

Supplementary Materials

Which casein micelle removal method is suitable for studies of human milk extracellular vesicles? A systematic comparison of four different treatments for casein depletion before extracellular vesicle isolation from human milk

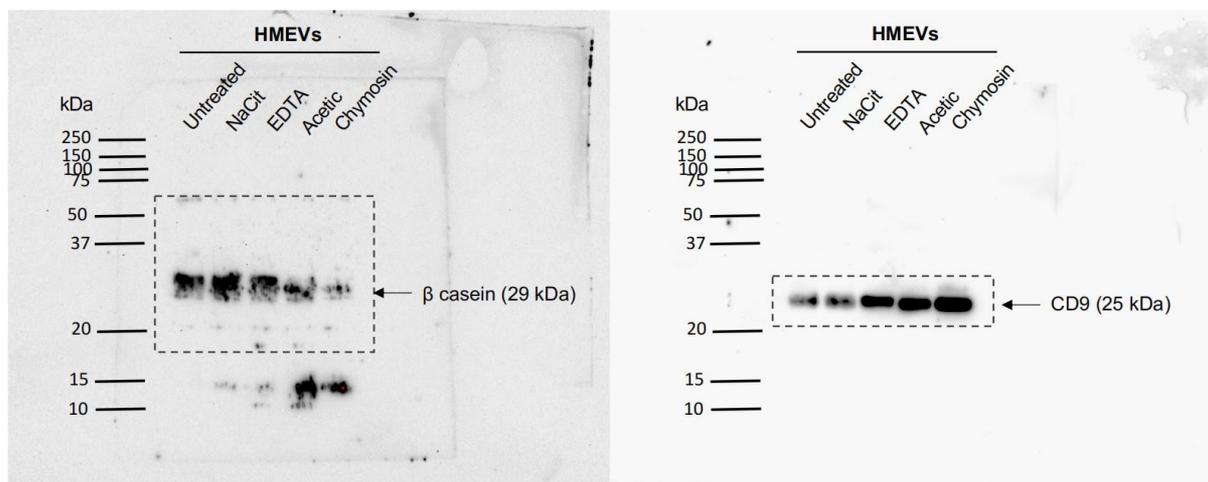
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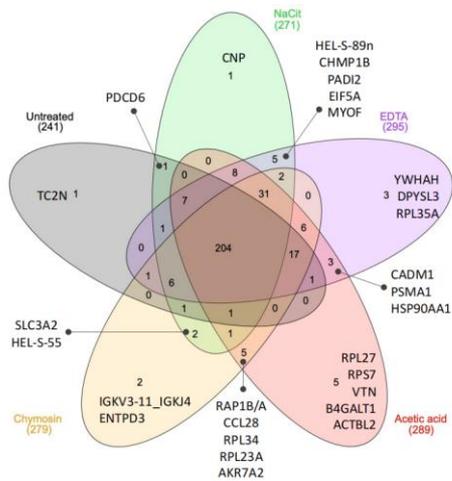
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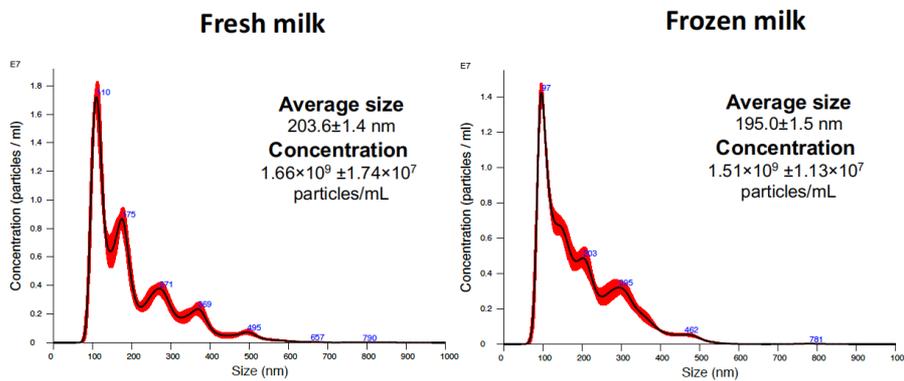


Supplementary Figure 1. Venn diagram illustrated the number of shared and unique HMEV proteins among different casein micelle removal methods (full details of protein names provided in Supplementary Table 3).



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 26 **Supplementary Figure 2.** The full-length immunoblot results of the cropped images (dash
 27 line), as presented in Figure 4C.

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 30 **Supplementary Figure 3.** Nanoparticle Tracking Analysis of HMEVs from Fresh and
 31 Frozen Milk Samples. NTA shows the size distribution, average diameter, and concentration
 32 (particles/mL) of human milk extracellular vesicles (HMEVs) isolated from fresh and frozen
 33 milk samples, stored at -80°C for 18 hours to make sure that it is frozen, obtained from the
 34 same donor.

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36 **Supplementary Table 1.** Full details of 393 HMEV proteins identified by DDA and
 37 BoxCAR proteomics with MaxQuant search.

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39 **Supplementary Table 2.** Eighty-seven proteins consistently identified by DDA and
 40 BoxCAR proteomics

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42 **Supplementary Table 3.** Lists of shared and unique proteins identified according to the
 43 Venn diagram as shown in Supplementary Figure 2.