

Lab. 5: Biochemical tests

6. IMViC Tests

Each of the letters in “IMViC” stands for one of these tests. “I” is for indole; “M” is for methyl red; “V” is for Voges-Proskauer, and “C” is for citrate, lowercase “i” is added for ease of pronunciation. IMViC is an acronym that stands for four different tests:

- a. **Indol test**
- b. **Methyl red test**
- c. **Voges-proskauer test**
- d. **Citrate utilization test**

To obtain the results of these four tests, three test tubes are inoculated: tryptone broth (indole test), methyl red – Voges Proskauer broth (MR-VP broth), and citrate. IMViC tests are employed in the identification / differentiation of members of family **enterobacteriaceae**.

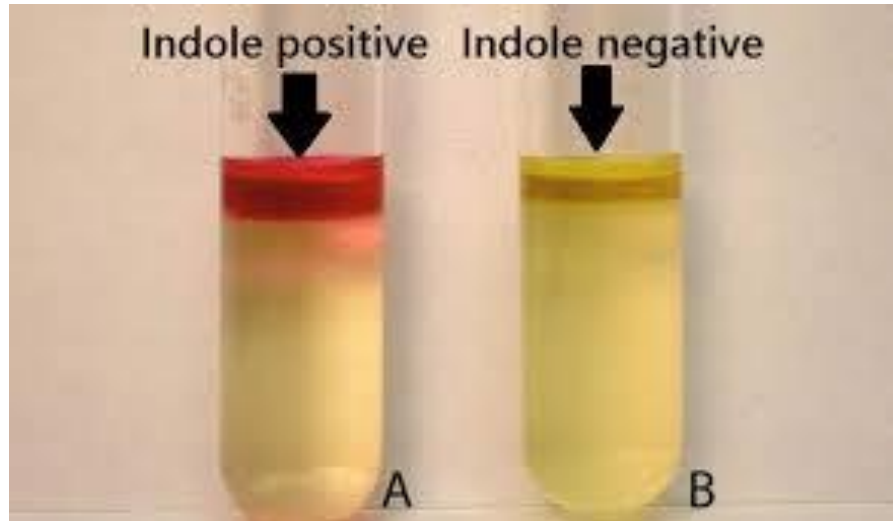
► Procedure

Cultures of any members of enterobacteriaceae have to grow for 24 to 48 hours at 37 °C and the respective tests can be performed:

a. Indole test

It is performed on **sulfide-indole-motility (SIM)** medium or in **tryptophan broth**. Result is read after adding Kovac’s reagent.

1. The positive result is indicated by the **red layer at the top of the tube** after the addition of Kovac's reagent.
2. A negative result is indicated by the lack of color change at the top of the tube after the addition of Kovac's reagent.



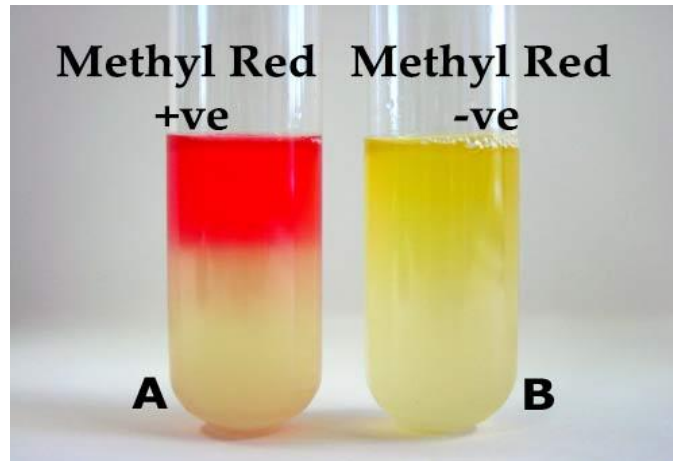
Indole test results: positive-development of Red-ring

b. Methyl red test and Voges-Proskauer test

both are done in **methyl red –Voges-Proskauer (MR-VP) broth**, but the reagents that we added differs

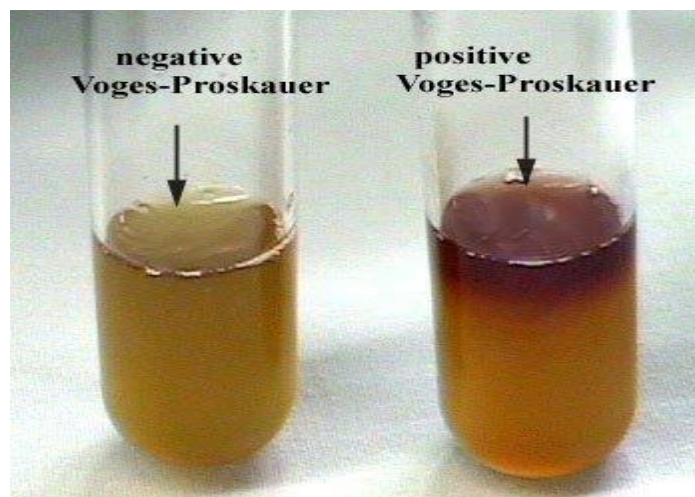
*** Methyl Red (MR) test:**

- Positive methyl red test are indicated by the development of the red color after the addition of **methyl red reagent**.
- A negative methyl red test is indicated by no color change after the addition of **methyl red reagent**.



* **Voges-Proskauer (VP) test:**

- Negative test is indicated by lack of color change after the addition of **Barritt's A** and **Barritt's B** reagents.
- A positive Voges-Proskauer test is indicated by the development of red-brown color after the addition of **Barritt's A** and **Barritt's B** reagents.

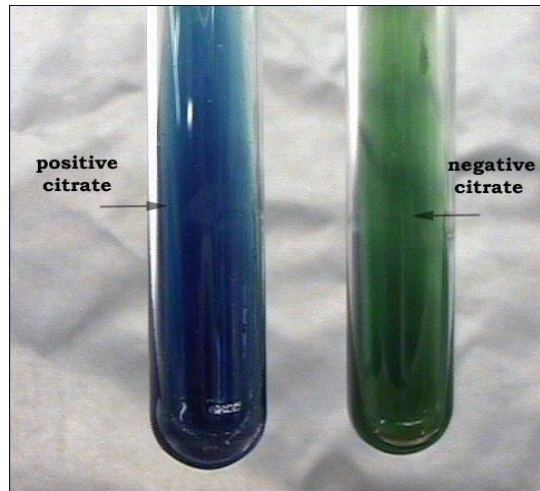


c. Citrate utilization test

The test performed on **Simmons citrate agar**:

- Negative citrate utilization test is indicated by the lack of growth and color change in the tube

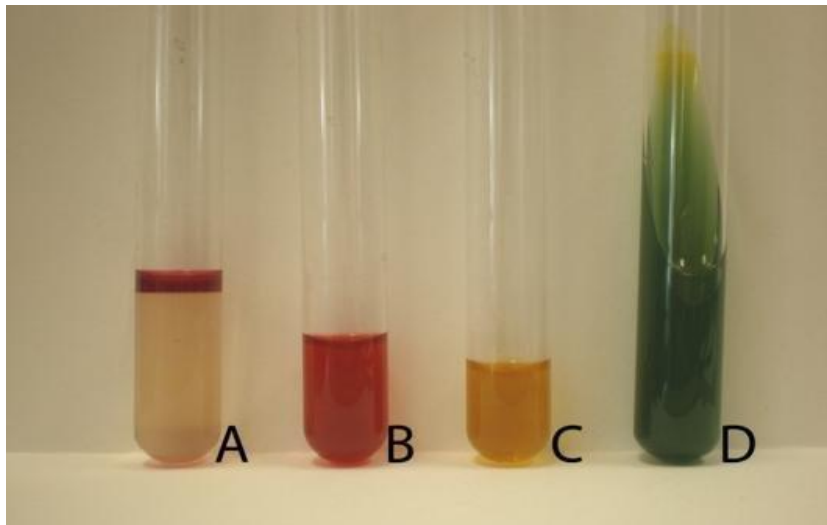
- Positive citrate result as indicated by growth and a blue color change.



► **IMViC Test results of some Genera of Enterobacteriaceae:**

1. IMViC tests of *Escherichia coli*

1. Indole: positive
2. Methyl-Red: positive
3. Voges-Proskauer test: Negative
4. Citrate test: Negative



IMViC Test of *E. coli*: (++ - -)

2. IMViC tests of *Enterobacter aerogenes*

1. Indole: Negative
2. Methyl-Red: Negative
3. Voges-Proskauer test: Positive
4. Citrate test: Positive

3. IMViC tests of *Proteus vulgaris*

1. Indole: Positive
2. Methyl-Red: Positive
3. Voges-Proskauer test: Negative
4. Citrate test: Negative

4. IMViC tests of *Citrobacter freundii*

1. Indole: Negative
2. Methyl-Red: Positive
3. Voges-Proskauer test: Negative
4. Citrate test: Positive