ACExtract Exosome Isolation Reagent(from medium)

Cat# CE1001 – 10 ml / CE1002 – 50 ml

Storage at 4℃

INTRODUCTION

ACExtract Exosome Isolation Reagent(from medium) is designed for extract exosome from cell medium and can be further applied for downstream experiments, such as RNA Sequencing, High-Throughput Screening and cell co-culture et al. Compared with traditional ultracentrifuge, this reagent provides shorter operation time, lower sample input, higher extraction efficacy and intact pattern of exosome.

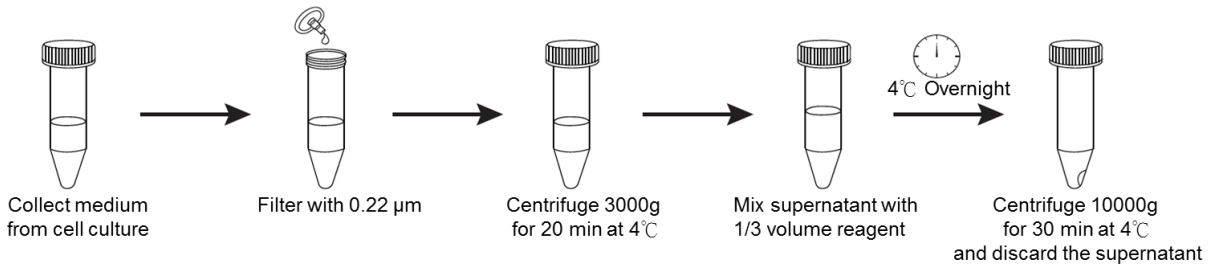
CONTENTS

No	Component	CE1001 – 10 ml	CE1002 – 50 ml
AA	ACExtract Exosome Isolation Reagent	10 ml	10ml x 5

QUALITY CONTROL

Extract exosome from 6 ml supernatant of 48hr culture HeLa cell line and then using qPCR to examine small RNA expression from exosome-extract RNA.

PROTOCOL



1. Collect medium from cell culture

   1.A For adherent cell :

   When cell is about 50-70% confluent, replace the FBS-contained medium with serum-free or exosome-depleted FBS medium. Collect the supernatant for downstream extraction when cell density reaches to 80-95% confluent.

   1.B For suspend cell :

   When cell is about 50-70% confluent, collect the cell after centrifugation at 300 xg, 4℃ for 10 min and replace the FBS-contained medium with serum-free or exosome-depleted FBS medium. Collect the supernatant after centrifugation at 300 xg, 4℃ for 10 min for downstream extraction when cell density reaches to 80-95% confluent.

2. Remove cell and debris by using 0.22um filter and transfer filtered medium into new centrifuge tube.

3. To complete removal of cell debris, centrifuge the filtered medium at 3000 xg, 4℃ for 20 min and gently collect the medium without disturbing the pellet.
4. Add 1/3 volume of **ACExtract Exosome Isolation Reagent** to cell-free medium.
   For instance:

cell-free medium	Reagent
1 ml	0.33 ml
9 ml	3 ml

5. Mix the medium/reagent mixture well by gently invert the tube till the mixture become clarification.
6. Place the tube in vertical direction and keep steady at 4℃ overnight.
7. On second day, centrifuge at 10000 xg, 4℃ for 30 min and discard the supernatant, the exosome is contained in pellet at the bottom of the tube.

*The exosome may be invisible. We recommend to use swinging bucket rotor and if using fixed angle rotor, it should be label the direction of tube placement.

8. To removal of residue supernatant, centrifuge at 1500 xg, 4℃ for 2min.
9. Suspend the exosome in a proper 1x PBS.

*Store isolated exosome at 4℃ for 1 week or -20/-70℃ for long-term storage.

PRODUCT USE LIMITATION

These products are intended for research use only.