

HiPer[®] Ezy MIC[®] Teaching Kit

Product Code: HTM006E

Number of experiments that can be performed: 10

Duration of Experiment:

Day 1: Preparation of media and revival of strains

Day 2: Protocol

Day 3: Observations and Results

Storage Instructions:

- The kit is stable for 12 months from the date of manufacture
 - Store the EzyMIC[®] strips at 2-8°C
- Other kit contents can be stored at room temperature (15-25°C)

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the strip to the pre-inoculated agar, the antibiotic releases from the strip forming a defined concentration gradient in the area around the strip. After appropriate incubation of the plates, a zone of inhibition is formed on either side of Ezy MIC® strip. The MIC is easily read as the value printed on the strip where the growth of the organism touches the strip. If the growth intersects on the line between two dilutions then the MIC is read as the value in the lower one. Ezy MIC® Teaching Kit contains Ezy MIC® strips which allow for the rapid and accurate determination of the MIC of an organism to an antibiotic for the improvement of patient management.

Kit Contents:

This kit can be used to determine the sensitivity of bacteria against various antibiotics.

Table 1: Enlists the materials provided in this kit with their quantity and recommended storage

Sr. No.	Product Code	Materials Provided	Quantity	Storage
			10 Expts	
1.		Ezy MIC® Strips		
a.	EM001	Amikacin	10*	2-8°C
b.	EM004	Azithromycin	10*	2-8°C
c.	EM017	Ciprofloxacin	10*	2-8°C
d.	EM029	Linezolid	10*	2-8°C
e.	EM060	Vancomycin	10*	2-8°C
3.	PW005	Sterile cotton swabs	55 Nos.	RT
4.	MV173	Mueller Hinton Agar	65 g	RT
5.	TKC376	Sterile Saline Solution	11 ml	2-8°C
6.	TKC375	Applicator sticks	10 Nos.	RT

* Ezy MIC® Strips are provided in separate packs. Each pack contains 10 strips of the corresponding antibiotic.

Materials Required But Not Provided:

Culture: *Staphylococcus aureus*

Glasswares: Sterile test tubes, sterile petriplates

Other requirements: Incubator, Micropipettes, Tips, Inoculation loop, Distilled Water

Storage:

HiPer® Ezy MIC® Teaching Kit is stable for 12 months from the date of manufacture without showing any reduction in performance. On receipt, store MIC strips at 2-8°C. Other reagents can be stored at room temperature (15-25°C)

Important Instructions:

1. Read the entire procedure carefully before starting the experiment.
2. Perform all the microbiological experiments under aseptic conditions.
3. **Preparation of Mueller Hinton (MH) Agar plates (150 ml):** Suspend 5.7 g of MH agar into 150 ml of sterile distilled water. Sterilize by autoclaving and allow the media to cool down to 40-45°C and pour 20 ml of media on sterile petri plates.
4. One applicator stick can be used multiple times.

Procedure:

Day 1: Revival of Strains

1. Pick up a loopful of culture and streak onto MH agar plate.

2. Incubate overnight at 37°C.

Day 2: Ezy MIC™ Test

1. Take five MH agar plates and label them as 1 to 5.
2. Take a loopful of culture from the plate into a sterile test tube containing 1 ml of sterile saline. Mix the tube thoroughly.
3. Take a sterile cotton swab and dip it into test tube. While taking out the swab from the tube rotate the soaked swab firmly against the upper inside wall of the tube to get rid of excess fluid.
4. Spread the cotton swab containing the culture evenly onto the Mueller Hinton Agar plate labeled as 1.
5. Repeat step 4 for other plates i.e. plate number 2 to 5. While spreading make sure to streak the entire agar surface of the plate with the swab three times, turning the plate at 60° angle between each streaking.
6. Remove Ezy MIC™ strips from cold storage and keep it at room temperature for 15 – 20 minutes before opening. Remove one applicator stick from the bag stored at room temperature.
7. Hold the applicator in the middle and gently press its broader sticky side on the centre of Amikacine Ezy MIC™ strip.
8. Lift the applicator along with attached Ezy MIC™ strip. Place the strip at a desired position on the Mueller Hinton agar plate labeled as 1 (swabbed with test culture). Gently turn the applicator clockwise with fingers. With this action, the applicator will detach from the strip.
9. Do not press the strip. Ezy MIC™ strip is adsorbed and will firmly adhere to the agar surface within 60 seconds. Once placed, the strip should not be repositioned or adjusted once placed.
10. Similarly, follow steps 6 to 8 for other antibiotics. Note down the plate number and the corresponding MIC strip that is impregnated.
11. Incubate the plates at 37°C and observe them after 16 - 18 hours (when the growth is sufficient)

Observation and Result:

Observe the plates for MIC where the ellipse intersects the MIC scale on the strip and note down the corresponding values.

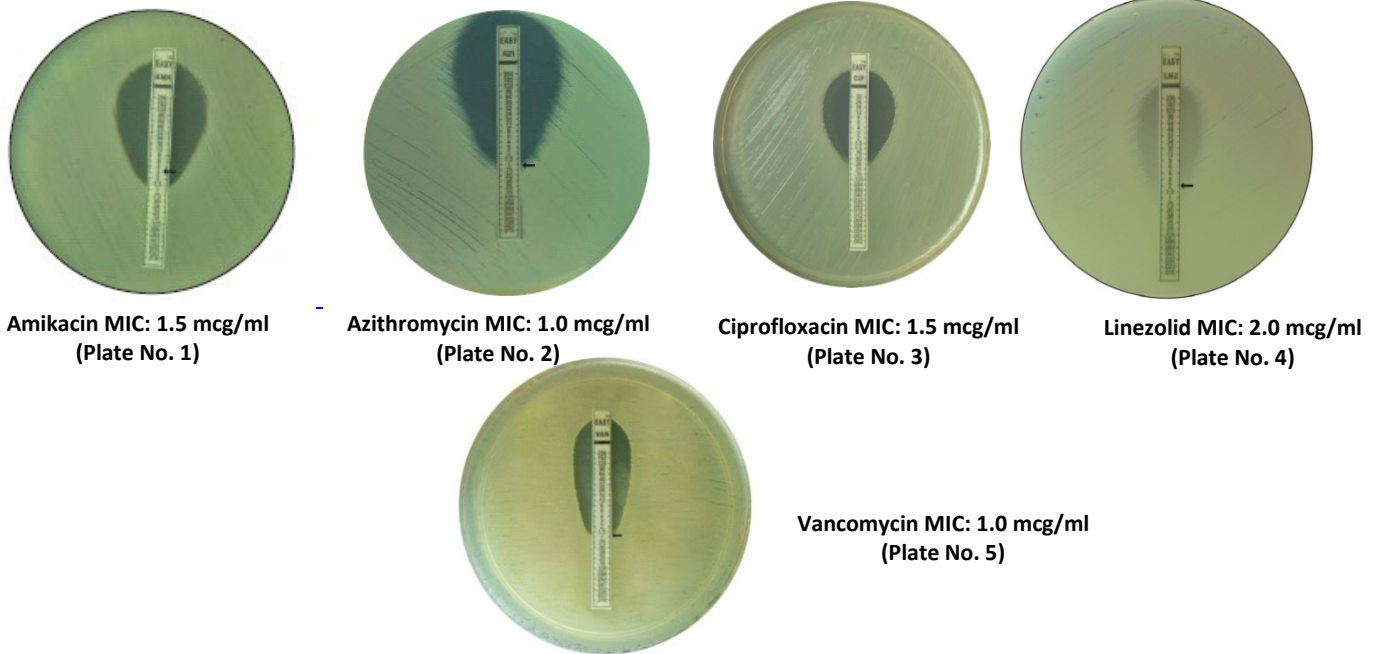


Fig 2: Plates showing MIC of different antibiotics against *S. aureus*

Reference MIC values for the antibiotics:

Antibiotics	Microorganisms to be tested	Std MIC values (mcg/ml)
Amikacin	<i>S. aureus</i> (ATCC 25923)	1.0 - 2.0 - 4.0
Azithromycin	<i>S. aureus</i> (ATCC 25923)	0.5 - 1.0 - 2.0
Ciprofloxacin	<i>S. aureus</i> (ATCC 25923)	0.12 - 0.25 - 0.5
Linezolid	<i>S. aureus</i> (ATCC 25923)	1.0 - 2.0 - 4.0
Vancomycin	<i>S. aureus</i> (ATCC 25923)	0.5 - 1.0 - 2.0

Use the following interpretive criteria for susceptibility categorization:

Microorganism Tested	Antibiotic Used	Interpretative Criteria		
		< S	I	>R
<i>S. aureus</i> (ATCC 25923)	Amikacin	16	32	64
<i>S. aureus</i> (ATCC 25923)	Azithromycin	2	4	8
<i>S. aureus</i> (ATCC 25923)	Ciprofloxacin	1	2	4
<i>S. aureus</i> (ATCC 25923)	Linezolid	4	-	8
<i>S. aureus</i> (ATCC 25923)	Vancomycin	2	4 - 8	16

S- Sensitive **I-Intermediate** **R- Resistant**

Interpretation:

The appearance of the zone of inhibition around the antibiotic strip indicates that *S. aureus* is sensitive to the antibiotics tested here namely, Amikacin, Azithromycin, Ciprofloxacin, Linezolid and Vancomycin. The minimum inhibitory concentration of these antibiotics can be determined for *S. aureus* which in turn helps in selecting most appropriate treatment regimen.

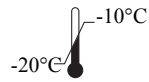
Troubleshooting Guide:

Sr. No.	Problem	Possible Cause	Solution
1	No clear zone of inhibition	Antibiotic may have been degraded	The EzyMIC® strips should be stored at 2-8°C
		Uneven bacterial growth	Ensure that the culture is evenly streaked on the plate

Please refer disclaimer Overleaf.

Technical Assistance:

At HiMedia we pride ourselves on the quality and availability of our technical support. For any kind of technical assistance, mail at mb@himedialabs.com



Storage temperature



Do not use if package is damaged



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