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Quality Over Quantity

Why Particle Count Is the Wrong Metric

Paper 05 in the CFT Advantage Series

W H I T E P A P E R

Wellbeing International Foundation • Hamilton, Bermuda

Abstract

Commercial exosome companies frequently market their products by emphasizing raw particle counts, 'trillions of exosomes per dose', as evidence of potency. This metric, while superficially compelling, is potentially overstated and obscures the factors that actually determine therapeutic efficacy. This paper examines why particle count fails as a meaningful measure of biological activity. We explore how allogeneic companies achieve inflated counts through industrial cell expansion and pooled donors, the biological context of the body's endogenous extracellular vesicle (EV) load, the critical role of immunological matching in cellular uptake, and the rapid immune clearance of mismatched allogeneic preparations. We contrast this with autologous approaches, which deliver smaller particle volumes of potentially greater biological relevance, including the full active secretome rather than isolated exosomes. The paper concludes that physicians and patients should prioritize composition, specificity, and biological potency over headlines about particle volume.

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1. Introduction: The Particle Count Arms Race

A troubling pattern has emerged in the commercial exosome market: companies compete primarily on a single, easily marketed metric, particle count per dose. Marketing materials boast of ‘trillions of exosomes,’ ‘the highest particle concentrations available,’ and ‘maximised particle yield.’ This messaging appeals to both physicians and patients: more particles sound better; if one exosome is good, surely a trillion must be better.

This logic fails on multiple biological and physical grounds. Raw particle volume is not a measure of therapeutic efficacy. It is not a proxy for quality, biological activity, or clinical benefit. Paradoxically, it may not reflect biological potency, and obscures the factors that actually determine whether a preparation will support biological function: the composition of the particles, the immunological compatibility of the preparation with the recipient, the half-life and biodistribution of the particles in vivo, and the functional relevance of the cargo they carry.

This paper examines the particle count claim from first principles: how allogeneic companies achieve such high counts, why the human body already contains a vast endogenous EV load that makes exogenous particle volume increasingly irrelevant, how immunological mismatch leads to rapid clearance of allogeneic preparations, and why autologous approaches, delivering far fewer total particles but with inherent immunological self-matching and full secretome cargo may offer superior therapeutic support.

2. How Commercial Exosome Companies Achieve High Particle Counts

2a. Industrial Cell Expansion and Phenotypic Drift

To generate ‘trillions’ of exosomes, allogeneic companies must culture vast numbers of cells. They typically expand primary human mesenchymal stem cells (hMSCs) or other donor cells through multiple passages, often 8–15 passages or more, to achieve the cell densities required for bulk exosome collection.

This extended expansion comes with a cost: cellular senescence and phenotypic drift. With each passage, cells accumulate stress, alter their gene expression profile, and gradually lose the molecular signatures that characterize fresh, minimally manipulated cells. Telomere shortening, epigenetic changes, and metabolic reprogramming occur progressively. The exosomes secreted by passage-12 cells are fundamentally different from those secreted by passage-2 cells from the same donor, they carry different mRNA cargo, display altered surface antigen patterns, and exhibit diminished biological potency.

There is a further mechanical cost to producing exosomes at this scale. Harvesting trillions of exosomes from large-volume cell cultures requires multiple rounds of high-speed ultracentrifugation, typically at forces in the region of 100,000×g. Even with differential centrifugation protocols, repeated spinning at these extreme forces places enormous shear stress on the vesicles themselves. Exosomes are delicate lipid-bilayer structures, typically just 30–150 nm in diameter, and they are inherently fragile under mechanical loading. The result is that a significant proportion of vesicles are ruptured, deformed, or stripped of their surface proteins during the isolation process. To preserve what remains intact, commercial preparations must be snap-frozen and maintained in cold-chain storage, because at room temperature, these mechanically stressed vesicles degrade rapidly.

CFT takes a fundamentally gentler approach. Because the goal is to concentrate the full secretome rather than isolate purified exosomes, the processing avoids repeated ultracentrifugation entirely. The preparation steps are designed to preserve the biological integrity of all secreted components, vesicles, growth factors, cytokines, and signalling molecules, in their native state. This is one reason why CFT preparations remain resilient and biologically active under standard storage conditions, without the extreme cold-chain requirements that industrial exosome products depend on. The vesicles and other bioactive components have not been subjected to the mechanical trauma that industrial exosome isolation inflicts.

Yet the marketing count treats all particles equally: a particle from passage-15 stressed cells is numbered the same as a particle from fresh cells. Quantity has displaced quality in the accounting.

2b. Pooled Donor Material

To further inflate particle yields and reduce batch-to-batch variability (a legitimate operational challenge), many commercial producers pool exosomes from multiple donors, multiple different individuals, into a single product. This achieves volume targets and theoretically improves consistency. It also reduces the biological relevance of the preparation to any individual recipient.

Each donor carries a unique MHC profile, HLA type, and immunological fingerprint. A recipient receiving pooled material from 10 different donors is receiving EVs that their immune system recognises as foreign from not one source, but ten. This multiplies the immunogenicity burden and accelerates opsonisation and clearance, phenomena discussed in detail below.

Moreover, pooling obscures the functional relevance of individual cargo components. An mRNA or protein present in exosomes from donor A may interact with those from donor B in ways never encountered in nature, creating an artificial biological milieu.

2c. What Nanoparticle Tracking Actually Measures

The standard method for determining particle count is nanoparticle tracking analysis (NTA), which detects particles in the 30–1,000 nm range based on light scattering. NTA is sensitive and produces impressive-looking numbers. However, as noted in the MISEV2018 guidelines (Théry et al., 2018),

it also measures everything in that size range: genuine exosomes (30–150 nm), larger microvesicles (150–500 nm), apoptotic bodies, protein aggregates, lipid vesicles, and cellular debris.

A company marketing ‘trillions of exosomes’ based on NTA data is being technically imprecise at best. The vast majority of counted particles may not be intact, functional exosomes at all. They may be broken vesicles, protein aggregates, or debris generated during cell expansion and harvest. Electron microscopy studies of pooled allogeneic exosome preparations frequently reveal that comparative analyses of EV measurement methods show that NTA particle counts can differ substantially from those obtained by electron microscopy or resistive pulse sensing (van der Pol et al., 2014), suggesting that a significant fraction of NTA-counted particles may not represent morphologically intact vesicles.

The particle count may therefore significantly overstate the number of functionally intact particles in a given preparation. When accounting for this, the advantage of allogeneic ‘trillion particle’ claims shrinks considerably.

3. The Body’s Existing EV Load: Context the Marketing Ignores

The human body does not exist in an exosome vacuum. Circulating extracellular vesicles are an endogenous signalling system; the bloodstream contains approximately 10^9 – 10^{12} particles per millilitre of blood, roughly 1–10 billion particles per mL (Yanez-Mo et al., 2015; Raposo & Stoorvogel, 2013). Cells continuously secrete EVs as part of normal homeostasis.

A typical ‘trillions of exosomes’ dose might contain 10^{12} – 10^{13} total particles across a few mL of concentrate, yielding a dose of roughly 10^9 – 10^{11} particles per mL. When administered to a body whose circulatory and tissue EV load already spans 10^9 , 10^{12} particles per mL, the exogenous contribution may provide diminishing returns unless the particles are biologically relevant and persist long enough to act.

The system is not particle-starved. Adding more particles to a system already saturated with endogenous EVs exhibits diminishing returns. The key question shifts from ‘How many particles?’ to ‘Are these particles biologically relevant to the specific tissue and physiological state of this patient?’ This is a question that raw particle count cannot answer.

Furthermore, the body possesses sophisticated mechanisms for clearing exogenous, non-self particles. The liver and spleen are primed to recognize and eliminate foreign biological material. In many cases, allogeneic EVs never achieve therapeutic residence times in target tissues; they are cleared to the reticuloendothelial system and degraded within minutes to hours of administration.

4. Immunological Matching and Cellular Uptake

4a. Self-Recognition and Uptake Efficiency

Extracellular vesicles are recognised by target cells through surface markers. EVs from autologous sources, derived from the patient's own cells, display the patient's own MHC Class I, HLA antigens, and other 'self' markers on their surface. These markers are the molecular equivalent of an identification badge: cells of the recipient recognise them as originating from self tissue and internalise them with high efficiency.

This is not arbitrary. The process reflects millennia of evolutionary refinement: the immune system recognises 'self' and preferentially internalises self-derived signals while flagging foreign material for surveillance. An autologous EV benefits directly from this biology, it gains rapid, efficient uptake into target cells without triggering immune clearance pathways.

Allogeneic EVs, by contrast, display foreign MHC and HLA. Recipient immune cells recognise them as non-self. While the body does not immediately attack allogeneic EVs with the ferocity it would deploy against a transfused mismatched blood cell, the EV is marked as suspicious. Uptake efficiency by target cells decreases. Innate immune activation begins. The particle faces additional barriers to achieving its intended biological effect. It should be noted that the immunogenicity of allogeneic EVs remains an active area of investigation, with some studies reporting limited immune activation and others documenting significant clearance effects. The picture is mixed and evolving, but the theoretical advantage of immunological self-recognition in autologous preparations is well-grounded.

4b. The Cargo Relevance Question

Even if an allogeneic EV reached a target cell intact, the question of cargo relevance would persist: does this particle carry signals that the recipient's cells are primed to respond to? An mRNA or microRNA from a healthy 25-year-old donor may have profound relevance to another young, healthy individual. It may be irrelevant or even counterproductive for a 65-year-old with metabolic dysfunction or immune dysregulation.

Autologous preparations inherently solve this problem: the cargo is derived from the patient themselves. The miRNAs, growth factors, and signalling molecules in the patient's own secretome are calibrated to the patient's own physiology, age, metabolic state, and tissue phenotype. Signal-recipient mismatch is eliminated: the cargo derives from the patient's own cells and is calibrated to their own physiology. The patient's cells have evolved, through their own gene expression patterns, to recognise and respond to the signals in their own secretome.

In short: autologous sources provide cargo relevance by definition. Allogeneic preparations, regardless of particle count, must assume that donor signals are appropriate for recipient tissues. This assumption may not hold for many recipients.

5. Immune Clearance of Allogeneic EVs

5a. Opsonisation and Phagocytic Clearance

When foreign particles enter the circulation, the immune system responds predictably. Complement proteins in the plasma bind to the surface of allogeneic EVs, a process called opsonisation (Wiklander et al., 2015; Conlan et al., 2017). It should be noted that some EVs express complement regulatory proteins such as CD55 and CD59 that may partially protect them from complement-mediated lysis (Clayton et al., 2003), meaning clearance kinetics likely vary by EV source and surface composition rather than following a single predictable pattern. These complement fragments tag the particle as foreign and mark it for destruction. Circulating phagocytes (principally monocytes and neutrophils) recognise these complement tags and engulf the marked EVs.

Concurrently, circulating antibodies may also bind to the foreign surface antigens on allogeneic EVs, further accelerating immune recognition and phagocytosis. In patients with prior exposure to allogeneic cells (such as those who have received transfusions, transplants, or previous allogeneic cellular therapies), pre-existing antibodies may be present, dramatically accelerating EV clearance.

The result: allogeneic EVs show a short half-life in circulation. Published biodistribution studies of allogeneic exosome preparations document rapid accumulation in the liver and spleen (the principal organs of the reticuloendothelial system) within 5–30 minutes of intravenous administration (reviewed in Kalluri & LeBleu, 2020; Doyle & Wang, 2019). Published biodistribution data indicate that a substantial proportion of the injected dose is cleared from the bloodstream within 1–2 hours. Residence time in target tissues is correspondingly brief, which may limit functional interaction, though it should be noted that rapid hepatic uptake does not by itself prove therapeutic failure, as liver-targeted effects may still occur and dosing optimisation may extend functional exposure.

The headline particle count matters less if a substantial proportion may be cleared to hepatic and splenic macrophages before reaching their intended target.

5b. The Autologous Advantage in Biodistribution

Autologous EVs, displaying the patient's own MHC and self-antigens, may avoid the opsonisation and phagocytic clearance mechanisms more effectively. They are expected to circulate longer and achieve greater residence time in target tissues, consistent with established immunological principles, though direct comparative pharmacokinetic data in humans remain limited. A smaller volume of autologous EVs may therefore achieve greater biological effect than a larger volume of rapidly-cleared allogeneic material, though direct comparative clinical data are limited.

This phenomenon is well-established in cellular transplantation: autologous cells generally show superior engraftment and persistence compared to allogeneic cells in transplantation settings. The

same principle may apply to cell-derived products like EVs, though direct comparative data in EV therapy remain limited. The immune match is likely more important than absolute particle count.

6. CFT's Secretome: Concentration, Composition, and Biological Potency

6a. The Full Secretome, Not Purified Exosomes

Cell-Free Therapy (CFT) takes a fundamentally different approach. Rather than isolating a single particle type (exosomes) from extensively expanded, pooled donor cells, CFT concentrates the complete secretome from a patient's own blood, a 150 mL peripheral blood draw, processed in a GMP facility in Germany within a defined, validated protocol.

This secretome is not a purified exosome preparation. It is the full biological product secreted by the patient's own cells: extracellular vesicles (exosomes, microvesicles), growth factors (VEGF, bFGF, HGF, and others), cytokines (TNF- α , IL-6, IL-10), adhesion molecules, and microRNAs. These components operate synergistically in vivo. They were not designed in a laboratory; they evolved through the patient's own physiology to support tissue homeostasis and regeneration.

The concentration of CFT preparations, approximately 10^9 – 10^{10} particles per mL (within the range reported for hypoxia-conditioned secretome preparations using NTA and flow cytometry; cf. Théry et al., 2018), is lower than commercial 'trillions of exosomes' claims. Yet the preparation contains richer bioactive content per particle than any commercial allogeneic product. The secretome is active; the commercial trillion-particle preparations often contain a substantial proportion of non-functional debris, as suggested by electron microscopy analyses of similar preparations.

6b. Characterised Composition and Bioactivity

Typical CFT preparations made this way contain a defined panel of bioactive factors at physiologically relevant concentrations. Concentrations include VEGF at 200–800 pg/mL, FGF-2 at 100–400 pg/mL, HGF at 50–150 pg/mL, PDGF-BB at 50–200 pg/mL, TGF- β 1 at 100–300 pg/mL, IL-6 at 50–500 pg/mL, IL-10 at 10–50 pg/mL, and IL-8 at 100–800 pg/mL, alongside EV concentrations of 10^9 – 10^{10} particles/mL and total protein of 1–10 mg/mL (Beer et al., 2016; Simader et al., 2017; see also Paper 02 in this series). This characterisation provides transparency: physicians and patients know what is in the preparation. By contrast, commercial allogeneic products often provide minimal characterisation beyond particle counts, leaving the actual biological content opaque.

Moreover, because CFT is autologous, derived from the patient's own blood, processed to concentrate the patient's own secretome, the biological signature of each patient's CFT preparation reflects that patient's unique physiology. A 30-year-old athlete's CFT will have a different molecular

signature from a 70-year-old with chronic disease, because their blood-derived secretory products are different. This is not a weakness; it is a strength. It is biological personalisation built into the product itself.

Commercial allogeneic products, by definition, can never achieve this personalisation. They deliver the same pooled donor material to every patient, applying a standardised formulation across diverse patient populations.

7. The Absence of Dose-Response Data

A critical gap exists in the exosome clinical literature: there are virtually no published, peer-reviewed dose-response studies establishing that higher particle counts produce greater therapeutic benefit. No clinical trial has compared 10^{11} particles to 10^{12} particles to 10^{13} particles and demonstrated a dose-dependent efficacy curve.

This absence is telling. In legitimate pharmaceutical development, dose-response is foundational. Establishing the dose-response relationship is how companies determine optimal dosing, how regulators approve drugs, and how clinical safety and efficacy are established. The exosome industry has largely bypassed this step, instead using particle count as a proxy for effect without empirical evidence that the proxy is valid.

The lack of dose-response data does not prove that higher counts produce no additional benefit. But it does demonstrate that the marketing claims about the superiority of ‘trillions’ rest on assumption, not evidence. The particle count arms race has outpaced the supporting clinical evidence.

8. The Bespoke Suit: An Analogy for Market Misdirection

Consider two suits of clothing. The first is bespoke: tailored specifically for you by a master craftsman, designed to fit your frame, made from fabrics matched to your colouring, and finished with precision. This suit has perhaps 50 buttons, 60 stitches per inch, and incorporates 8–10 distinct quality materials. It fits perfectly. It will serve you well.

The second suit is off-the-rack. It is inexpensive and mass-produced. It comes in 50 identical copies, manufactured from 5 different sizes and delivered to every body type in the country. Each suit has 200 buttons, 120 stitches per inch, uses 15 different fabrics, and employs complex synthetic materials designed to be moderately acceptable across a wide population. One suit is marketed as superior because it has four times as many buttons. The manufacturer emphasises the button count in all advertising.

You would immediately recognise this marketing as misleading. The number of buttons is irrelevant. What matters is fit, quality, durability, and whether the suit serves your specific body. The bespoke suit, with far fewer buttons, would be vastly superior.

This is the distinction between CFT and commercial allogeneic exosomes. CFT is the bespoke suit: personalised, precisely fitted to the patient's own physiology, smaller in volume but with greater biological relevance. Commercial 'trillions of exosomes' are the off-the-rack suit: mass-produced, optimised for volume metrics rather than function, marketed primarily on particle count. The suit with more buttons is not better. The preparation with more particles is not inherently better. Specificity, quality, and biological match determine efficacy.

9. Implications for Physicians and Patients

Physicians evaluating exosome preparations should critically evaluate marketing claims based on particle count alone. A trillion particles from pooled, extensively expanded allogeneic donors, many of which are not functionally intact, rapidly cleared by the immune system, and immunologically mismatched to the patient, may deliver less biological benefit than a far smaller volume of autologous secretome with complete immunological matching and full bioactive complement.

The right questions to ask are: What is the actual composition of this preparation? How much of the particle count represents intact, morphologically normal vesicles versus debris? What is the immunological match between preparation and patient? What is the in vivo half-life of these particles in circulation? What is the tissue biodistribution? Are there dose-response data supporting the claims made? Is the preparation autologous or allogeneic? If allogeneic, has the immunogenicity been characterised?

Patients should understand that they are not being offered a volume product; they are being offered a biological preparation that either does or does not support their tissue function. A massive particle count may be evidence of industrial efficiency in manufacturing, not evidence of therapeutic superiority. Ask your physician whether the specific preparation has been studied in a clinical setting relevant to your condition, and whether comparative data exist showing benefit over alternatives.

For CFT specifically, patients should understand that they are receiving their own biological secretome, concentrated from a blood draw, processed in a GMP facility, and delivered as an autologous preparation. There is no batch variability from multiple donors; there is variability between patients, because each patient receives their own unique biological signature. This is not a standardised pharmaceutical product. It is a personalised biological preparation.

10. Conclusion

The exosome industry has increasingly converged on a single, easily marketed metric that bears no necessary relationship to clinical efficacy. ‘Trillions of exosomes per dose’ is a headline, not a measure of quality. It obscures the factors that actually determine whether a preparation will support normal biological function: composition, immunological matching, cargo relevance, in vivo half-life, and biological specificity.

Allogeneic exosome companies achieve high particle counts through industrial cell expansion (causing phenotypic drift), pooled donor material (which may reduce biological relevance), and inclusive counting methods (measuring debris alongside functional particles). These high volumes are then rapidly cleared from circulation by the immune system, limiting therapeutic residence time. The patient receives a large number of particles, a significant proportion of which are likely cleared or non-functional, in an immune environment that works against foreign biological material.

The human body already circulates 10^9 , 10^{12} EV particles per mL endogenously. Adding exogenous particles to a system already carrying a substantial endogenous EV load may provide diminishing returns unless those particles are biologically relevant, immunologically matched, and persist long enough to act. This is the rationale for autologous preparations.

CFT delivers a smaller particle volume, 10^9 , 10^{10} particles per mL, but with inherent immunological self-matching (autologous), full biological activity (complete secretome rather than purified debris), and inherent cargo relevance (the patient’s own bioactive factors). The characterised presence of 20–30 distinct bioactive factors across the preparation, including growth factors, cytokines, and EV-associated miRNA, represents a richer biological composition than purified exosome preparations that isolate a single particle type. CFT does not compete on particle count because particle count is the wrong metric. It competes on what actually matters: specificity, potency, and biological match.

For physicians, the lesson is clear: when evaluating exosome and extracellular vesicle preparations, look past the particle count. Demand characterisation, immunological matching data, biodistribution evidence, and, most importantly, clinical outcome data demonstrating benefit in your patient population. For patients, the lesson is equally straightforward: a preparation with trillions of particles does not automatically support your biological function better than a bespoke, personalised alternative with far fewer particles but potentially greater biological relevance. Quality, not quantity, determines outcome.

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Disclaimer: Individual results vary. Cell-Free Therapy is not intended to diagnose, treat, cure, or prevent any disease. The information in this paper is provided for educational purposes and does not constitute medical advice. CFT supports the body's normal biological function through autologous, cell-free biological preparations.

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