OriCell Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Medium

Catalog No. MKCMA-90031

Product Description:

Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Medium consists of optimized Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Basal Medium, pre-selected Fetal Bovine Serum and supplements. This product has been developed for the optimal differentiation of Cynomologus Monkey Mesenchymal Stem Cells (Cat. No. MKCMA-01001) into adipocytes.

The product is intended for laboratory research use only, not for drug, house hold, or other uses.

Kit Components:

Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Medium A:

(Cat. No. MKCMA-03031-200)

Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Basal Medium A (Cat. No. MKCMA-03031-175)	175 mL
Mesenchymal Stem Cell-Qualified Fetal Bovine Serum (Cat. No. MKCMA-05001-20)	20 mL
Penicillin-Streptomycin	2 mL
Glutamine	2 mL
Insulin	400 µL
IBMX	200 µL
Rosiglitazone	200 µL
Dexamethasone	200 µL

Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Medium B:

(Cat. No. MKCMA-03032-200)

Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Basal Medium B	175 mL
(Cat. No. MKCMA-03032-175)	
Mesenchymal Stem Cell-Qualified Fetal Bovine Serum	20 mL
(Cat. No. MKCMA-05001-20)	
Penicillin-Streptomycin	2 mL
Glutamine	2 mL
Insulin	400 µL

Instructions:

Preparation of Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Medium A (induction medium)

1. Prior to use, thaw Mesenchymal Stem Cell-Qualified Fetal Bovine Serum at 2 to 8°C under refrigeration until thawed. Gently swirl the bottle to ensure homogeneity. Mesenchymal Stem Cell-Qualified Fetal Bovine Serum has been heat-inactivated and is ready to use after thawing.

Note: The thawed serum may contain some flocculent precipitates. The presence of these substances in serum does not alter the performance characteristics of the product. It is not recommended to filter the serum to remove these precipitates. Doing so may result in the loss of some serum nutrients.

2. About 30 minutes prior to use, thaw Dexamethasone, Insulin, IBMX, Rosiglitazone, Penicillin-Streptomycin solution and Glutamine solution at room temperature. Gently invert the vials to ensure homogeneity.

Note: Centrifuge the vials briefly at low speed before removing the caps to ensure recovery of entire content.

3. Disinfect with 70% v/v ethanol the external surfaces of the bottles/vials for every component in the kit. Allow ethanol to evaporate away.

4. In a laminar flow hood aseptically open the bottles/vials.

5. Transfer the entire amount of Mesenchymal Stem Cell-Qualified Fetal Bovine Serum, Penicillin-Streptomycin solution and Glutamine solution into Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Basal Medium A.
6. Rinse each vial with the medium and transfer the rinse medium back to the bottle of basal medium A.
7. Transfer the entire amount of Dexamethasone, Insulin, IBMX and Rosiglitazone into Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Basal Medium A.
8. Rinse each vial with the medium and transfer the rinse medium back to the bottle of basal medium A as much as possible.
9. Repeat step 8 several times.
10. Gently swirl the fully supplemented complete medium to ensure a homogeneous mixture. The complete medium is now ready to use.

Note: Although each component in this kit is supplied sterile, it is strongly recommended to filter the fully supplemented complete medium.

Preparation of Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Medium B (maintenance medium)

1. Prior to use, thaw Mesenchymal Stem Cell-Qualified Fetal Bovine Serum at 2 to 8°C under refrigeration until thawed. Gently swirl the bottle to ensure homogeneity. Mesenchymal Stem Cell-Qualified Fetal Bovine Serum has been heat-inactivated and is ready to use after thawing.

Note: The thawed serum may contain some flocculent precipitates. The presence of these substances in serum does not alter the performance characteristics of the product. It is not recommended to filter the serum to remove these precipitates. Doing so may result in the loss of some serum nutrients.

2. About 30 minutes prior to use, thaw Insulin, Penicillin-Streptomycin solution and Glutamine solution at room temperature. Gently invert the vials to ensure homogeneity.

Note: Centrifuge the vials briefly at low speed before removing the caps to ensure recovery of entire content.

3. Disinfect with 70% v/v ethanol the external surfaces of the bottles/vials for every component in the kit. Allow ethanol to evaporate away.
4. In a laminar flow hood aseptically open the bottles/vials.
5. Transfer the entire amount of Mesenchymal Stem Cell-Qualified Fetal Bovine Serum, Penicillin-Streptomycin solution and Glutamine solution into Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Basal Medium B.

6. Rinse each vial with the medium and transfer the rinse medium back to the bottle of basal medium B.
7. Transfer the entire amount of Insulin into Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Basal Medium B.
8. Rinse the vial with the medium and transfer the rinse medium back to the bottle of basal medium B as much as possible.
9. Repeat step 8 several times.
10. Gently swirl the fully supplemented complete medium to ensure a homogeneous mixture. The complete medium is now ready to use.

Note: Although each component in this kit is supplied sterile, it is strongly recommended to filter the fully supplemented complete medium.

Adipogenesis Protocol (for 6-well tissue culture plate)**:**

1. Cynomologus Monkey Mesenchymal Stem Cells are cultured in Cynomologus Monkey Mesenchymal Stem Cell Growth Medium (Cat. No. MKCMA-90011) (growth medium thereafter) at 37°C in a 5% CO_2 humidified incubator.
2. When cells are approximately 80-90% confluent, they can be dissociated with Trypsin-EDTA (Cat. No. TEDTA-10001-100).
3. Cynomologus Monkey Mesenchymal Stem Cells are replated in growth medium at $2x10^4$ cells/cm^2 in 6-well tissue culture plates with a medium volume of 2 mL per well.
4. Incubate the cells at 37°C in a 5% CO_2 humidified incubator.
5. Refeed the cells every 3 days until they are 100% confluent or post confluent. It's strongly suggested to induce the adipogenic differentiation of Cynomologus Monkey Mesenchymal Stem Cells 3-5 day post confluent.
6. When the cells reach 100% confluent or post confluent, carefully aspirate off the spent growth medium from the wells and add 2 mL induction medium per well.
7. Three days later, change the medium to maintenance medium by completely replacing the spent induction medium.
8. 24 hours later, change the medium back to induction medium.
9. To optimally differentiate Cynomologus Monkey Mesenchymal Stem Cells into adipogenic cells, repeat the cycle of induction/maintenance three times.
10. After three to five cycle of induction/maintenance, culture the cells in maintenance medium for additional 7 days by replacing the medium every 3 days.

Oil red O Stain Analysis

1. After differentiating, remove Adipogenic Differentiation Medium from well and rinse with 1×PBS. Fix cells with 2ml of 4% formaldehyde solution for 30 minutes.
2. Then rinse wells twice with 1×PBS and stain cells with 1ml oil red O working solution(3:2 dilution with distilled water and Filter with filter paper) for 30 minutes.
3. Rinse wells 2-3 times with 1×PBS, then visualize under light microscope and capture images.

Stability/Storage:

All products should be stored in the dark.

Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Basal Medium A&B is stable at 2 to 8°C for up to one year. Other components are stable at -20°C for up to two years. These products should be discarded beyond the labeled expiration date.

Once prepared, the fully supplemented complete medium can be stored for up to one month when stored in the dark at 2 to 8°C.

For optimal performance, repeated warming/cooling and freeze-thawing should be avoided.

Quality Control:

Cynomologus Monkey Mesenchymal Stem Cell Adipogenic Differentiation Medium is performance tested on Cynomologus Monkey Mesenchymal Stem Cells.

Standard evaluation includes:

1. Sterility test (bacteria, fungi, mold and mycoplasma)
2. pH test
3. Osmolality
4. Endotoxin