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PARTICLE SIZE

PARTICLE CONCENTRATION

FLUORESCENCE DETECTION

NANOSIGHT
NANOPARTICLE TRACKING ANALYSIS
FOR EXOSOME RESEARCH

NANOSIGHT
NANOPARTICLE TRACKING ANALYSIS

Providing data at all steps of exosome characterization, from improving isolation methods, to tracking sample stability and purity, through to identifying positive subsets using specific fluorescent labeling.

Over recent years, interest in exosomes and other vesicles has grown dramatically. To drive cutting-edge research, you need easy-to-use, robust tools which will generate reliable data. The NanoSight range of instruments provides high resolution particle size and concentration data for early stage researchers, as well as those looking to translate research to clinical settings. NanoSight Nanoparticle Tracking Analysis (NTA) provides size, concentration and polydispersity data in minutes (Figure 1).

The NanoSight NTA software suite requires minimal user input to obtain these data, and allows methods to be saved and easily re-used. This allows for samples to be monitored over time where the only variable is any intrinsic difference in the sample (see Figure 2, which shows the effect of storage at 4°C compared to room temperature on the size and concentration of plasma exosomes).

The selected method of exosome isolation may impact both the size and concentration of the exosomes that are prepared. NanoSight provides easy overlay of data from different experiments, as shown in Figure 3.
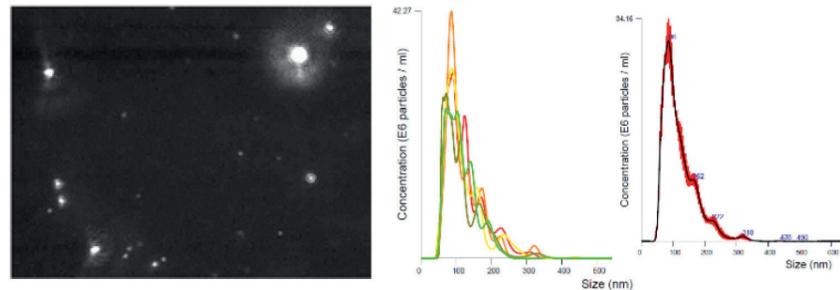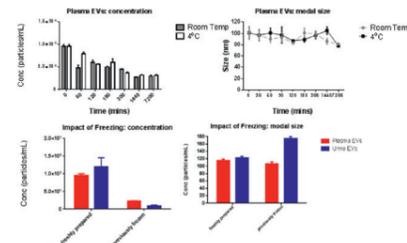

Figure 1: Image of light scattered by exosomes viewed on NanoSight NS300 (left hand panel). Size vs Concentration graphs of individual measurements (center panel) and merged average data +/- SEM (right hand panel)

Figure 2: Top panels: Time v Concentration and Time V Size graphs for plasma exosomes stored at either 4°C or at room temperature. Lower panels: Impact of freezer storage on exosome size and concentration, for both plasma-derived and urine-derived exosomes.

Figure 3: Comparison of size, concentration and polydispersity of exosomes when isolated by different methods
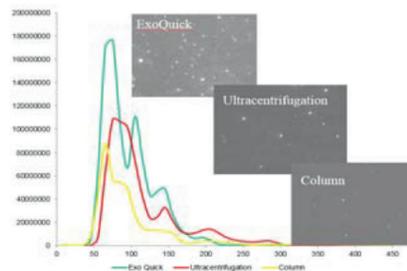
Key benefits

- Size, concentration and polydispersity data…in minutes
- Easy to use NTA software suite
- SOP-driven methods for better data comparisons

- Discriminate between exosomes and contaminants with membrane dyes in fluorescence mode
- Characterize fluorescently-labeled exosome subsets after direct or indirect labeling

- Monitor sample purity
- Monitor sample stability

All the previous examples were generated using NTA in traditional Light Scatter mode. However, NTA can also give size, concentration and polydispersity data in Fluorescence Mode when used with suitable fluorescently-labeled samples and with an appropriate filter inserted into the optical path. Using this approach, NTA assures users of the identity of particles as exosomes rather than contaminants (Figure 4).

When used to identify specific markers on the exosome surface, NanoSight can be used to speciate subsets of exosomes by directly labeling with fluorescently-tagged antibodies, or by using an indirect 2-step approach. Exosomes containing the tetraspanins CD63 and CD9 have been identified using these methods (Figure 5).
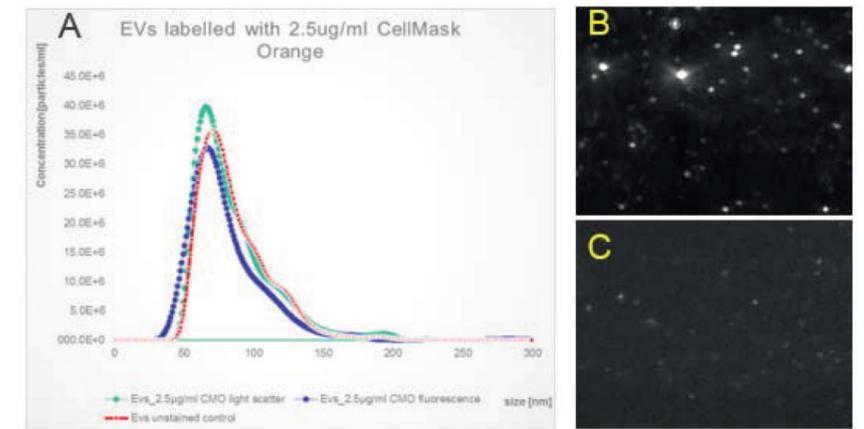

Figure 4: A. Comparison of labelled and nonlabelled exosomes in scatter and fluorescent modes (membrane labelling dye) with NanoSight images of the labelled sample viewed in B, light scatter mode and C, fluorescent mode.

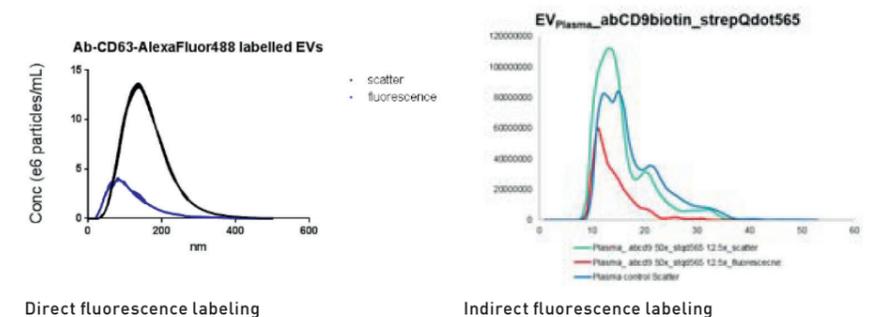Direct fluorescence labeling   Indirect fluorescence labeling

Figure 5: (Left) Exosomes labeled with anti- CD63-Alexa488™ measured by NanoSight NTA in fluorescence and light scatter mode; (Right) Exosomes labeled with anti-CD9 biotinylated primary antibody and streptavidin-conjugated Quantum dot (QDOT565) measured by NanoSight NTA in fluorescence and light scatter mode
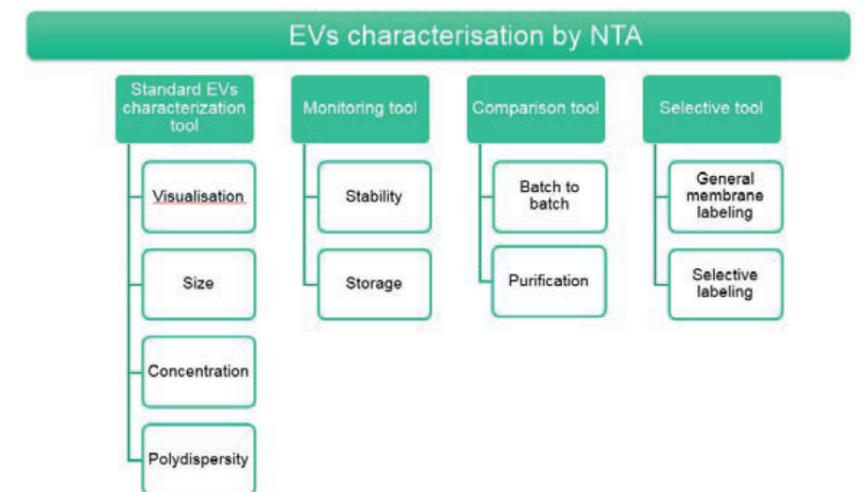
NanoSight NTA, your go-to instrument for exosomes research