

# Precise and Efficient EVs Purity Detection with Exoplorer™

Extracellular Vesicles (EVs) play a vital role in assessing EV quality, ensuring the reliability and validity of research findings and therapeutic applications. This significance highlights the critical need for accurate exosome purity detection.

The Exoplorer™ Nano-flow Cytometer, a cutting-edge instrument for nanoparticle-level analysis, holds great promise for precise measurement of EV purity. Comparative tests were conducted between the Exoplorer™ and NAT using two methods: one involves calculating the particles/protein ratio as recommended by the International Society for Extracellular Vesicles, and the other measures the membrane ratio, which is also widely recognized as a valid approach.

# Measure EVs Purity By Calculating Particles/Protein Ratio with the Exoplorer<sup>™</sup> and NTA

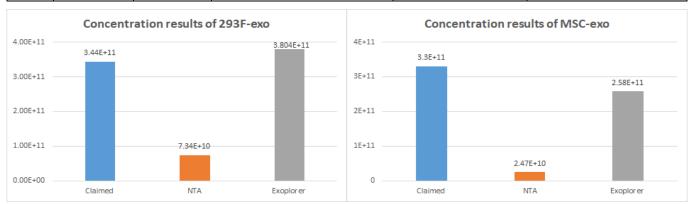
EVs purity can be detected by the calculation of particle/protein ratio based on the following formula:

$$particles/mg = \frac{Particles/ml}{m\,g/m\,l}$$

Among them, particles/ml refers to the EVs concentration and mg/ml is the protein concentration in the EVs sample. EVs concentration is tested by the Exoplorer<sup>™</sup> and NTA, and also the particle/protein ratio is calculated using the results from the two instruments as comparison, while the protein concentration is counted using the Bicinchoninic Acid Assay.

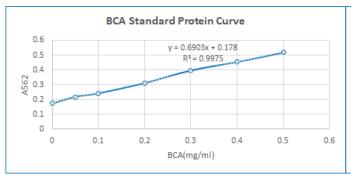
## Comparative Concentration Results between the Exoplorer and NTA

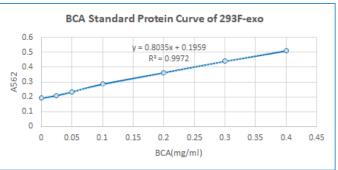
| No.  | Sample<br>Name | Company | Claimed Data by Nanofcm<br>(Particle/mL) | Data by NTA<br>(Particle/mL) | Data by Exoplorer<br>(Particle/mL) |
|------|----------------|---------|--|------------------------------|------------------------------------|
| 1    |                |         |  | 7.83E+10                     | 3.132E+11                          |
| 2    | 293F-exo       | Echo    | 3.44E+11                                 | 8.34E+10                     | 3.708E+11                          |
| 3    | 293F-exo       | Biotech | 3.44E+11                                 | 5.84E+10                     | 4.572E+11                          |
| Mean |                |         |  | 7.34E+10                     | 3.80E+11                           |
| 1    |                |         |  | 3.01E+10                     | 2.466E+11                          |
| 2    | MSC-exo        | Echo    | 3.3E+11                                  | 2.52E+10                     | 2.448E+11                          |
| 3    | ivisc-exo      | Biotech | 3.35+11                                  | 1.88E+10                     | 2.826E+11                          |
| Mean |                |         |  | 2.47E+10                     | 2.58E+11                           |





#### **Protein Concentration Results using Bicinchoninic Acid Assay**

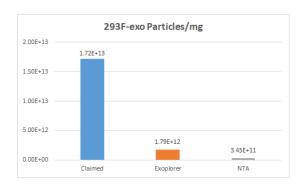




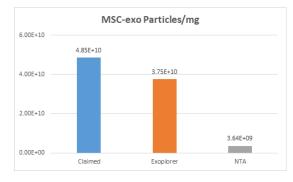
| Sample   | Test Item       | Claimed Data | 1         | 2        | 3        | Mean |
|----------|-----------------|--------------|-----------|----------|----------|------|
| MSC-exo  | Protein (mg/mL) | 6.8          | 6.2581486 | 7.301173 | 6.153846 | 6.57 |
| 293F-exo | Protein (mg/mL) | 0.02         | 0.1647791 | 0.194648 | 0.179714 | 0.18 |

### Comparative Particles/Protein Ratio between the Exoplorer and NTA

| Sample   | Instrument | Test Item    | Claimed Data | Mean     |  |
|----------|------------|--------------|--------------|----------|--|
|          |            | Protein      | 0.02         | 0.21     |  |
|          | Exoplorer  | Particle     | 3.44E+11     | 3.80E+11 |  |
| 2025 040 |            | Particles/mg | 1.72E+13     | 1.79E+12 |  |
| 293F-exo |            | Protein      | 0.02         | 0.21     |  |
|          | NTA        | Particle     | 3.44E+11     | 7.83E+10 |  |
|          |            | Particles/mg | 1.72E+13     | 3.68E+11 |  |



| Sample    | Instrument | Test Item    | <b>Claimed Data</b> | Mean     |  |
|-----------|------------|--------------|---------------------|----------|--|
|           |            | Protein      | 6.8                 | 6.93     |  |
|           | Exoplorer  | Particle     | 3.30E+11            | 2.58E+11 |  |
| MSC-exo   |            | Particles/mg | 4.85E+10            | 3.75E+10 |  |
| ivisc-exo |            | Protein      | 6.8                 | 7.01     |  |
|           | NTA        | Particle     | 3.30E+11            | 3.01E+10 |  |
|           |            | Particles/mg | 4.85E+10            | 4.29E+09 |  |



#### Note:

- 1. Of the two samples, 292F-exo is a high-purity sample and the MSC-exo sample purity is much lower than it according to their manufactures Echo Biotech.
- 2. The protein concentration of 293F-exo from Echo Biotech, as tested with the BCA Protein Assay Kit from Beyotime, was approximately 10 times the claimed value. This discrepancy may be due to the presence of trehalose in the 293F-exo, which could have affected the concentration obtained.



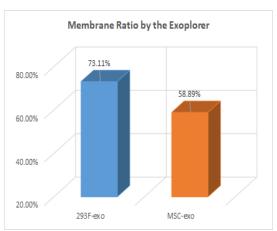
# Measure EV Purity Results by Calculating the Membrane Ratio with the Exoplorer<sup>TM</sup>

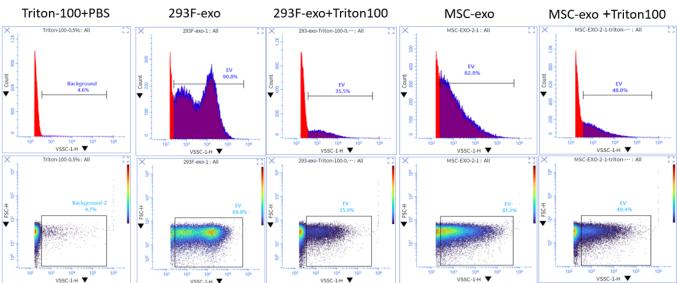
EVs purity can also be measured by the calculation of membrane ratio based on the following formula:

$$\label{eq:membrane} \textit{Membrane ratio} = \frac{(\text{concentration before lysis} - \text{background concentration}) - \text{concentration after lysis}}{\textit{Concentration before lysis} - \textit{background concentration}}$$

Among them, the concentration before lysis represents the count of EVs sample, background concentration refers to the count of Triton-100+PBS sample and concentration after lysis is the count of Triton-100+EV sample.

| Commis   | Triton-100 | EV    | Triton-100 | Membrane Ratio |
|----------|------------|-------|------------|----------------|
| Sample   | +PBS       |       | +EV        |                |
| 293F-exo | 710        | 38607 | 10901      | 73.11%         |
| MSC-exo  | 710        | 35220 | 14897      | 58.89%         |





**Note:** Of the two samples, 292F-exo is a high-purity sample and the MSC-exo sample purity is much lower than it according to their manufactures Echo Biotech.

## **Summary**

The experimental data presented above clearly demonstrate that the Exoplorer<sup>TM</sup> Nano-flow Cytometer is highly capable of serving as an effective tool for measuring EVs purity. Its precision and reliability make it an invaluable asset in the field of EVs research. By providing accurate measurements, this instrument can significantly contribute to the quality control of EVs, ensuring that research findings and therapeutic applications are based on high-quality materials.