

# Culturing Escherichia coli cells

Created on: 20-03-2019 - Last modified on: 08-11-2019

## SCOPE OF THE METHOD

<b>The Method relates to</b>	Other: Recombinant DNA technology
<b>The Method is situated in</b>	Basic Research, Regulatory use - Routine production
<b>Type of method</b>	In vitro - Ex vivo
<b>This method makes use of</b>	Other (e.g. bacteria): Escherichia coli, DH5alpha, C43 (DE3), BL21 (DE3)

## DESCRIPTION

### Method keywords

bacterial cells  
cell culture  
protein expression  
E. coli  
prokaryote

### Scientific area keywords

microbiology  
biotechnology  
Recombinant DNA technology

### Method description

E. coli is one of the organisms of choice for the production of recombinant proteins.

DH5 alpha cells are commonly used for maintenance, propagation and mutation, whilst BL21(DE3) and C43(DE3) are mainly used for expression of the transgene. The advantage of C43(DE3) is that it is used to produce proteins that are expressed poorly in BL21 (DE3) or that are very toxic to the host organism. All strains can be cultured in Lysogeny Broth (LB) medium or LB agar plates with an appropriate antibiotic for positive selection of the clones. For induction of protein expression, isopropyl-b-thiogalactoside (IPTG) in a concentration of 0.2 mM - 1 mM can be used. In case you have a problems with leaky expression, 1 % w/v glucose can be added to the LB medium for excellent growth of the bacteria. Transformation of the cells can be achieved by heat shock or electroporation.

### **Lab equipment**

Biosafety cabinet ;  
Bunsen burner ;  
Petri dishes.

### **Method status**

Still in development  
History of use  
Internally validated

### **PROS, CONS & FUTURE POTENTIAL**

#### **Advantages**

Fast growth kinetics (doubling time 20 mins) ;  
High cell density cultures are easily achieved ;  
Readily available and inexpensive components for media ;  
Easy transformation.

#### **Challenges**

No post-translational modifications (i.e. prokaryote).

#### **Future & Other applications**

Every researcher that will need a purified protein can obtain it in a recombinant form.

## REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION

### Associated documents

## PARTNERS AND COLLABORATIONS

### Organisation

**Name of the organisation** Vrije Universiteit Brussel

**Department** Pharmaceutical and Pharmacological Sciences (FARM)

**Specific Research Group or Service** In Vitro Toxicology and Dermato-cosmetology

**Country** Belgium

**Geographical Area** Brussels Region

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