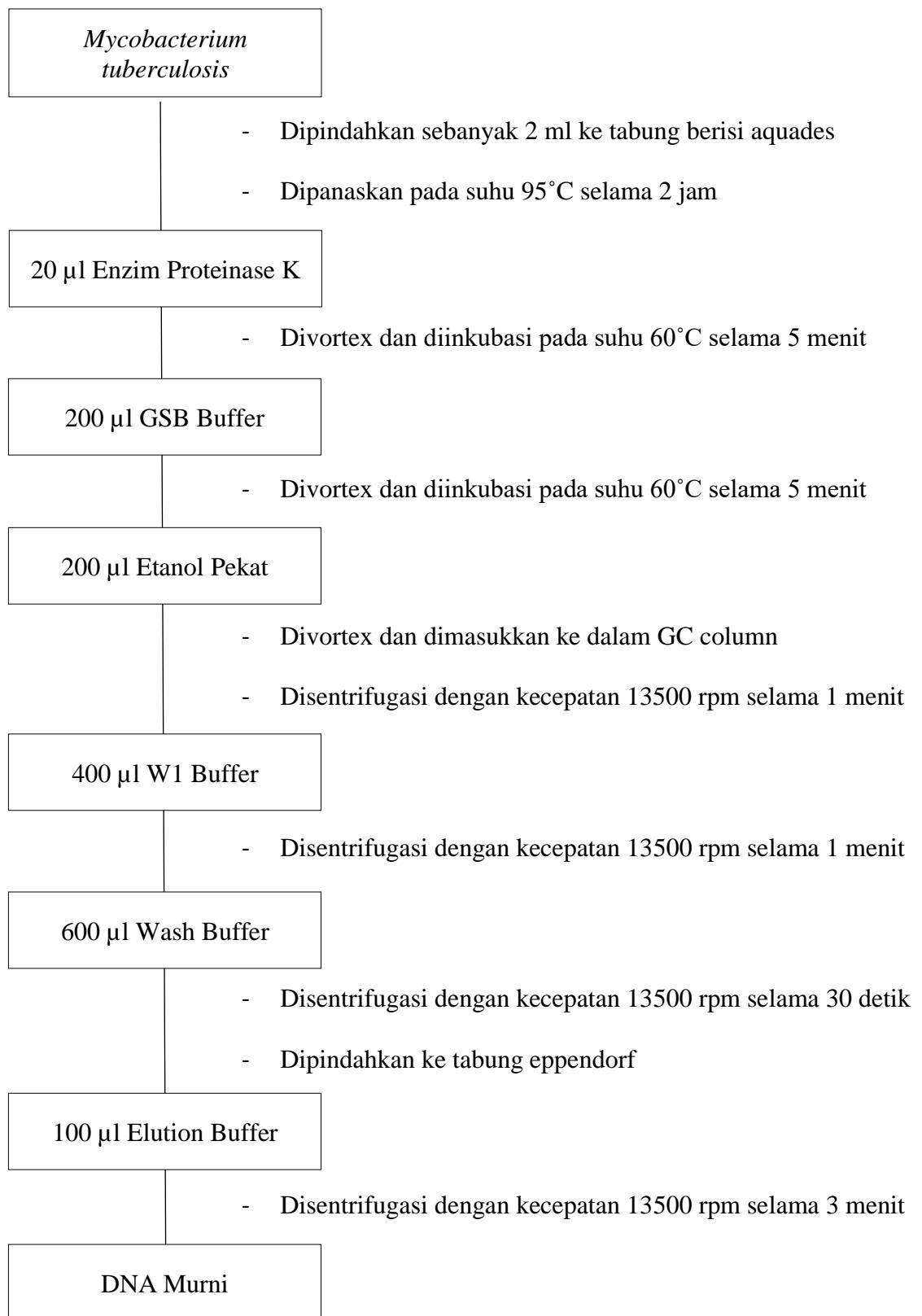
## A. Dekontaminasi Sampel Sputum



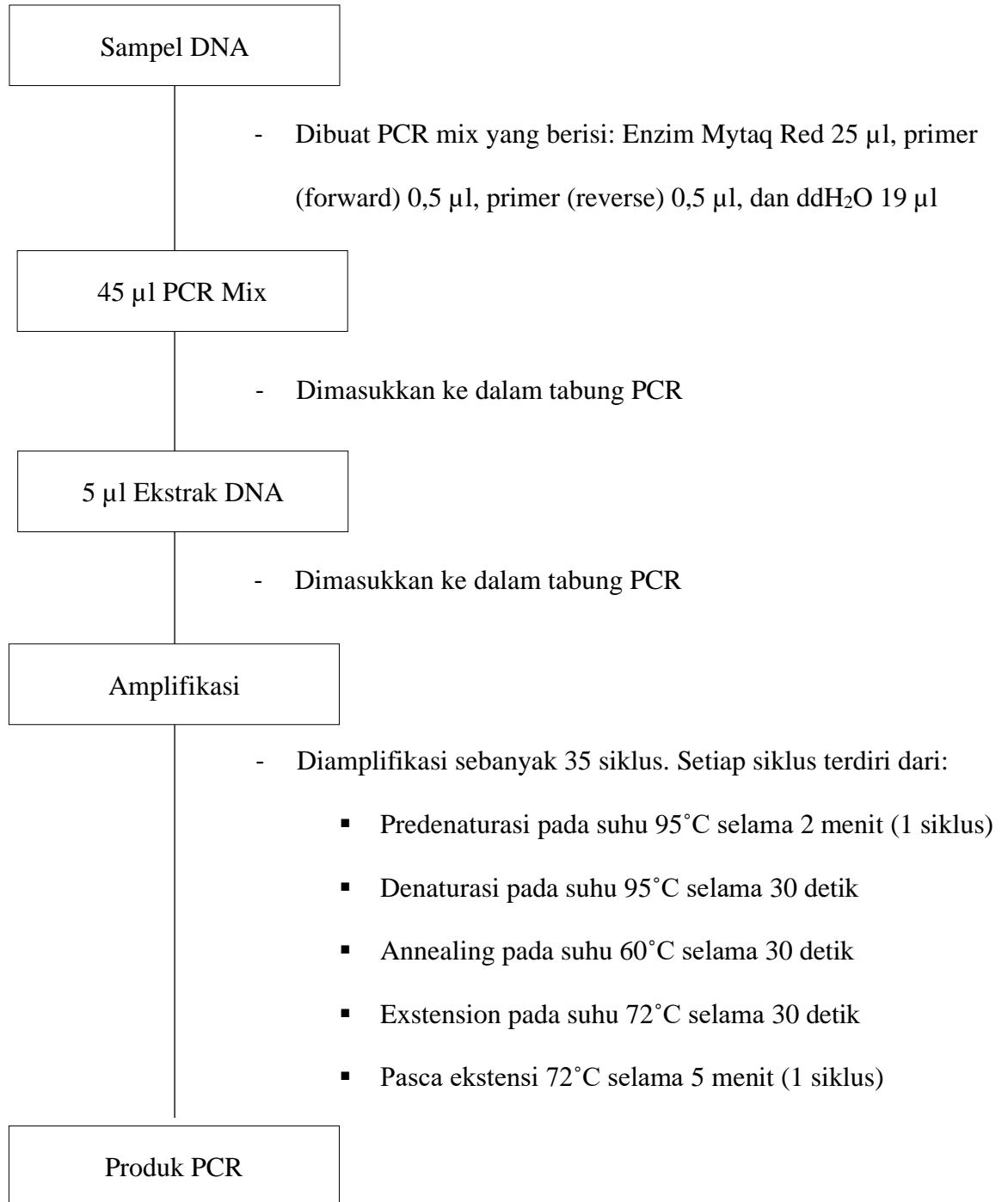
## B. Pewarnaan Ziehl-Neelson



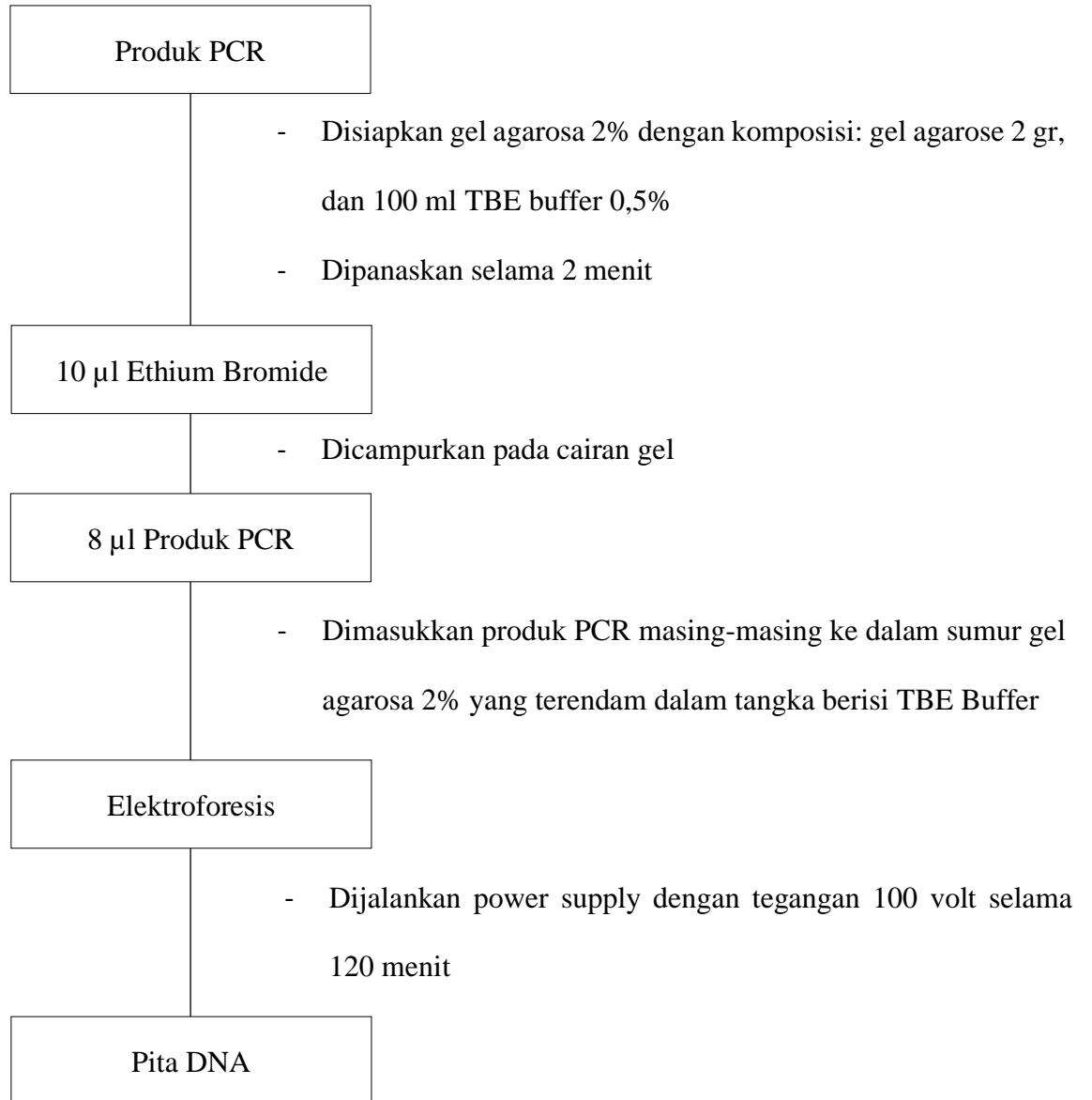
### C. Ekstraksi DNA *Mycobacterium tuberculosis*



#### D. Amplifikasi DNA dengan PCR



## E. Deteksi Produk PCR dengan Elektroforesis



**Lampiran 2.** Peta Origin Antara Primer dan Gen fbpA dari DataBase

**A. Origin Gen fbpA dari DataBase**

**Mycobacterium tuberculosis H37Rv, complete genome**

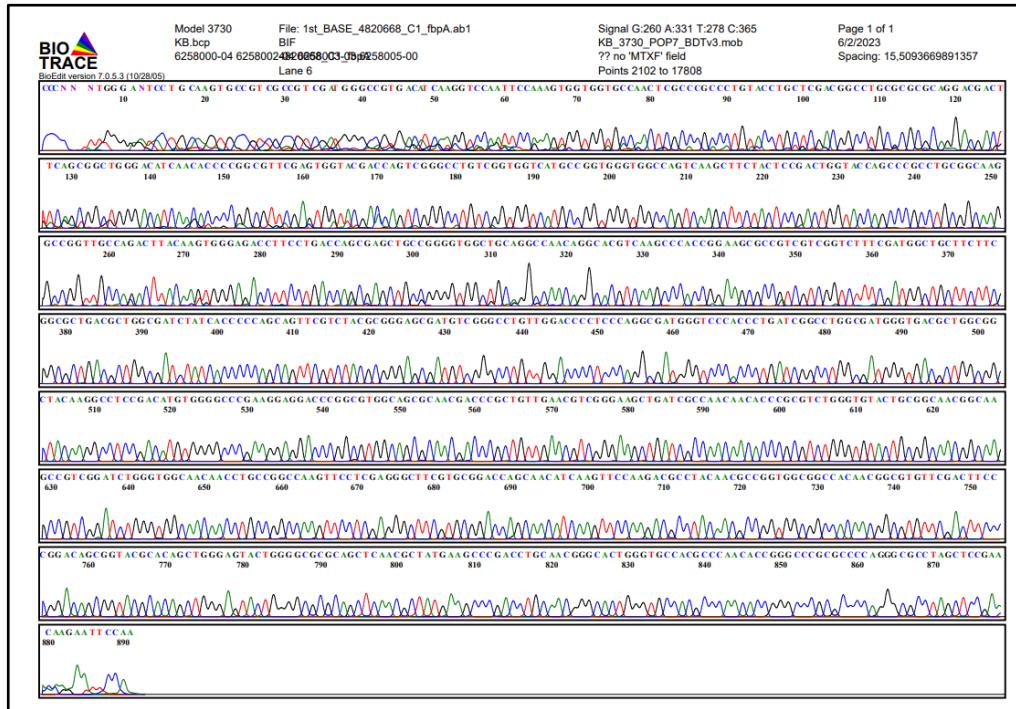
GenBank: CP003248.2

[GenBank](#) [Graphics](#)

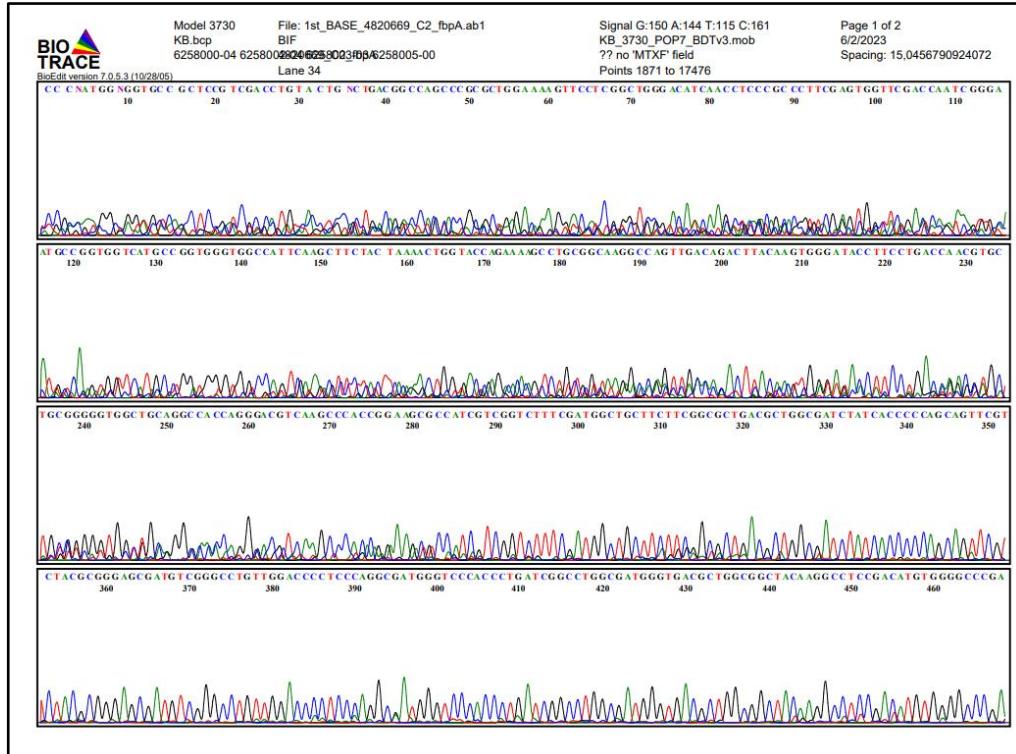
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>CP003248.2:4265809-4266707 Mycobacterium tuberculosis H37Rv, complete genome
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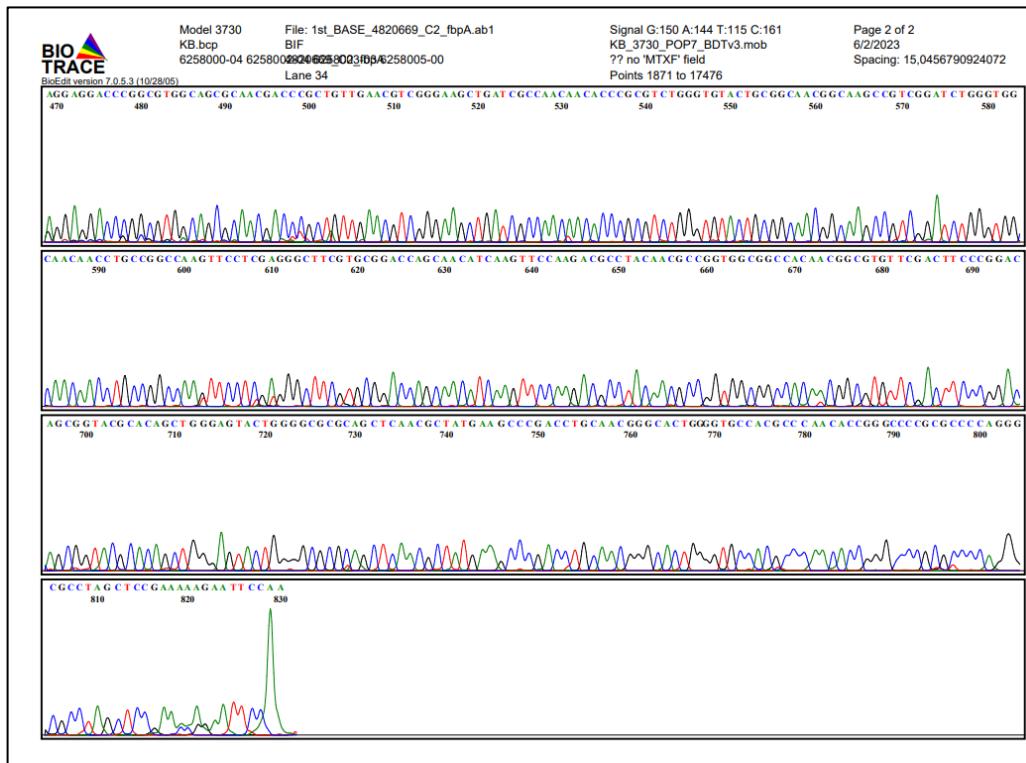
### Lampiran 3. Hasil Sekuens Sampel

#### A. Visualisasi Sekuens Sampel C1

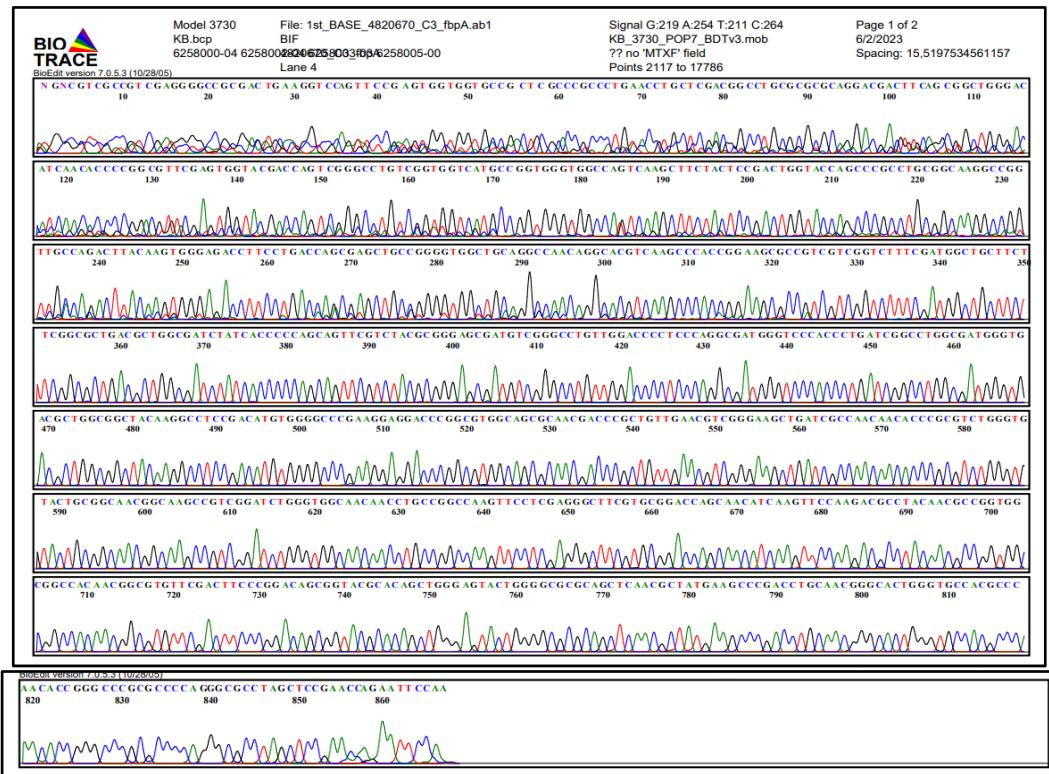


#### B. Visualisasi Sekuens Sampel C2

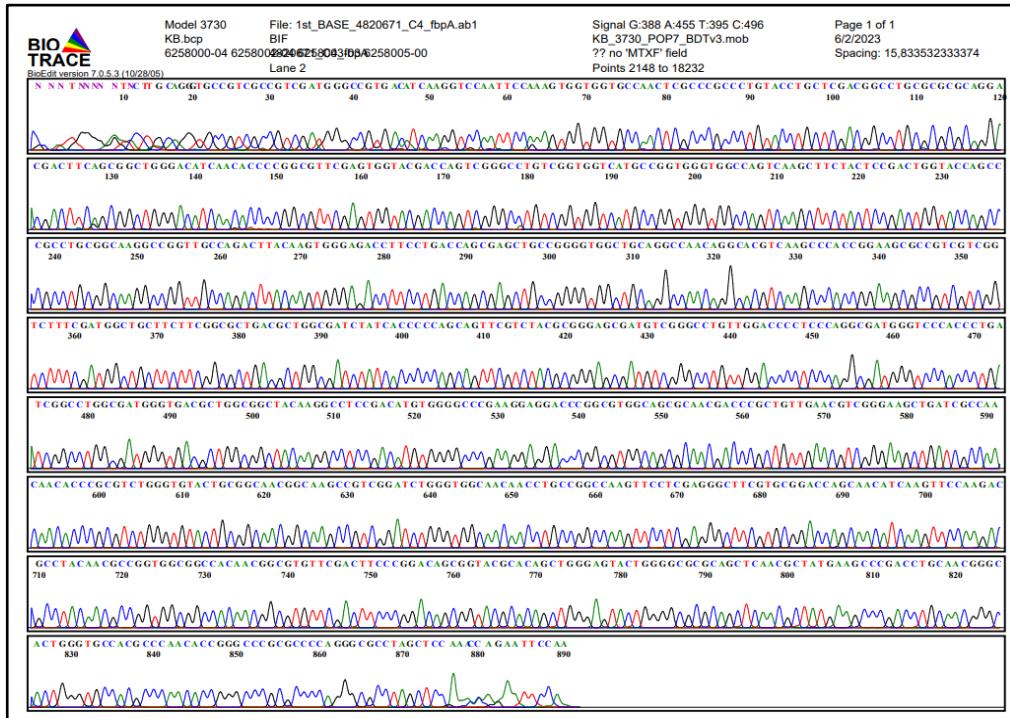




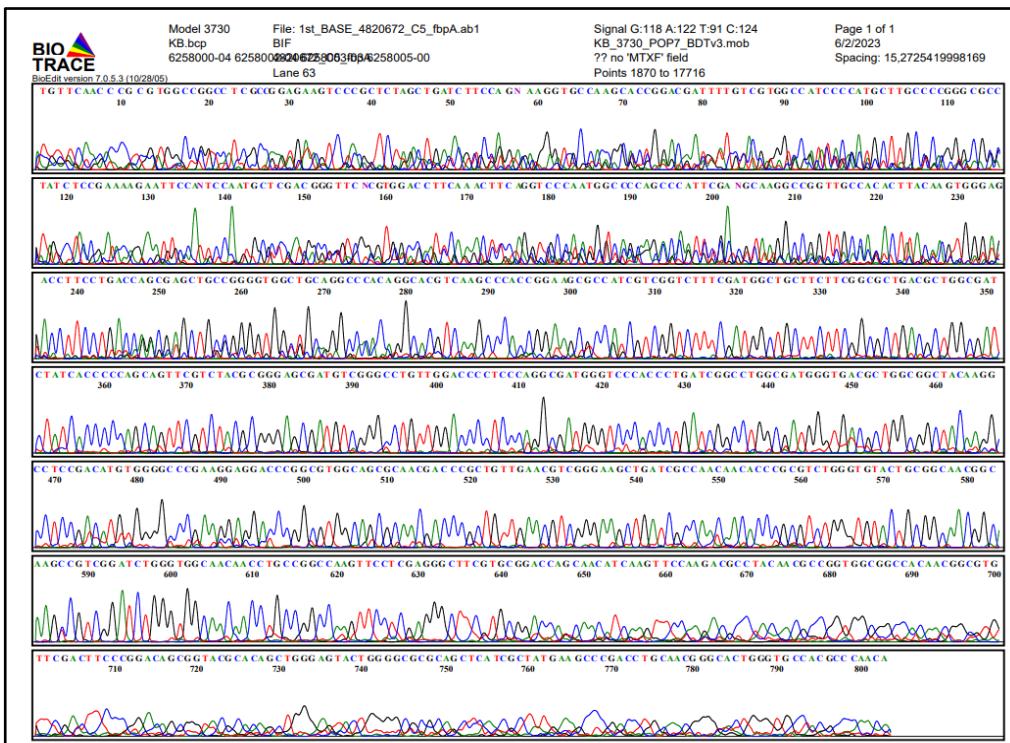
### C. Visualisasi Sekuensi Sampel C3



## D. Visualisasi Sekuens Sampel C4



## E. Visualisasi Sekuens Sampel C5



## Lampiran 4. Hasil BLAST Sampel

### A. Hasil BLAST Sampel C1

**Mycobacterium tuberculosis H37Rv, complete genome**

Sequence ID: [CP003248.2](#) Length: 4411709 Number of Matches: 1

Range 1: 4265811 to 4266678 <a href="#">GenBank</a> <a href="#">Graphics</a>					<a href="#">▼ Next Match</a>	<a href="#">▲ Previous Match</a>
Score 1598 bits(865)	Expect 0.0	Identities 867/868(99%)	Gaps 0/868(0%)	Strand Plus/Minus		
Query 14	CCTGCAAGTGCCGTGCCGTGATGGCCGTGACATCAAGGTCAAATTCCAAAGTGGTGG	73				
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Sbjct 4266618	TGCCAACTCGCCGCCCTGTACCTGCTCGACGCCCTGCGCGCAGGACGACTTCAGCGG	4266559				
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Sbjct 4266138	CAACGACCCGCTGTTAACGTCGGGAAGCTGATGCCAACACACCCCGCTGGGTGA	4266079				
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## B. Hasil BLAST Sampel C2

### Mycobacterium tuberculosis H37Rv, complete genome

Sequence ID: [CP003248.2](#) Length: 4411709 Number of Matches: 1

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Query 26	CCTGTA-CTGNCT-GACGGCCAGCCC CGC CTGGAAAAGTT CCTCGGCTGGGACATCAACC			83		
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Sbjct 4266365	GTCAAGCCCACCGGAAGGCCGTGTCGGTCTTCGATGGCTGCTTCTCGGCCTGACG				4266306	
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Query 444	AAGGCCTCCGACATGTGGGCCCCAAGGAGGACCCGGCTGGCAGCGCAACGACCCGCTG			503		
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Sbjct 4265827	GCGCCTAGCTCGAA 4265813					

### C. Hasil BLAST Sampel C3

#### Mycobacterium tuberculosis H37Rv, complete genome

Sequence ID: [CP003248.2](#) Length: 4411709 Number of Matches: 1

Range 1: 4265809 to 4266667 <a href="#">GenBank</a> <a href="#">Graphics</a>					<a href="#">▼ Next Match</a>	<a href="#">▲ Previous Match</a>
Score 1524 bits(825)	Expect 0.0	Identities 849/860(99%)	Gaps 4/860(0%)	Strand Plus/Minus		
Query 4	CGTCGCCGTCGAGGGGCCGCGAC	-TGAAGTCCAGTTCC-GAGTGGTGGTGCC-GCTCGC	60			
Sbjct 4266667						4266608
Query 61	CCGCCCTGAACCTGTCGACGGCCTGCGCGCGCAGGACGACTTCAGCGGCTGGGACATCA	120				
Sbjct 4266607						4266548
Query 121	ACACCCGGCGTTCGAGTGGTACGACCGAGTCGGGCCTGTCGGTGGTATGCCGGTGGTG	180				
Sbjct 4266547						4266488
Query 181	GCCAGTCAAGCTTCACTCCGACTGGTACCGAGCCCCCTGCGGCAAGGCCGGTTGCCAGA	240				
Sbjct 4266487						4266428
Query 241	CTTACAAGTGGGAGACCTTCTGACAGCGAGCTGCCGGGTGGCTGCAGGCCAACAGGC	300				
Sbjct 4266427						4266368
Query 301	ACGTCAAGCCCACCGGAAGCGCCGTCGTCGGTCTTCGATGGCTGCTTCTGGCGCTGA	360				
Sbjct 4266367						4266308
Query 361	CGCTGGCGATCTATCACCCCCAGCAGCTTCGTCACCGGGAGCAGTCGGGCCTGTTGG	420				
Sbjct 4266307						4266248
Query 421	ACCCCTCCCAGGCATGGGTCCCACCCCTGATCGGCCCTGGCGATGGGTGACGCTGGCGCT	480				
Sbjct 4266247						4266188
Query 481	ACAAGGCCTCCGACATGTGGGGCCCGAAGGAGGACCCGGCTGGCAGCGCAACGACCCGC	540				
Sbjct 4266187						4266128
Query 541	TGTTGAACGTCGGGAAGCTGATGCCAACAAACCCCGCTGGGTGTAUTGCGGCAACG	600				
Sbjct 4266127						4266068
Query 601	GCAAGCCGTCGGATGGGTGGCAACAAACCTGCCGCAAGTCTCGAGGGCTCGTGC	660				
Sbjct 4266067						4266008
Query 661	GGACCAGCAACATCAAGTCCAAGACGCCCTAACACGCCGGTGGCGGCCAACACGGCGTGT	720				
Sbjct 4266007						4265948
Query 721	TCGACTTCCCGGACAGCGGTACGCACAGCTGGGAGTACTGGGGCGCGCAGCTAACGCTA	780				
Sbjct 4265947						4265888
Query 781	TGAAGCCGACCTGCAACGGGCACTGGGTGCCACGCCAACACCGGGCCCGCCCCAGG	840				
Sbjct 4265887						4265828
Query 841	GCGCCTAGCTCCGAACCAGA	860				
Sbjct 4265827						4265809

## D. Hasil BLAST Sampel C4

### Mycobacterium tuberculosis H37Rv, complete genome

Sequence ID: [CP003248.2](#) Length: 4411709 Number of Matches: 1

Range 1: 4265809 to 4266676 <a href="#">GenBank</a> <a href="#">Graphics</a>					<a href="#">▼ Next Match</a>	<a href="#">▲ Previous Match</a>
Score 1589 bits(860)	Expect 0.0	Identities 867/870(99%)	Gaps 2/870(0%)	Strand Plus/Minus		
Query 14	TGCAGGGTGCGCTGCCGTCGATGGCCGTGACATCAAGGTCAAATTCCAAAGTGGTGGT	73				
Sbjct 4266676		4266618				
Query 74	GCCAACCTGCCGCCCTGTACCTGCTGACGGCCTGCGCGCGCAGGACGACTTCAGCGGC	133				
Sbjct 4266617		4266558				
Query 134	TGGGACATCAACACCCCGGCGTTCGAGTGGTACGACCAGTCGGGCTGCGGTGGTCATG	193				
Sbjct 4266557		4266498				
Query 194	CCGGTGGGTGGCCAGTCAAGCTCTACTCCGACTGGTACCGAGCCCGCTGCGGCAAGGCC	253				
Sbjct 4266497		4266438				
Query 254	GGTTGCCAGACTTACAAGTGGAGACCTTCTGACCAAGCGAGCTGCCGGGGTGGCTGCAG	313				
Sbjct 4266437		4266378				
Query 314	GCCAACAGGCACGTCAGGCCACCGGAAGGCCGTGCGTGGTCTTCGATGGCTGTTCT	373				
Sbjct 4266377		4266318				
Query 374	TCGGCGCTGACGCTGGCGATCTATCACCCCCAGCAGTCGCTACCGGGAGCGATGTCG	433				
Sbjct 4266317		4266258				
Query 434	GGCCTGTTGGACCCCTCCCAGCGATGGGTCCCACCCCTGATCGGCTGGCGATGGGTGAC	493				
Sbjct 4266257		4266198				
Query 494	GCTGGCGCTACAAGGCCTCGACATGTGGGCCGAAGGAGGACCCGGCTGGCAGCGC	553				
Sbjct 4266197		4266138				
Query 554	AACGACCCGCTGTTGAACGTCGGAAGCTGATCGCCAACAACACCCCGCTGGGTGAC	613				
Sbjct 4266137		4266078				
Query 614	TGCAGGGCAACGGCAAGCCGTCGGATCTGGGTGGCAACAACCTGCCGCAAGTCTCGAG	673				
Sbjct 4266077		4266018				
Query 674	GGCTTCGTGCGGACAGCAACATCAAGTCCAAGACGCGCTACAACGCCGGTGGCGCAC	733				
Sbjct 4266017		4265958				
Query 734	AACGGCGTGTGACTTCCCGGACAGCGGTACGCACAGCAGCTGGGAGTACTGGGCGCGAG	793				
Sbjct 4265957		4265898				
Query 794	CTCAACGCTATGAAGCCCGACCTGCAACGGGACTGGGTGCCACGCCAACACCGGGCCC	853				
Sbjct 4265897		4265838				
Query 854	GCGCCCCAGGGCGCTAGCTCAAACCAGA 883					
Sbjct 4265837	GCGCCCCAGGGCGCTAGCTCGAAC-AGA 4265809					

## E. Hasil BLAST Sampel C5

### Mycobacterium tuberculosis H37Rv, complete genome

Sequence ID: [CP003248.2](#) Length: 4411709 Number of Matches: 1

Range 1: 4265846 to 4266445 <a href="#">GenBank</a> <a href="#">Graphics</a>					<a href="#">▼ Next Match</a>	<a href="#">▲ Previous Match</a>
Score 1086 bits(588)	Expect 0.0	Identities 596/600(99%)	Gaps 0/600(0%)	Strand Plus/Minus		
Query 204	GCAAGGCCGGTTGCCACACTTACAAGTGGGAGACCTTCCTGACCAGCGAGCTGCCGGGT	263				
Sbjct 4266445	GCAAGGCCGGTTGCCACACTTACAAGTGGGAGACCTTCCTGACCAGCGAGCTGCCGGGT					4266386
Query 264	GGCTGCAGGCCACAGGCACGTCAAGCCCACCGGAAGCGCCATGTCGGTCTTCGATGG	323				
Sbjct 4266385	GGCTGCAGGCCACAGGCACGTCAAGCCCACCGGAAGCGCCGTGTCGGTCTTCGATGG					4266326
Query 324	CTGCTTCTCGCGCCTGACGCTGGCGATCTATCACCCCCAGCAGTTCGTCTACGCGGGAG	383				
Sbjct 4266325	CTGCTTCTCGCGCCTGACGCTGGCGATCTATCACCCCCAGCAGTTCGTCTACGCGGGAG					4266266
Query 384	CGATGTCGGGCCTGTTGACCCCTCCAGGCGATGGTCCCACCCCTGATGGCCTGGCGA	443				
Sbjct 4266265	CGATGTCGGGCCTGTTGACCCCTCCAGGCGATGGTCCCACCCCTGATGGCCTGGCGA					4266206
Query 444	TGGGTGACGCTGGCGCTACAAGGCCTCCGACATGTGGGGCCGAAGGAGGACCCGGCGT	503				
Sbjct 4266205	TGGGTGACGCTGGCGCTACAAGGCCTCCGACATGTGGGGCCGAAGGAGGACCCGGCGT					4266146
Query 504	GGCAGCGAACGACCCGCTGTTGAACTCGGGAGCTGATGCCAACACACCCGGCT	563				
Sbjct 4266145	GGCAGCGAACGACCCGCTGTTGAACTCGGGAGCTGATGCCAACACACCCGGCT					4266086
Query 564	GGGTGTACTGCGCAACGGCAAGCCGCTGGATCTGGGTGGCAACAACCTGCCGGCAAGT	623				
Sbjct 4266085	GGGTGTACTGCGCAACGGCAAGCCGCTGGATCTGGGTGGCAACAACCTGCCGGCAAGT					4266026
Query 624	TCCTCGAGGGCTTCGTGCGGACCAGAACATCAAGTCCAAGACGCCATAACGCCGGTG	683				
Sbjct 4266025	TCCTCGAGGGCTTCGTGCGGACCAGAACATCAAGTCCAAGACGCCATAACGCCGGTG					4265966
Query 684	GCGGCCACAACGGCGTGGTCACTTCCGGACAGCGGTACGCACAGCTGGAGTACTGG	743				
Sbjct 4265965	GCGGCCACAACGGCGTGGTCACTTCCGGACAGCGGTACGCACAGCTGGAGTACTGG					4265906
Query 744	GCGCGCAGCTCATCGCTATGAAGCCGACCTGCAACGGCACTGGGTGCCACGCCAAC	803				
Sbjct 4265905	GCGCGCAGCTCATCGCTATGAAGCCGACCTGCAACGGCACTGGGTGCCACGCCAAC					4265846

## **Lampiran 5. Komposisi Bahan**

### **A. Komposisi Larutan Stok Dekontaminasi**

#### **Stok larutan NaOH-Sodium Sitrat:**

NaOH 1 N : 20 gram, 500 ml akuades

Sodium Sitrat : 14,5 gram, 500 ml akuades

#### **Stok larutan NaOH-NALC:**

NaOH-Sodium sitrat : 100 ml

Massa NALC : 1 gram

#### **Larutan stok PBS *buffer*:**

Na<sub>2</sub>HPO<sub>4</sub> : 300 ml (0,947%)

KH<sub>2</sub>PO<sub>4</sub> : 200 ml (0,947%)

**PBS *buffer* : 500 ml (0,067 M, pH 6,8)**

### **B. Komposisi Reagen Ekstraksi DNA**

Sampel sputum : 200µl

Enzim Proteinase K : 20µl

GSB *buffer* : 200µl

Etanol absolut : 200µl

W1 *buffer* : 400µl

Wash *buffer* : 600µl

Elusion *buffer* : 100µl

### **C. Komposisi Larutan Stok Primer**

Massa primer kering : 0,23 mg (26,1 nmol)

Volume ddH<sub>2</sub>O : 261 µl

**Konsentrasi stok : 100 µl (261 µl)**

Pengenceran larutan stok:

$$V_1 N_1 = V_1 N_1$$

$$V_1 \cdot 100 = 100 \mu\text{l} \cdot 20$$

$$V_1 = 20 \mu\text{l} \rightarrow \text{Volume ddH}_2\text{O} = 80 \mu\text{l}$$

#### **D. Komposisi PCR Mix**

Enzim *mytaq red* : 25 µl

Primer *forward* : 0,5 µl

Primer *reverse* : 0,5 µl

ddH<sub>2</sub>O : 19 µl

Sampel DNA : 5 µl

**Volume Total : 25 µl**

#### **E. Komposisi Gel Agarosa**

Berat agarosa : 2 gram

Volume TBE *buffer* : 100 ml

## Lampiran 6. Bahan yang Digunakan

### A. Pewarnaan Ziehl-Neelson



### B. Ekstraksi DNA



**Lampiran 7. Dokumentasi Penelitian**

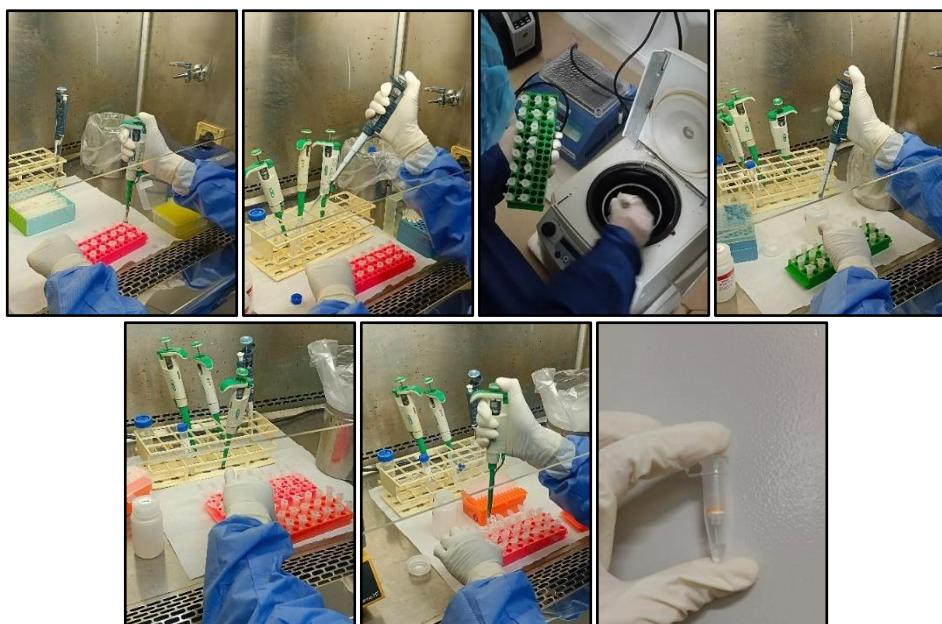
**A. Dekontaminasi Sputum**



**B. Pewarnaan Ziehl-Neelsens**



### C. Ekstraksi DNA



### D. Amplifikasi Gen fbpA dengan PCR



### E. Elektroforesis

