

# MAKING *ESCHERICHIA COLI* COMPETENT FOR DNA TRANSFORMATION

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## INTRODUCTION

It is found that bacteria can carry out different functions by inserting a plasmid. In this experiment, our team prepared competent *Escherichia coli* that readily take up foreign plasmids, pUC 19, which has ampicillin resistant strain, so that it can be ampicillin resistant. The objective of our project is to determine the different parameters that could enhance the *E.coli* transformation into a competent strain.

In our experiment, we investigated 3 different parameters: the tubes used, the growth medium and the Optical Density of the *E.coli*.

Before the experiment, our group hypothesized the results that we would obtain after the transformation.

The hypothesis are as follows:

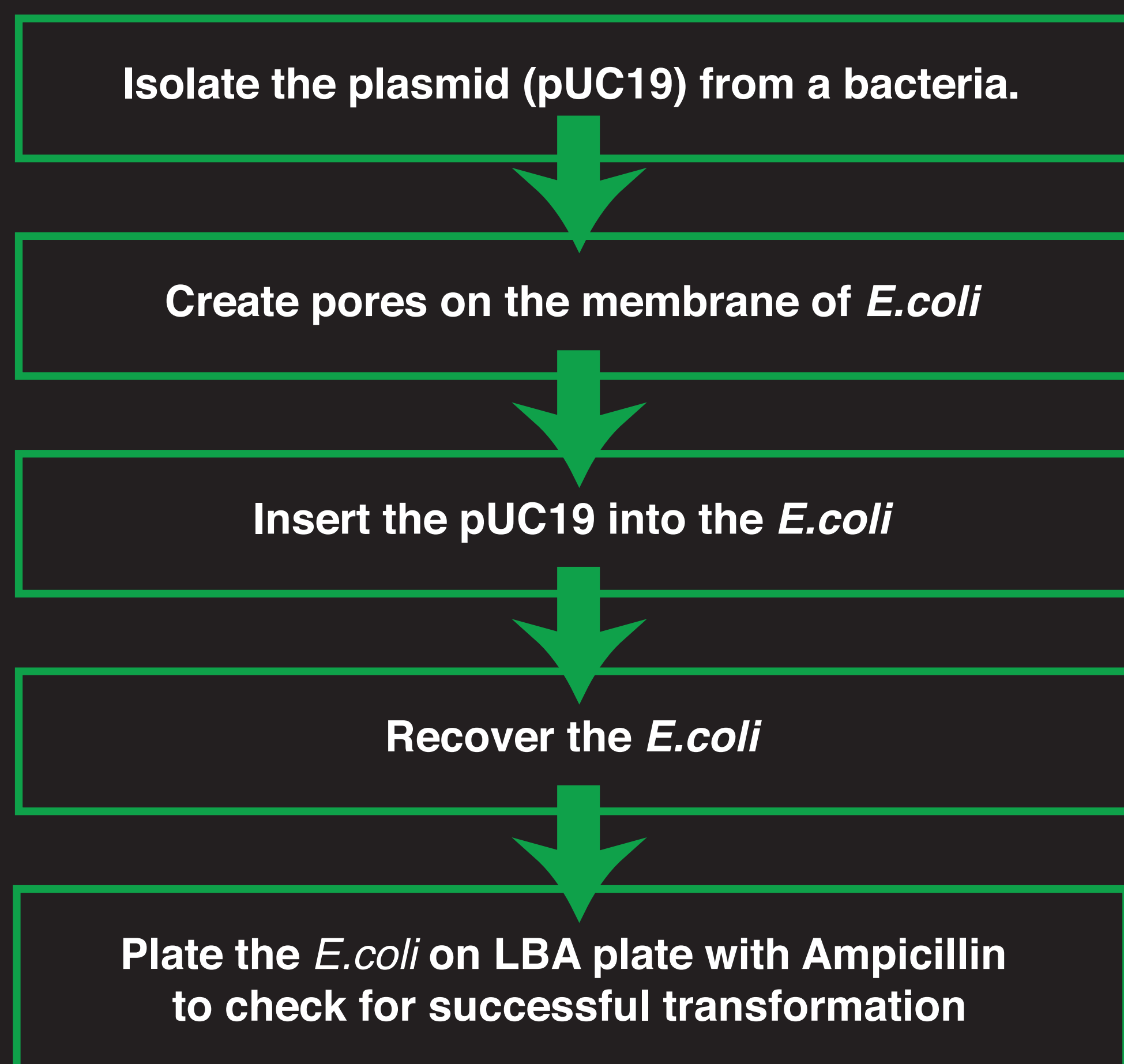
**Tubes used:** the falcon tube will have better yield of transformed bacteria.

**Growth medium:** SOC will result in more successfully transformed *E.coli* colonies.

**Optical Density:** Cells with OD<sub>610</sub> 0.450 will have the highest number of successfully transformed bacteria.

## METHODOLOGY

### Preparation



### Collecting Data

Our results were obtained through the counting of the number of *E. coli* colonies on LBA plate containing Ampicillin. We decided that for the variable to be considered to have an effect on the transformation efficiency of *E. coli* DH5-α strain, there must at least be a difference of 100 colonies on each plate.

## RESULTS / DATA



Figure 1: 50 µl *E. coli* DH5-α strain plated on LBA plates containing Amp. The successfully transformed *E. coli* have multiplied to form colonies on plates. The plates shown contain *E. coli* grown to OD<sub>610</sub> 0.450. This is repeated for different OD values such as OD<sub>610</sub> 0.450 and OD<sub>610</sub> 0.817.

OD <sub>610</sub> of <i>E. coli</i> DH5-α strain	Number of transformants on LBA plate containing Amp				
	R <sub>1</sub>	R <sub>2</sub>	R <sub>3</sub>	R <sub>4</sub>	Average
0.205	42	161	51	61	80
0.450	201	302	209	285	249
0.817	290	136	140	262	207

Table 2: Effect of OD<sub>610</sub> of *E. coli* DH5-α strain on transformation efficiency.

Growth Medium	No. of bacteria colonies formed				
	R <sub>1</sub>	R <sub>2</sub>	R <sub>3</sub>	R <sub>4</sub>	Average
LB Broth	134	160	257	273	206
SOC	261	153	467	171	263

Table 3: Effect of growth medium on the number of *E. coli* colonies formed.

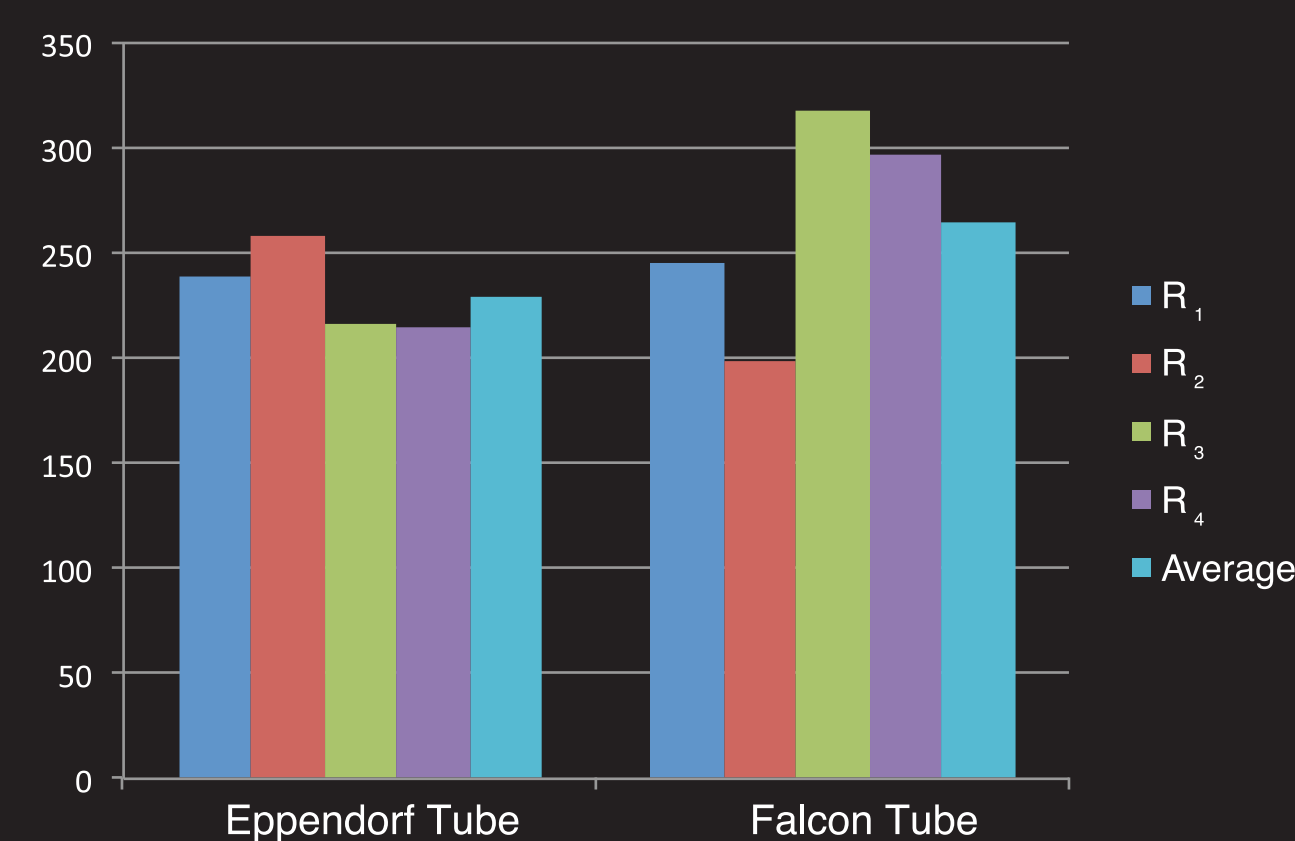


Figure 3: The number of bacteria colonies seen on LBA plate from using different storage tubes: Eppendorf and Falcon tubes.

## CONCLUSION

Through the investigation of three parameters, our experiments have proven that:

### Effect of Optical Density of *E. coli*

*E. coli* at OD 0.45 and OD 0.8 will yield the most ampicillin resistant cells as between OD<sub>610</sub> 0.3 to OD<sub>610</sub> 1.0, *E. coli* are in the active log phase and OD<sub>610</sub> 0.205, *E. coli* are in the lag phase. Hence, we can conclude that *E. coli* at exponential phase yield the highest transformation efficiency.

### Effect of aeration (Falcon/Eppendorf)

There was no difference whether falcon tubes or eppendorf tubes were used. This could be because there was sufficient oxygen for the *E. coli* to recover. Hence, we can conclude that the effect of tubes for storing *E. coli* is negligible.

### Effect of growth medium (SOC/LB)

There was also no difference in the number of ampicillin resistant *E. coli* grown on LBA plates with Amp. Thus, we concluded that the effect of growth medium on transformation efficiency is minimal.