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# **Certificate of Analysis**

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**Product:** CIRCLEGROW Sterile Solution **Catalog No.:** 3000-1X4; 3000-104; 3000104

**Lot No.:** 96137

CIRCLEGROW Sterile Solution: Molecular Biology Certified Bacterial Growth Medium

Rich Medium for the growth of E. coli and certified for plasmid and other bacterial cultures in liquid

medium.

# Formulation and Molecular Biological Specifications of Components:

Proprietary formulation containing Yeast Extract, Tryptone Broth, and Vitamins. Biological buffers have been added for proper pH adjustment.

### Recommended Use:

For laboratory use only.

#### Storage:

Store CIRCLEGROW sterile solution at ambient temperature (15-30°C).

#### Shelf Life:

2 years.

#### Quality Assurance Information

All MP Biomedicals products are thoroughly tested to ensure reliable results in the laboratory.

#### Media Preparation:

Reagents are tested and chosen for their ability to promote optimum growth of *E. coli* when combined in CIRCLEGROW formulations. Chosen reagents are combined in the accurate proportions according to the formula listed above and thoroughly blended for a uniform distribution. A sample of the blended formulation was used to prepare liquid. The combinations were mixed to dissolve components and then autoclaved at 121C for 15 minutes.

Note: Autoclaving at hotter temperatures or for longer periods will result in darkening of the sterile medium, (a darker color does not normally affect growth of cells, only the appearance of the sterile medium).

#### **Quality Control Assay:**

CIRCLEGROW liquid solutions were mixed by swirling to evenly disperse all components and visually inspected after autoclaving for complete dissolution of components. The sterility of CIRCLEGROW liquid solutions were checked.

#### **Cell Growth and Plasmid Yield Tests:**

Duplicate 40 ml aliquots of autoclaved CIRCLEGROW were inoculated with E. *coli* host cells containing a test standard pUC-based high-copy number plasmid (TSP) containing the Kanamycin resistance gene. The cultures were aerated for 24 hours at 37°C in a shaking incubator with 20 ug/ml Kanamycin. A 10 ml aliquot was removed from each culture and a 3.6ml of 10ml aliquot was processed by the RPM "Rapid Pure Miniprep" procedure to measure plasmid DNA yield. The pH of the cultures was measured and the cells in the remaining 30 ml of each culture were pelleted at 4000 x g, supernatant discarded, and pellets drained and weighed.

# **Results of Quality Control Assay**

	Acceptable Ranges for Test Standard Lot	Small Capsules	Powder Pouch	Large Capsules
pH and Temp.	6.6-7.2 @ 25°C	n/a	6.74 @25°C	n/a
Clarity	Clear	n/a	Clear	n/a
pH (Harvest)	report result	n/a	6.67 @25°C	n/a
TSP Yield per 1.5 ml	up to 15 ug	n/a	0.8 ug	n/a
Cell Pellet Weight/30 ml	>0.07 g	n/a	0.500 g	n/a

The CIRCLEGROW Sterile Solution passed the sterility test.

# Conclusions:

This lot of CIRCLEGROW Sterile Solution is released for product sales.

*12/29/2016 - John Huang, PhD* QBioGene/MP Biomedicals, LLC

Quality Control Manager

MSDS available online at www.mpbio.com