Cinnabar High-Yield *E. coli* Growth Media

PRODUCT NUMBER: 3C8488

APPLICATION:

Cinnabar high-yield growth medium is a rich, complex, animal-free growth medium for plasmid DNA and protein production in *E. coli*. This medium contains glycerol, yeast extract, soytone, salts, and trace metals.

Cinnabar medium is suitable for fermenters and bioreactors. Teknova developed this medium based on Studier's original medium for protein expression using induction in *E. coli*[1].

INFORMATION AND INSTRUCTIONS:

General usage

- Cinnabar medium can be used following standard microbiological protocols for *E. coli*. Optimal growth conditions require 37°C incubation for approximately 18 hours under shaking conditions (300 rpm).
- For best results, use a fresh inoculum culture (prolonged storage on ampicillin or carbenicillin plates can affect subsequent cell growth). Add inoculum such that the titer after addition to media is >1000 cells/mL; lower titers can increase doubling times and reduce yields[2].
- The media contains 0.7% glycerol as a carbon source. Additional sugars (e.g., glucose, sucrose, arabinose, mix of lactose/glucose/glycerol) are available separately.

PROTEIN INDUCTION

- Cinnabar media can be used with fast induction protocols.
- Supplement Cinnabar media with 0.8% glucose to suppress basal levels of expression in T7/lac inducible systems, especially if the protein expressed may be toxic.

Note: in some systems, protein induction can be detected when cells are grown in Cinnabar media supplemented with <0.8% glucose and no IPTG.

- In the presence of 0.8% glucose, induction occurs with IPTG concentrations ranging from 0.01–0.10 mM with 0.06 mM being optimal. Under these conditions, induction begins at 14 hours.
- For typical fast induction protocols, total culturing time ranges from 14 to 20 hours with induction occurring after IPTG addition in the last 3 to 4 hours.
- Cultures grown at 37°C may need to be diluted after overnight growth. Ideally, OD600 should be between 0.4 to 0.8. Follow guidelines for your bacterial strain.
- In cases where you need to express your protein at lower temperatures (e.g., to minimize aggregation of insoluble, recombinant proteins), BL21 (DE3) has a long lag phase of ~7 hours at 30°C in Cinnabar media (compared to ~2 hours at 37°C in Cinnabar media).

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PLASMID PURIFICATION

If using a kit to purify plasmid DNA, we strongly recommend that you precipitate the cell lysate with isopropanol and resuspend the pellet before loading onto the column. This step prevents overloading of the column and almost doubles the yield capacity of the columns.

REFERENCES:

1. Studier FW (2005) Protein production by auto-induction in high density shaking cultures. Protein Expr Purif 41:207–234.
2. Irwin PL, Nguyen LHT, et al. (2010) Evidence for a bimodal distribution of *Escherichia coli* doubling times below a threshold initial cell concentration. BMC Microbiology 10:207.

Questions?

We're here to help. Get in touch with us at info@teknova.com or call 1.800.209.4488.