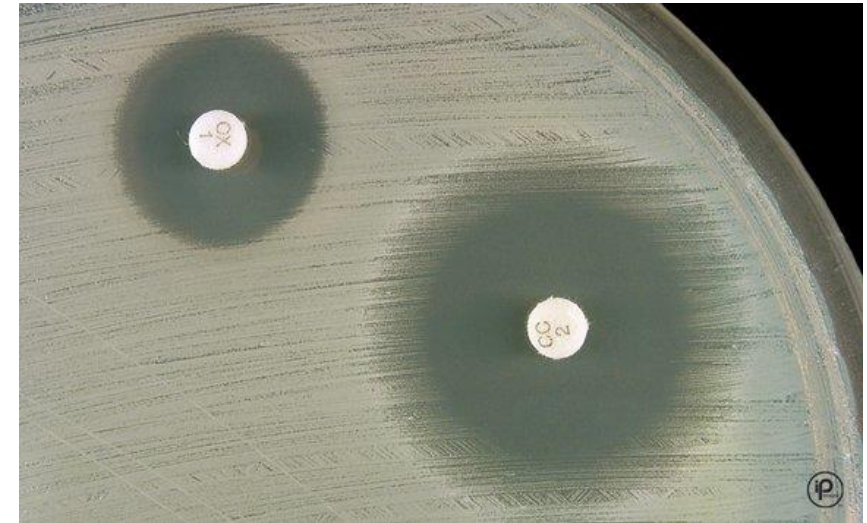


Lecture 8



Antimicrobial drug resistance



Objectives

- Enumerate Mechanisms of Antimicrobial Drug resistance
- Interpret a cultured disc diffusion plate
- Explain the principle of the following susceptibility tests: Disc diffusion, serial dilution methods and E test
- Define ***Minimal inhibitory concentration and Minimal bactericidal concentration***



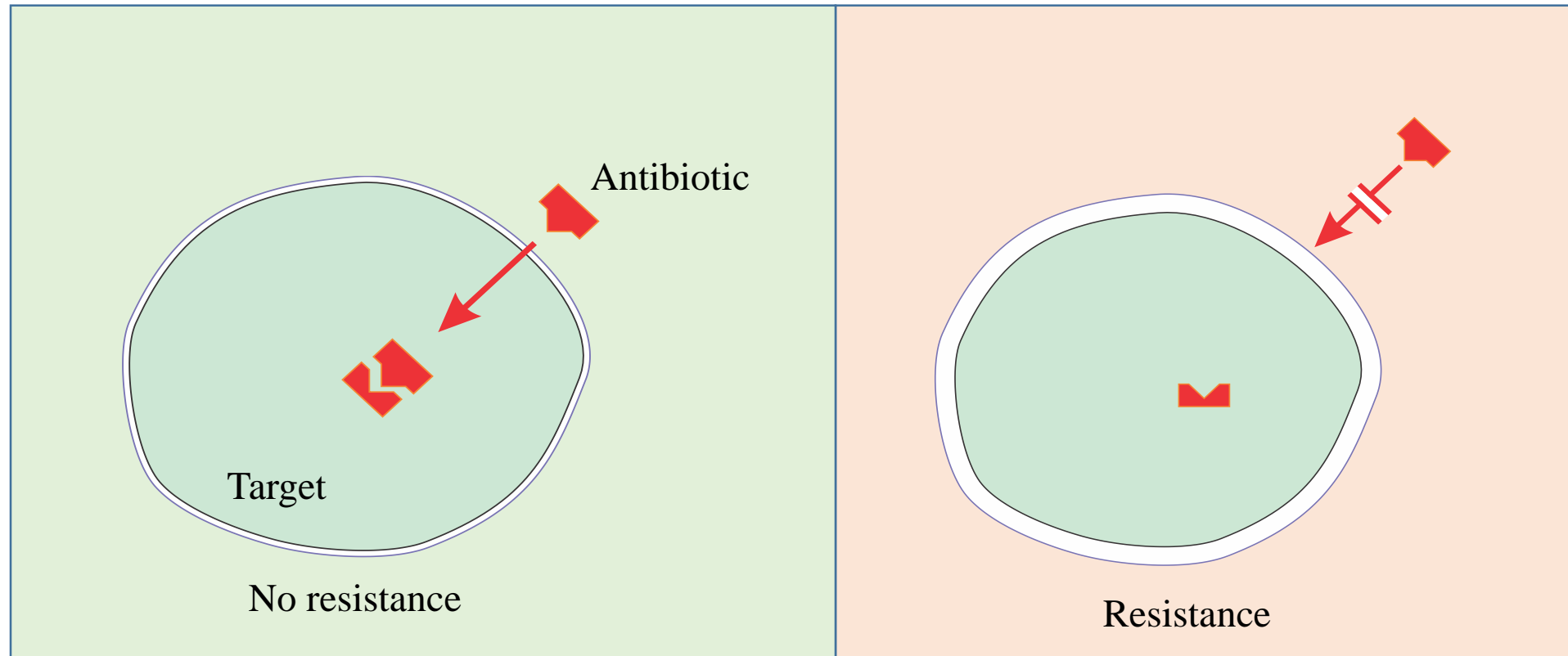
Antimicrobial drug resistance

- It is the unresponsiveness of the organisms to the administered drug (antibiotics)

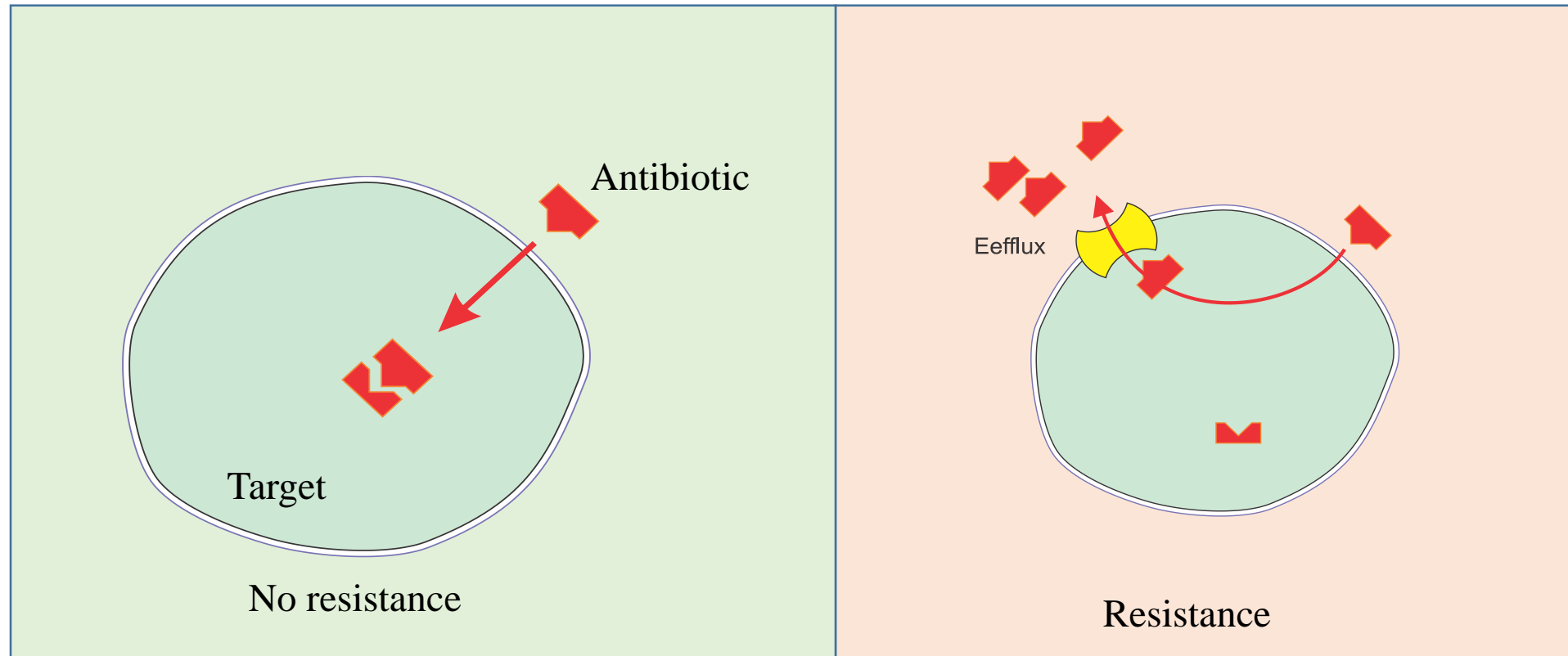
Mechanisms of Antimicrobial Drug resistance

- ① Microorganisms change their **permeability** to the drug
- ② **Pumping out** (Active efflux) of the drugs across the cell surface.
- ③ Microorganisms change their **target receptor** for the drug
- ④ Microorganisms produce enzymes that **destroy the drug**
- ⑤ Microorganisms alter the **metabolic pathway** to bypass the reactions inhibited by the drug.

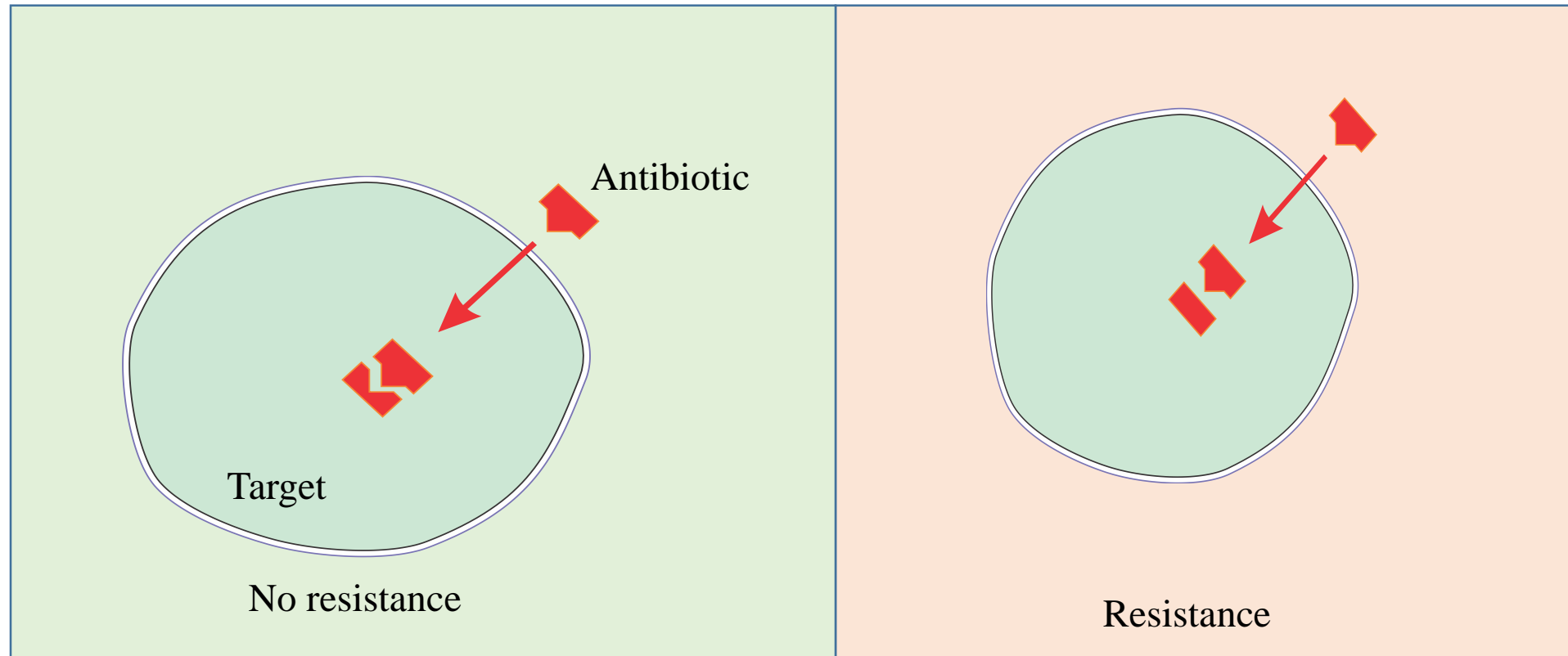
1. Microorganisms change their permeability to the drug



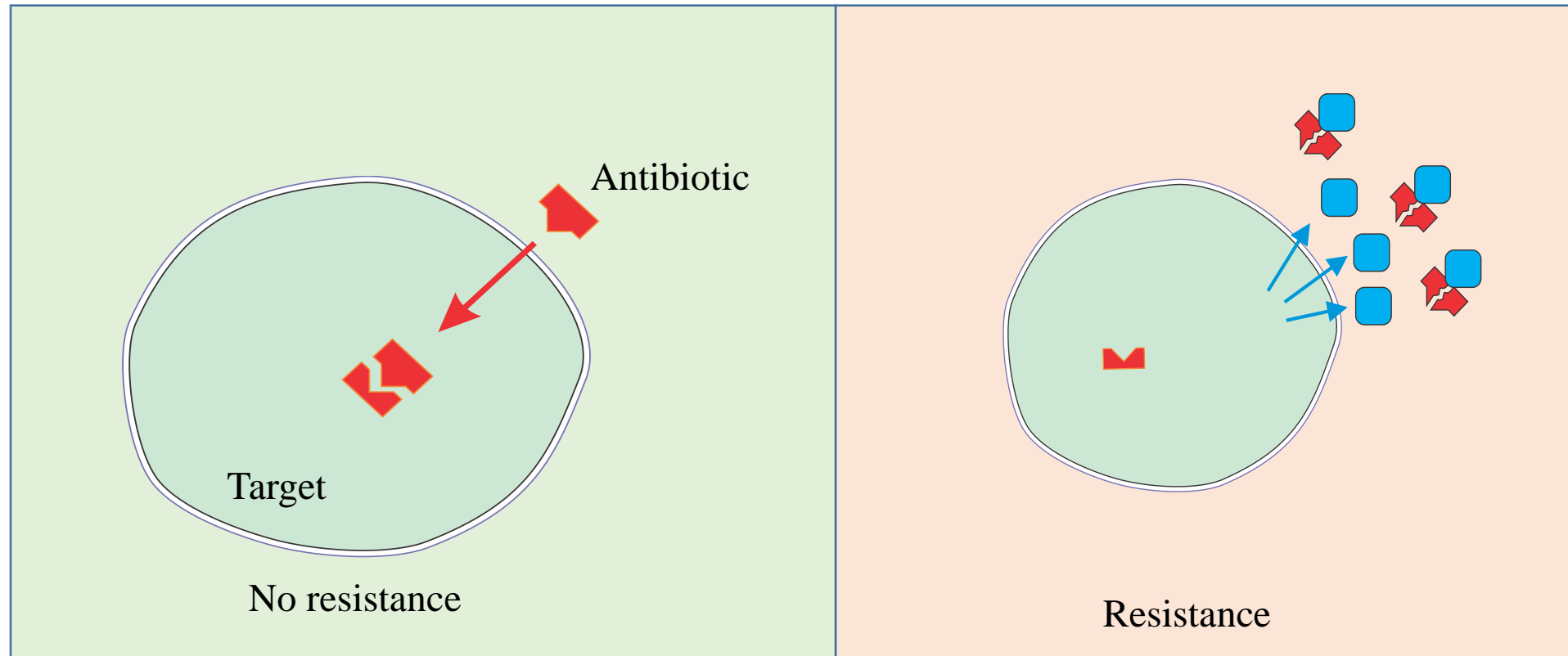
2. **Pumping** out (Active efflux) of the drugs across the cell surface.



3. Microorganisms change their target **receptor** for the drug.

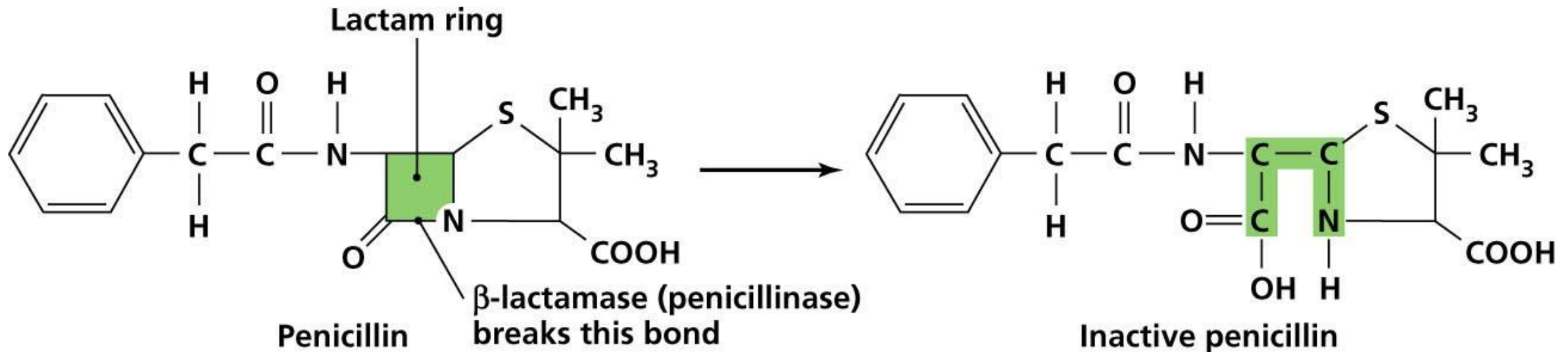


4. Microorganisms produce enzymes that **destroy** the drug.

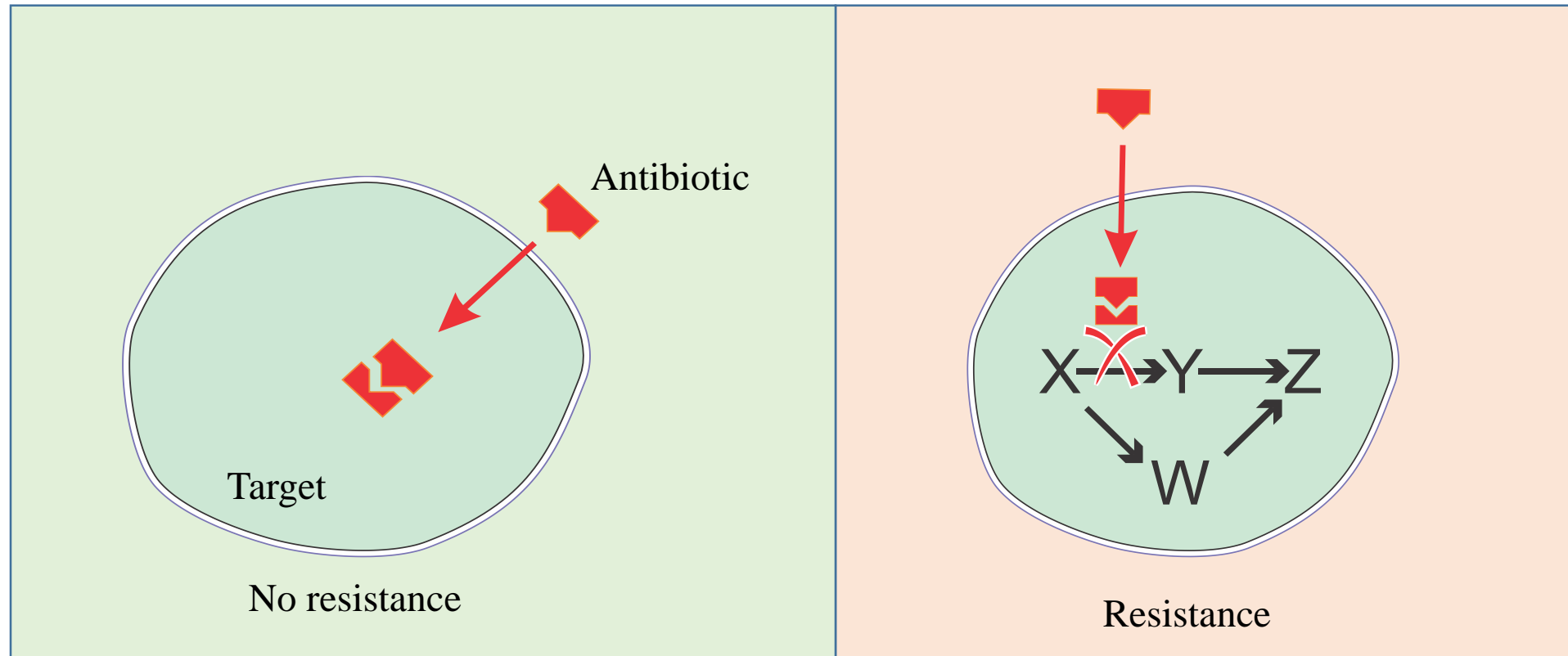


Example → beta lactamase

Beta-lactamases are enzymes produced by some bacteria and are responsible for their resistance to **beta-lactam** antibiotics like penicillin.



5. Microorganisms change the **metabolic pathway** to bypass the reactions inhibited by the drug.



Antimicrobial susceptibility tests

AIM

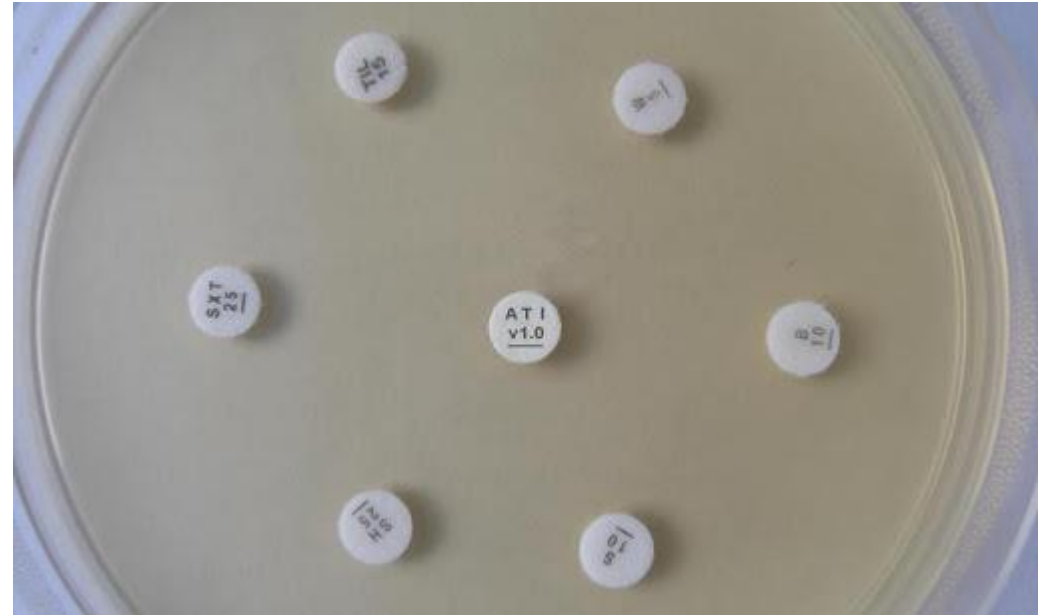
To know bacterial sensitivity to known
concentration of the Antibiotic

Method of evaluation

1. Disk-diffusion method (Kirby-Bauer)
2. Dilution Method
3. E test

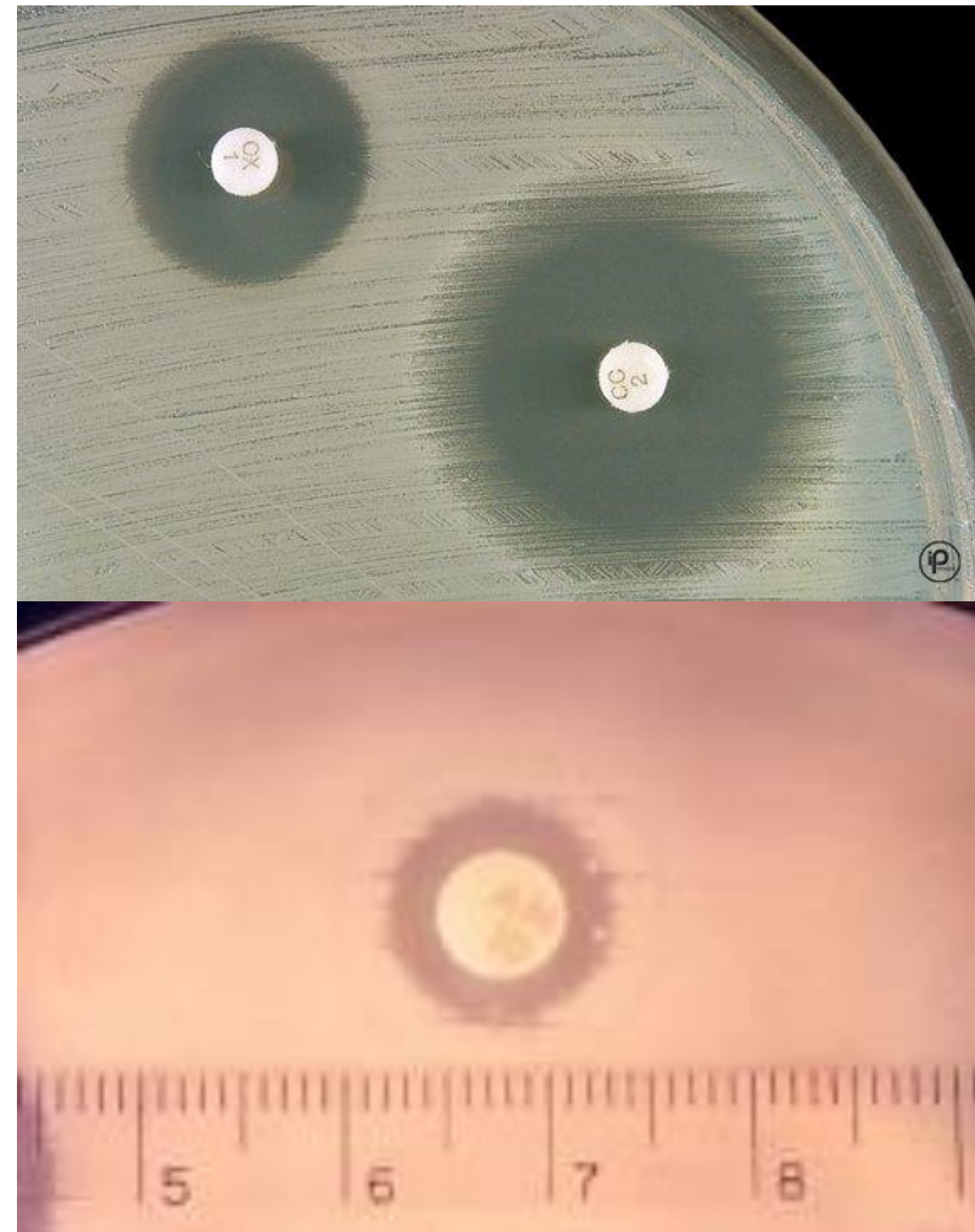
1. Disk-diffusion method (Kirby-Bauer):

- An agar plate is uniformly inoculated with the test organism.
- A paper disk impregnated with a fixed concentration of an antibiotic is placed on the agar surface.



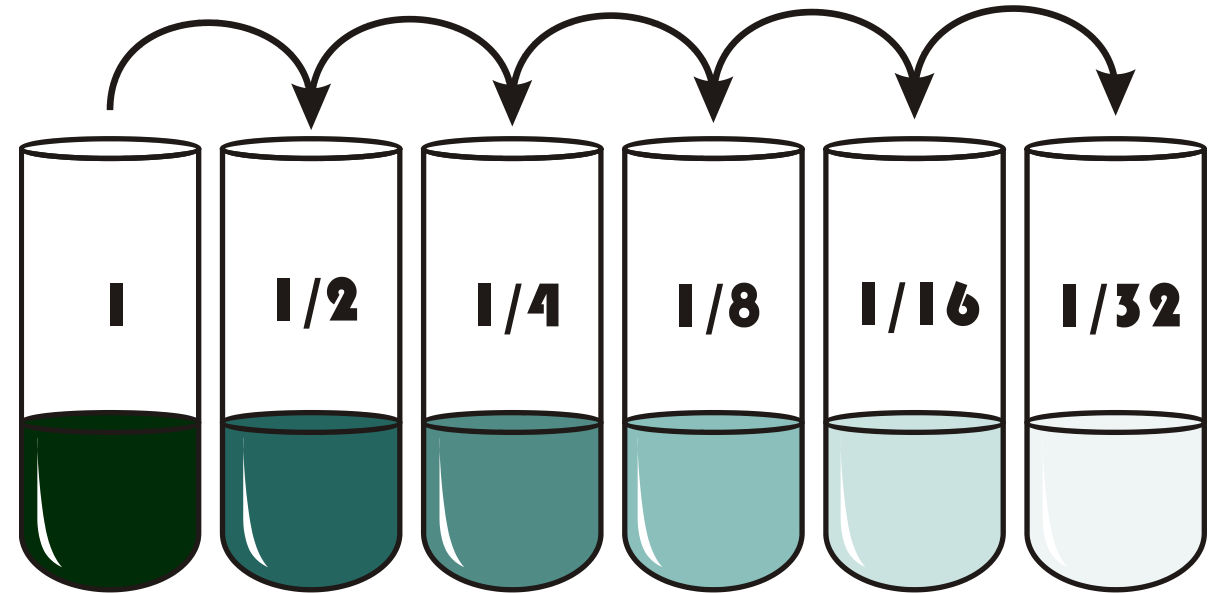
Disk-diffusion method

- If the test organism is susceptible to the antibiotic, the growth of the test organism will be inhibited around the disk (**inhibition zone**).
- The **diameter** of **inhibition zone** correlates with susceptibility of the organism.
→ A larger zone indicates a more susceptible organism.



Dilution Method

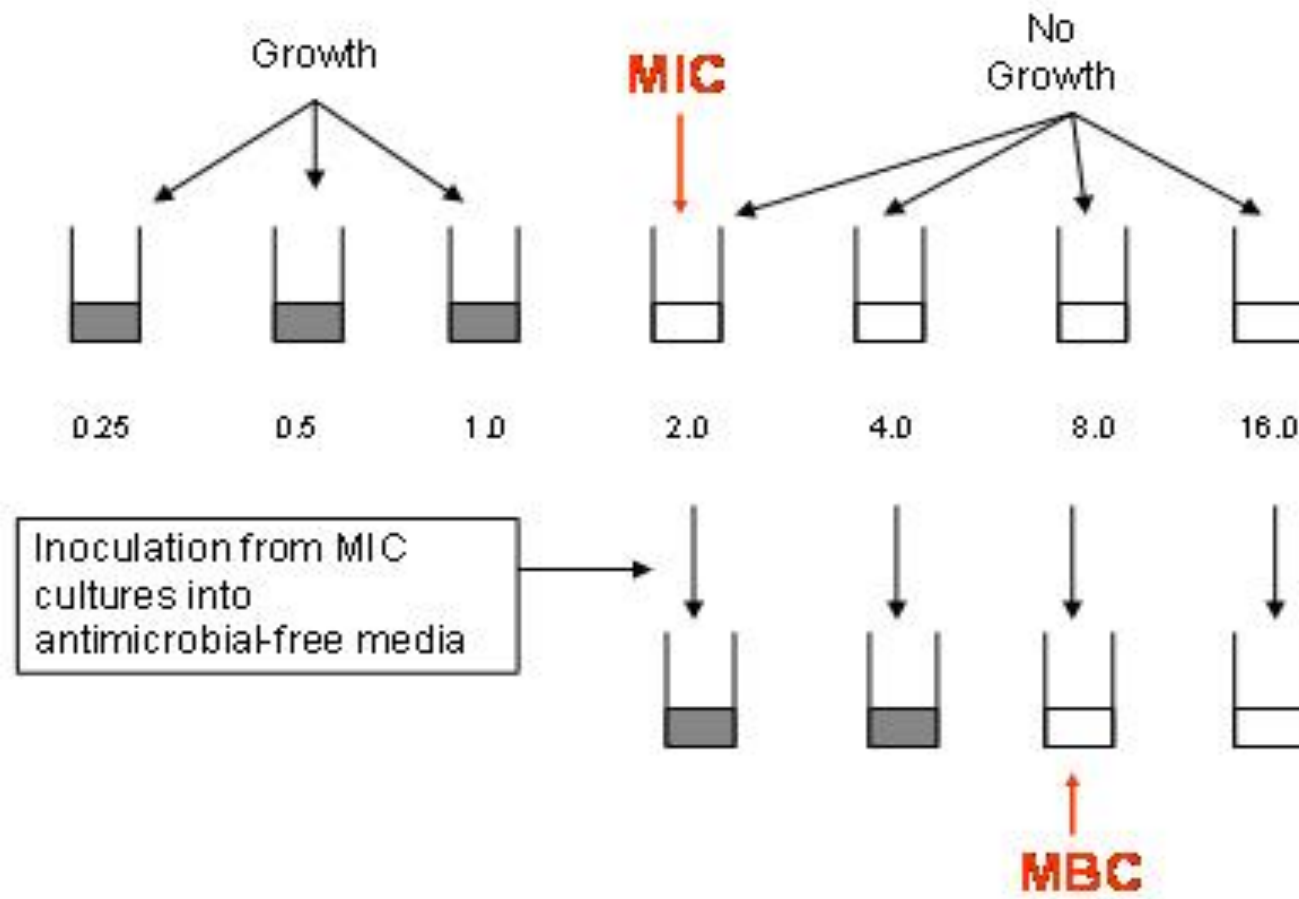
- Serial dilutions of the antibiotic are made in a liquid medium.
- A standardized number of bacteria is added to each dilution.
- After incubation the tubes are examined for visible bacterial growth (i.e. turbidity).



MIC and MBC

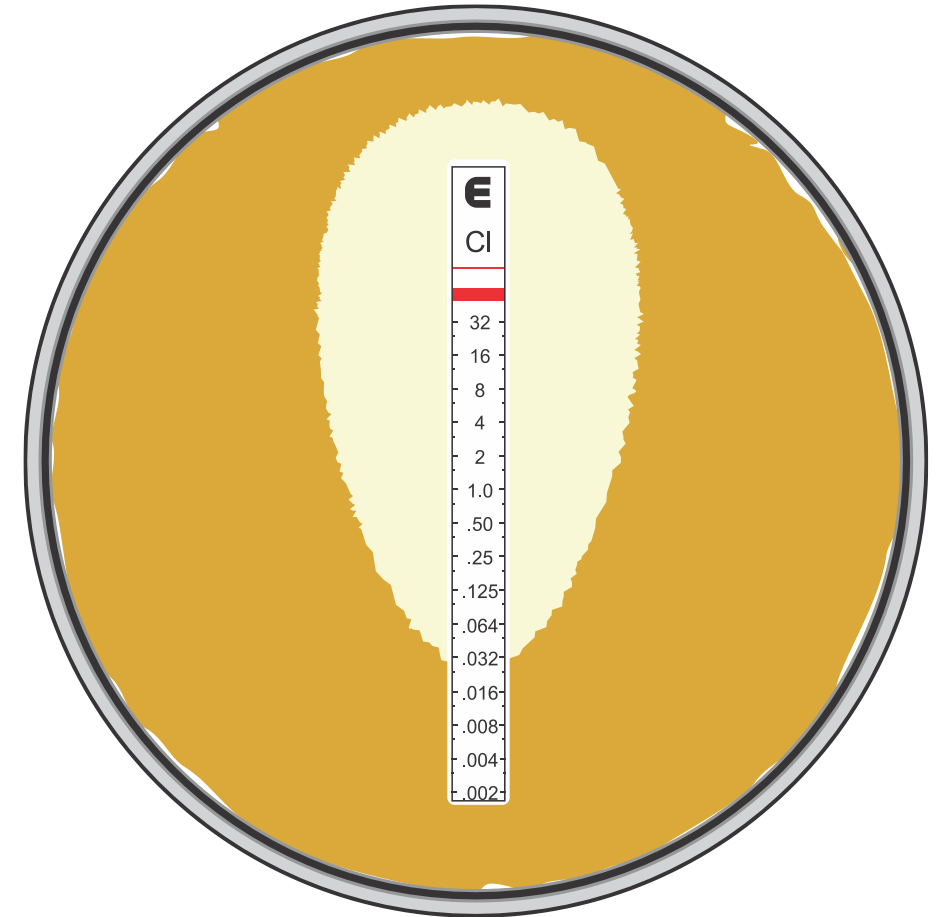
- ***Minimal inhibitory concentration (MIC)***: The **lowest** concentration of antibiotic that **inhibit** the growth of the bacteria (i.e. preventing appearance of turbidity).
- ***Minimal bactericidal concentration (MBC)***: The **lowest** concentration of antibiotic required to **kill** the bacteria.
- **MBC** can be determined by subculturing the contents of the tubes with no turbidity onto antibiotic-free medium and examining for bacterial growth. (**$MBC \geq MIC$**)

Serial Dilution Susceptibility Testing



E test

- The E test consists of a strip containing a concentration gradient of one antibiotic.
- An agar plate is inoculated with an organism, a strip is placed on the plate.
- after overnight incubation, the intersection of the growth on the scale is read to determine the MIC.



Quizzes





1. MBC is :

- A. The **lowest** concentration of antibiotic required to **inhibit** the growth of the bacteria
- B. The **highest** concentration of antibiotic required to **inhibit** the growth of the bacteria
- C. The **lowest** concentration of antibiotic required to **kill** the bacteria
- D. The **highest** concentration of antibiotic required to **kill** the bacteria





2. MIC is :

- A. The **lowest** concentration of antibiotic required to **inhibit** the growth of the bacteria
- B. The **highest** concentration of antibiotic required to **inhibit** the growth of the bacteria
- C. The **lowest** concentration of antibiotic required to **kill** the bacteria
- D. The **highest** concentration of antibiotic required to **kill** the bacteria



3. Beta lactamase enzyme is used by some bacteria to resist:



- A. Penicillin
- B. Tetracycline
- C. Polymixin
- D. Rifampicin

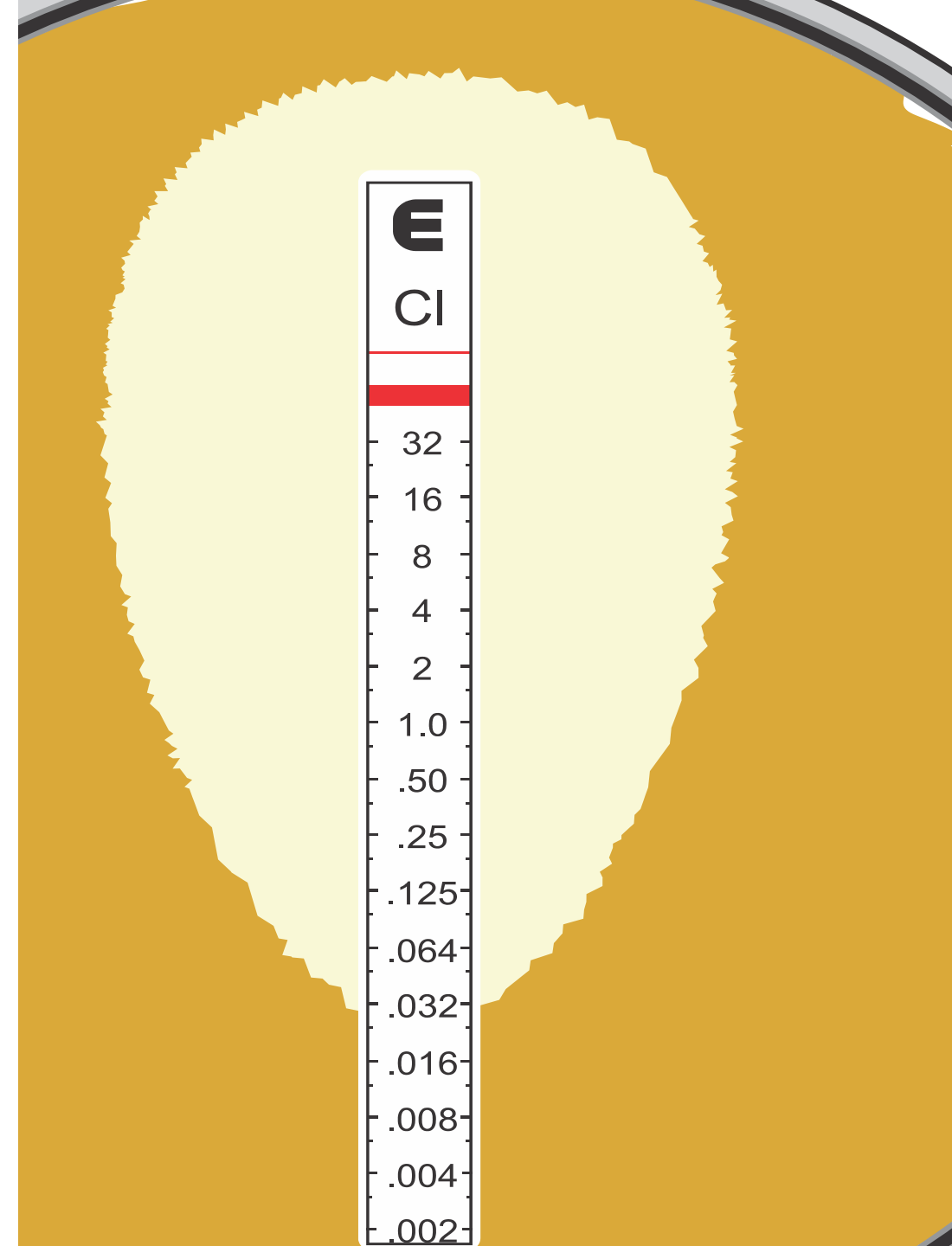


4. In Disc diffusion method of antibiotic susceptibility tests, increase in the diameter of the inhibition zone means

- A. More resistance to the antibiotic
- B. More susceptibility to the antibiotic

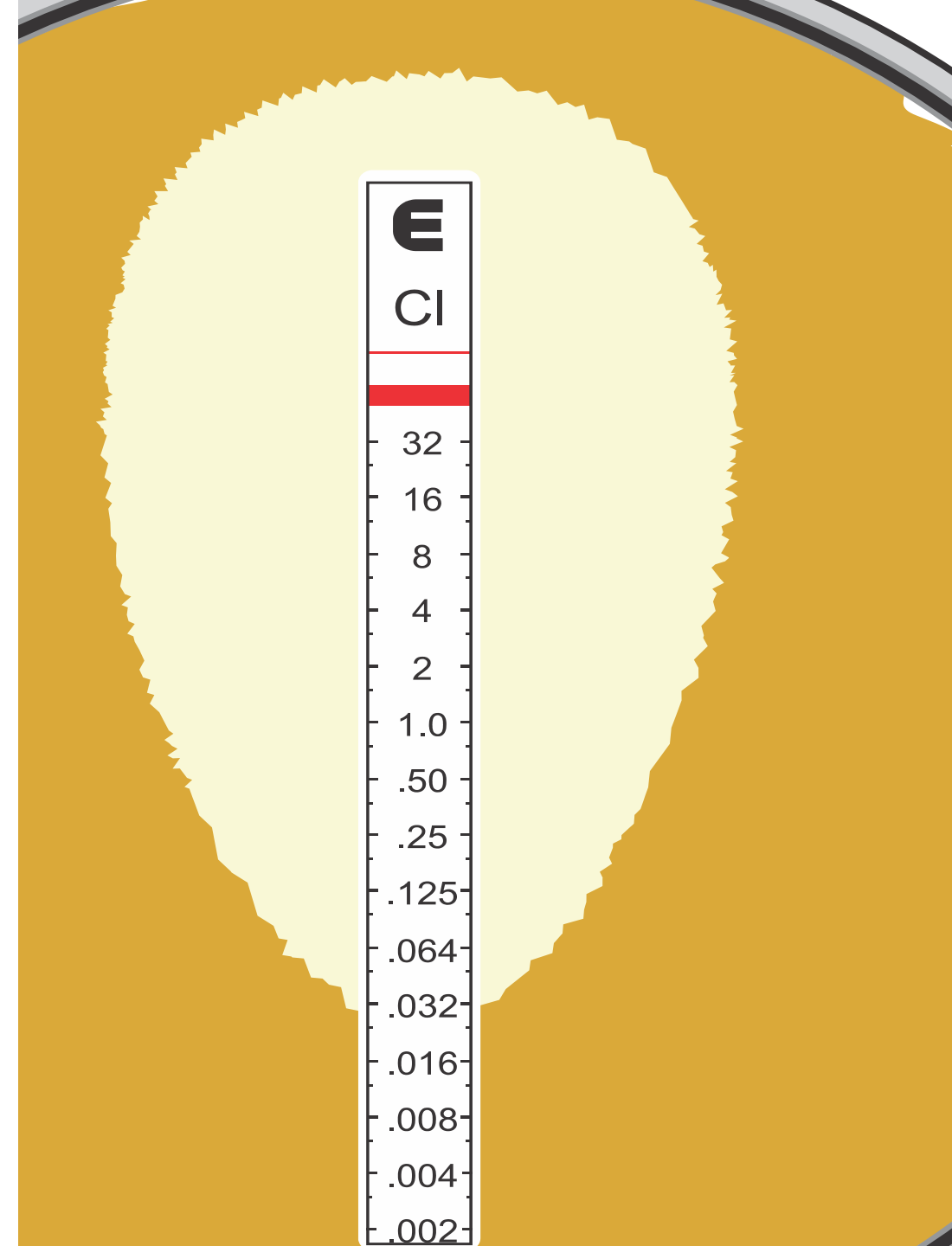
5. The name of this test

- A. Disc diffusion Method
- B. E test
- C. Broth dilution Method



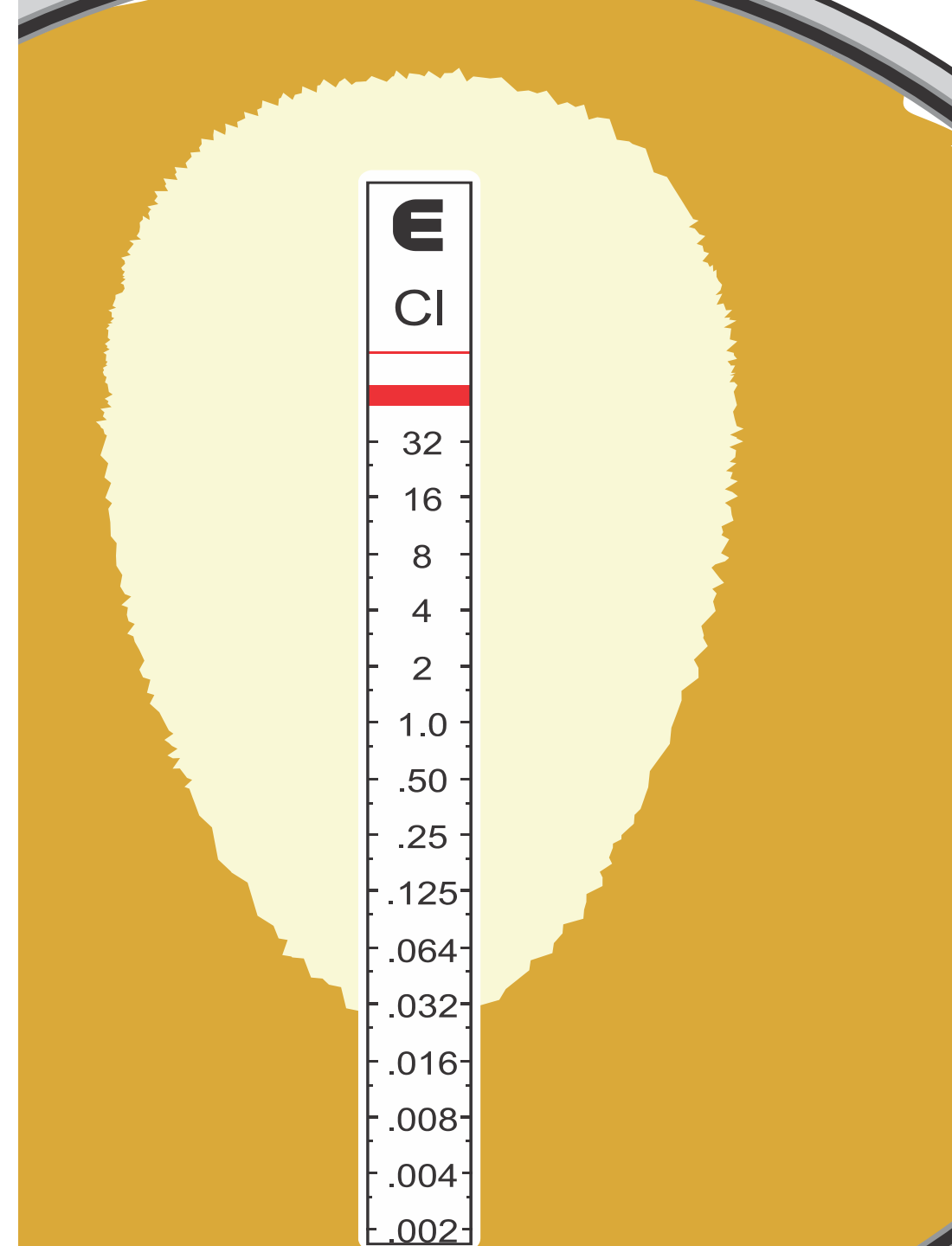
5. This test is used to:

- A. Measure bacterial susceptibility to antibiotics
- B. Identify the type of bacteria
- C. Detect bacterial antigen



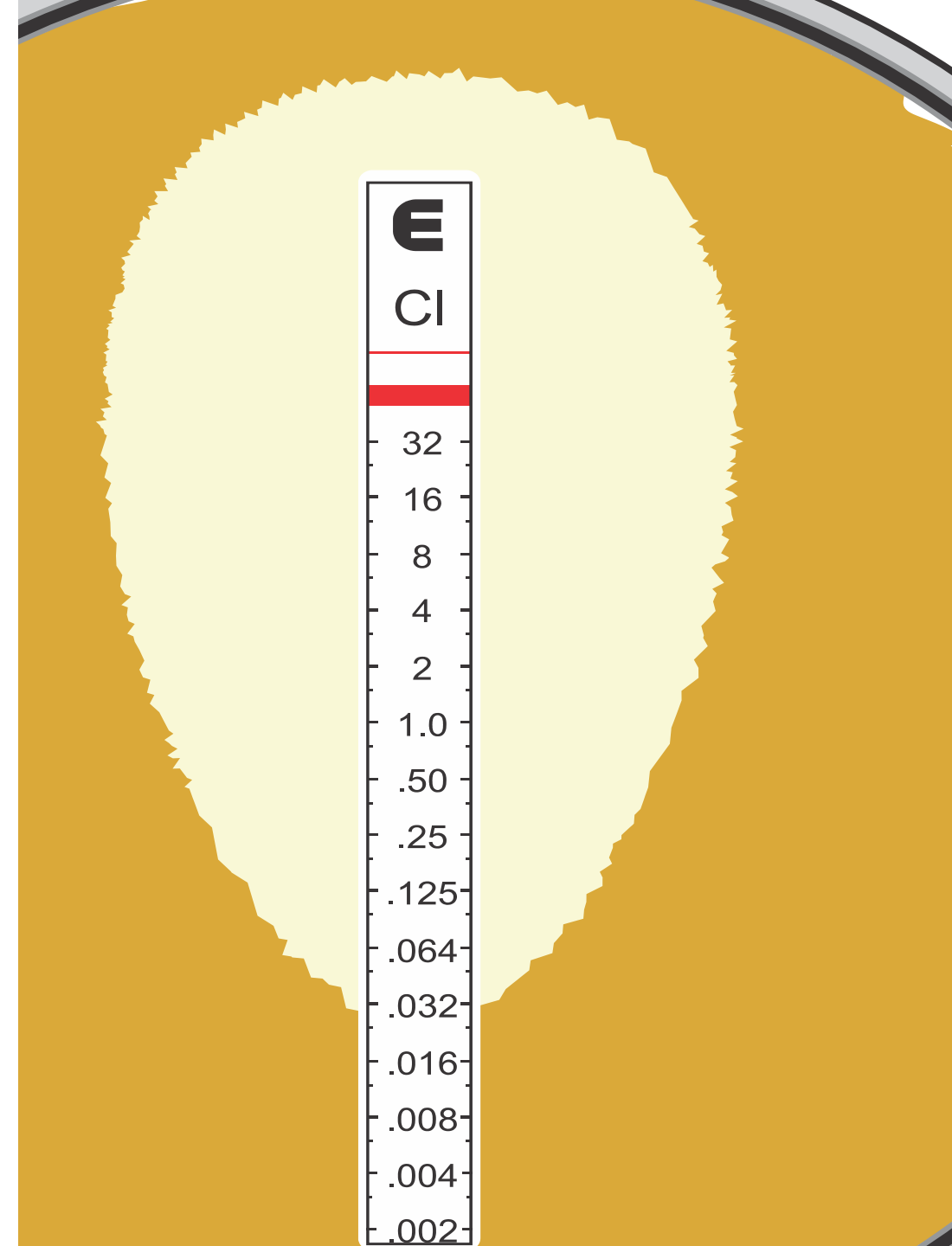
5. The MIC in this test is

- A. 0.002
- B. 32
- C. 0.032
- D. 2



5. Higher MIC means

- A. More resistance
- B. More susceptibility

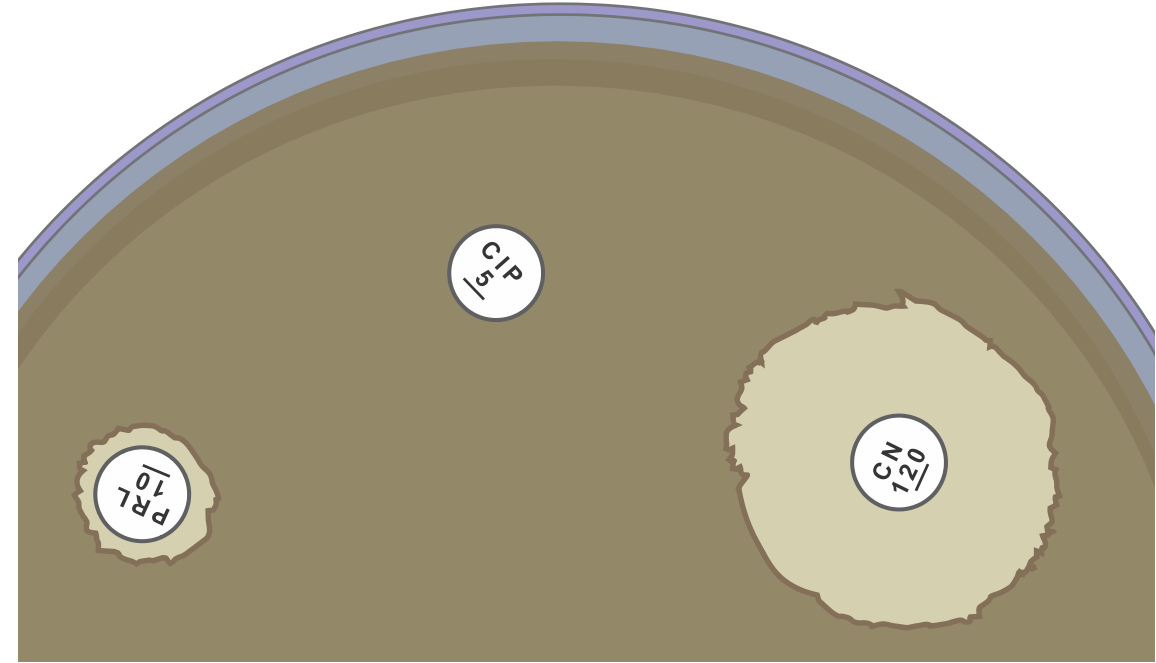


6. Mention 4 mechanisms of antibiotic resistance:

1.
2.
3.
4.

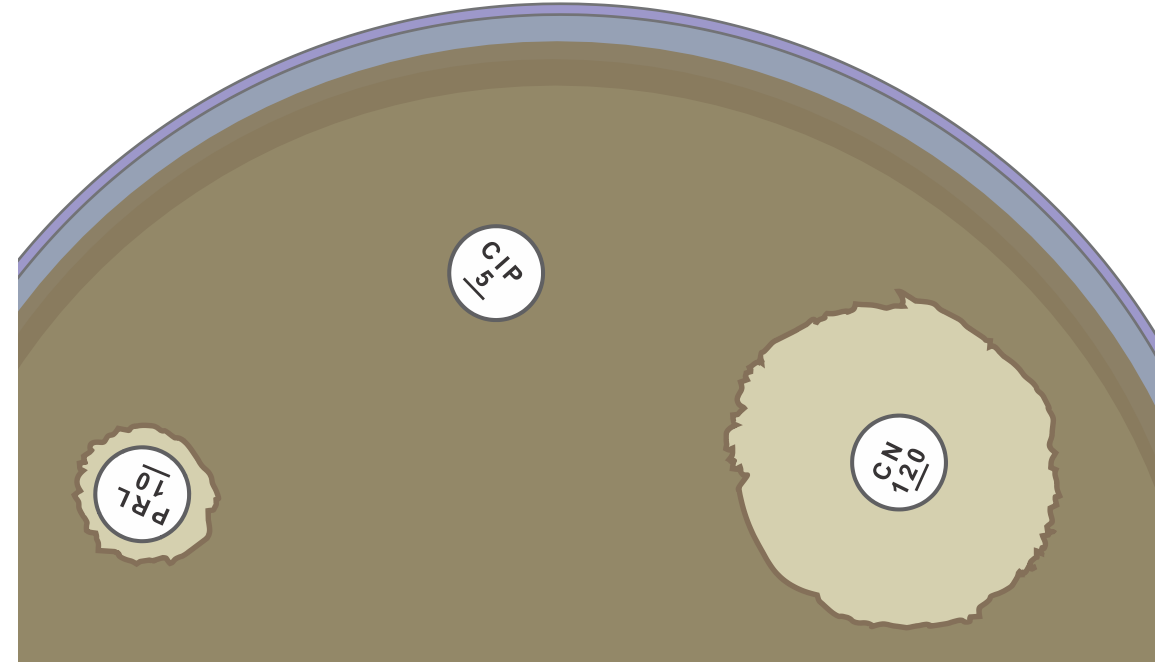
7. This test is used to:

- A. Measure bacterial susceptibility to antibiotics
- B. Identify the type of bacteria
- C. Detect bacterial antigen



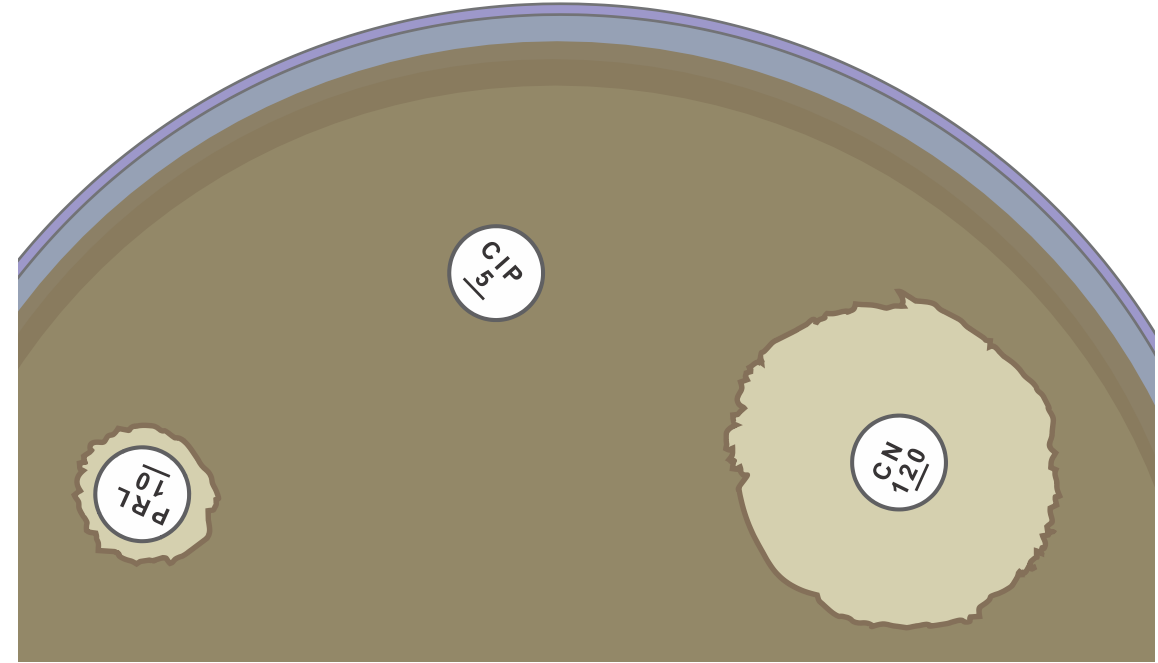
7. The name of this test

- A. Disc diffusion Method
- B. E test
- C. Broth dilution Method



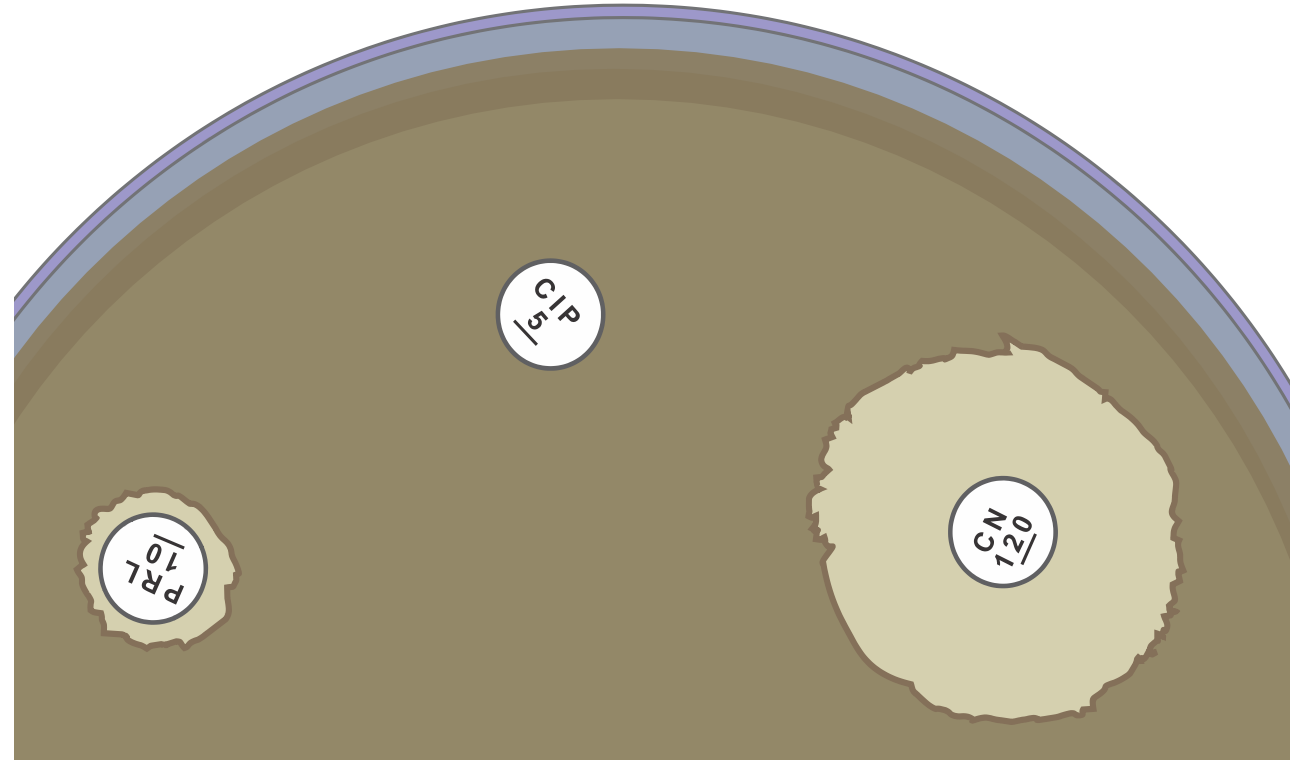
7. The increase in diameter means:

- A. More resistance
- B. More susceptibility



7 Apparently, Which is most effective antibiotic:

- A. Ciprofloxacin
- B. Piperacillin
- C. Gentamycin



CIP = ciprofloxacin

PRL = piperacillin

CN = gentamycin