



## Product Specification

<b>Product name</b>	Lolina® Human AD-MSC Cool-Exo® Enhancer kit 1 (500×), Xeno Free, Exo Plus
<b>Cat.No.</b>	NaC20140301
<b>Size</b>	100 µL/1mL/5ml
<b>Storage and shipping</b>	Store at -20 °C. Once added to medium, store at 4 °C, do not refreeze after thawing. Dry ice transportation.

## Product Description

Lolina® Human AD-MSC Cool-Exo® Enhancer kit 1 (500×) is a sterile, concentrated (500X) solution which contains growth factors, hormones, or proteins for directed induction of paracrine secretion. The supplement is formulated to provide an optimally balanced environment that selectively enhance the production of exosomes with anti-inflammatory properties and anti-inflammatory related miRNAs.

Preconditioning AD-MSCs with this kit aims to activate the cells' immune response pathways, leading to the secretion of exosomes with enhanced anti-inflammatory and regenerative properties. These preconditioned exosomes can be more effective in clinical applications, such as in the treatment of inflammatory diseases and tissue repair.

## Components

<b>Compound No.</b>	<b>Size</b>	<b>Compounds</b>
NaC20140301-A	100 µL/1mL/5ml	Lipopolysaccharide (LPS)
NaC20140301-B	100 µL/1mL/5ml	IL-1β

## Instructions for Use

### 1. Stock solution Preparation.

Thaw all the two compounds at 37°C. Gently tilt the tube several times to ensure complete mixing. (Note: the growth supplement may have lipoprotein precipitates; this does not affect the efficacy of

the supplement). 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. Divide the NaC2012030 into the required volumes, **do not** mix them. The stock solution should be stored at -20 °C. When stored at -20°C, the stock solution is stable for 6 months.

## 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: Lolina® Human AD-MSC Cool-Exo® Enhancer kit 1 Treatment

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

Add the stock solution of Lolina® Human AD-MSC Cool-Exo® Enhancer kit 1 to the culture medium to a final concentration. The dilution ratio is 1:500.

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

Replace the regular culture medium with the treatment medium.

Incubate the AD-MSCs with the treatment medium for 24 hours under standard culture conditions (37 °C, 5% CO<sub>2</sub>).

### Step 3: Post-Treatment Handling

**Remove Treatment Medium:** After 24 hours of treatment, remove the medium containing LPS and IL-1 $\beta$ .

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

#### **Conditioning Phase:**

Replace with fresh, serum-free, or exosome-depleted medium.

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.