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# **Product Specification**

Product name	Lolina® Human AD-MSC heal-exo® enhancer kit 1, Xeno Free, Exo Plus		
Cat.No.	NaC20140304		
Storage and	Store at -70 °C. Once added to medium, store at 4°C, do not refreeze after thawing.		
shipping	Dry ice transportation.		

# **Product Description**

Lolina® Human AD-MSC heal-exo® enhancer kit 1 is a set of sterile powders or concentrated solution which contains growth factors, hormones, or proteins for significant induction of paracrine secretion.

As an treatments additive for AD-MSC in vitro culture, this supplement has been proved has following functions:

- 1. It increased EV production more than fivefold and cargo protein levels in EV more than twofold compared to levels in MSC alone.
- 2. The contents enhanced proliferation, migration, and tube formation of human umbilical vein endothelial cells in vitro and improved skin wound healing in vivo.
- 3. It was neither cytotoxic nor altered the properties of MSCs, including their surface marker profile and in vitro chondrogenesis, adipogenesis, and osteogenesis.
- 4. It promoted the stability of MSCs and also helped stabilize the exosome membrane.

# **Components**

Compound No.	Compounds	Format	Size
NaC20140304-A	r- human Thrombin	Lyophilized	1000 units/mg, 200 μg
NaC20140304-B	r-Human serum albumi (rHSA)	Lyophilized	62.5 mg

# **Instructions for Use**

# 1. Stock solution Preparation.

One kit is for 50ml cell culture medium.

The compounds are offered as lyophilized powder, their stock solutions are prepared as follows:

- a. The compounds are offered as powder/liquid in tubes. Please centrifuge before opening the cap to ensure the accuracy of the dosage.
- b. Please carry out dissolution and packaging operations on a clean bench.
- c. Spray the medium bottle and supplement tube with 70% ethanol and wipe to remove excess liquid. In a sterile field, remove the caps without touching the interior threads with fingers.
- d. Reconstitute NaC20140304-A in 500 μl sterile 1 ×PBS. Aliquot into appropriate volumes of storage solution. Aliquot into appropriate volumes of storage solution. When stored at -70°C, the stock solution is stable for 6 month. When stored at 4 °C, the stock solution is stable for 1 week.
- e. Reconstitute NaC20140304-B in sterile 0.9% NaCl solution. The recommend volume is 5 mL. Aliquot into appropriate volumes of storage solution. When stored at 2-8 °C, -20°C, the stock solution is stable for 24 month.

#### 2. Protocol

# **Step 1: AD-MSC Culture**

**Seeding:** Seed AD-MSC in culture flasks or plates at a density allowing them to reach 70-80% confluence.

**Growth:** Allow AD-MSC to grow until they reach the desired confluence.

#### Step 2: Treatment AD-MSC with NaC20140304

#### **Prepare Treatment Medium:**

- a. Thaw the stock solution.
- b. Add the stock solution of NaC20140304 to the regular culture medium to obtain a treatment medium. The dilution ratio is: NaC20140304-A 1:100; NaC20120303-C 1:10.

#### **Treat AD-MSCs:**

- c. Replace the regular culture medium with the treatment medium.
- d. Incubate the AD-MSCs with the treatment medium for 24 hours under standard culture conditions (37 °C, 5% CO2).

#### **Step 3: Post-Treatment Handling**

**Remove Treatment Medium:** After 24 hours of treatment, remove the treatment medium.

Wash MSCs: Wash the cells gently with PBS to remove any residual the treatment medium.

### **Conditioning Phase:**

- a. Replace with fresh, serum-free, or exosome-depleted medium.
- b. Incubate the AD-MSCs for an additional 24-48 hours to collect the conditioned medium containing exosomes.

# Step 4: Exosome Isolation and Purification

Collect the conditioned medium after the post-treatment incubation period.

# Note

If handled improperly, some components of the medium may present a health hazard. Take appropriate precautions when handling it, including the wearing of protective clothing and eyewear. Dispose of properly.