

## **Tips for Using UltraGRO™ to Grow Mesenchymal Stem Cells (MSCs)**

HELIOS® Bioscience Brand, AventaCell Product, UltraGRO™ shows optimal growth of MSC at 5 % (v/v) in typical cell culture media, i.e. Alpha-MEM, which contains 2 mM L-Glutamine.

We recommend seeding MSCs at approximately  $3 \times 10^3 \sim 6 \times 10^3$  cells per  $\text{cm}^2$ .

For UltraGRO™ product, Heparin at a final concentration of 2 U/ml in the culture media supplemented with 5 % UltraGRO is required. Failure to add Heparin will cause coagulation during cell culture in typical medium.

### **UltraGRO™ Storage**

UltraGRO™ product is most stable when stored frozen until needed.

The recommended storage temperature is  $-20\text{ }^\circ\text{C}$ .

Please thaw frozen UltraGRO™ product in a  $37\text{ }^\circ\text{C}$  water bath before use. Once UltraGRO™ product is thawed, it is recommended to use it for completed medium preparation (e.g. 5 %) immediately, or to divide it into single-use aliquots and store unused aliquots at  $-20\text{ }^\circ\text{C}$ .

### **Precipitation in Cell Culture**

Clotting or insoluble particles may form in thawed UltraGRO™, it is recommended to centrifuge at  $3,400 \times g$  for 3 ~ 5 minutes or to filter the liquid concentrate with a sterile  $40\text{ }\mu\text{m}$  Cell Strainer to remove insoluble particles.

Filtering the completed medium (e.g. 5%), after UltraGRO™ is diluted in the basal medium, will not affect the cell culture performance. However,  $0.22\text{ }\mu\text{m}$  filtering is **NOT** recommended for the 100 % UltraGRO™ concentrate, as this may reduce 5% UltraGRO™ cell culture performance.

Repeated freeze-thaw cycles should be avoided as they will cause an increase in insoluble particles and potential decrease in UltraGRO™ performance.