DePlasma™

For Primary Cell Lines and Prophylactic Use

PRODUCT INFORMATION

Product name	DePlasma™
Product number	D022-125mg
Package quantity	DePlasma™ : 25mg/vial X 5
Appearance	Beige powder
Storage temperature	Powder: 4°C
(Keep container tightly sealed)	Solution: 4°C
Shelf life	Powder: up to 1 year
	Solution: up to 3 months

GENERAL APPLICATION

DePlasma™ is designed to eliminate *Mycoplasma* contamination from cultured cell lines. It can also be used prophylactically to protect culture cells from future *Mycoplasma* contamination.

DESCRIPTION

DePlasma[™] is an antibiotic cocktail comprised of one macrolide, one quinolone, and one tetracycline compound in a single vial. This three compound mixture offers lower cytotoxicity and higher potency when compared with other products on the market. It is effective against all known *Mycoplasma* species and related non cell-walled prokaryotes.

MODE OF ACTION

The modes of action for each of the antibiotics in **DePlasma™** are different. Tetracyclines and macrolides block protein synthesis by interfering with translation at the prokaryotic ribosome. Quinolones inhibit bacterial DNA gyrase and topoisomerase IV, blocking DNA replication. Eukaryotic cells do not contain DNA gyrase or topoisomerase IV and are therefore unaffected by **DePlasma™** treatment.

METHOD

DePlasma[™] can be added directly to the bottle of culture medium or to the flask containing the cells. It is recommended that a 1000x stock solution (or 5 mg/ml) be made of **DePlasma[™]**. To make the stock solution, add 1.0 ml water to one 25 mg vial, shake it, and transfer to a >10 ml container and then add another 4.0 ml water to make a final 5 ml stock solution of 5.0 mg/ml. The solution can be stored at 4°C for up to 3 months.

Working concentration of DePlasma^m: 5 μ g/ml.

Decontamination period: 4 passages (about 2 to 3 weeks depending on the growth of cells).

Concentration for maintenance of Mycoplasma free culture: 2.5 μ g/ml.

Caution

In order to maximize the effectiveness of **DePlasma™**, you should:

- Increase the FBS concentration 10% higher to enhance the cell growth.
- Mix the fresh cultures thoroughly with DePlasma[™].
- Break the clumps and clusters of the cell lines.
- Keep cells' densities 5% to 10% higher than usual.
- Observe the cells under inverted microscope daily.

Protocol – Primary cell lines

- Split an actively dividing culture of cells into media containing 5 µg/ml of *DePlasma™*.
- Remove and replace with fresh *DePlasma™* containing medium (5 µg/ml) every passage for 3 passages. (For some slow-growing cells, it is recommended to replace the media every ~2-3 days).

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- Confirm the elimination of *Mycoplasma* by using TOKU-E's PCR *Mycoplasma* Detection Kit.
- If *Mycoplasma* has not been eliminated after 3 passages of treatment with *DePlasma™*, then treat with *DePlasma™* at 40 µg/ml for one additional passage.
- 5. Check for the elimination of *Mycoplasma* by using TOKU-E's **PCR** *Mycoplasma* **Detection Kit**.

For maintenance of *Mycoplasma* free cultures, use **DePlasma™** at a concentration of 2.5 μg/ml.

Protocol – Prophylactic Use

- Split an actively dividing culture of cells into media containing 2.5 µg/ml of *DePlasma™*.
- Confirm that cells grow normally in presence of DePlasma[™].
- For maintenance of *Mycoplasma* free culture, continue using *DePlasma*[™] at a concentration of 2.5 µg/ml.
- 4. Check for the elimination of *Mycoplasma* by using TOKU-E's **PCR** *Mycoplasma* **Detection Kit**.

